A. **PURPOSE**:

The primary use of the Fourier Transform Infrared Spectrometer (FTIR) in the Controlled Substance laboratory is for the confirmation of the free base form of cocaine. Rarely this instrument will be used in the identification of gas samples (such as nitrous oxide). The FTIR is a confirmatory technique which provides a structural identification of the unknown.

Most important to the CS laboratory is the ability of the FTIR to distinguish cocaine salt form (CSF) from the free base form (CFB). The FTIR is used in conjunction with the GC/MS, with the GC/MS being used to confirm the presence of cocaine in samples, and a combination of solubility and FTIR being used to confirm the free base form.

Historically the distinction of CFB from CSF or a mixture of the two forms was important prior to 2005 since the criteria weights and associated penalties associated with the possession of cocaine were set at 0.5g for CFB and 1 ounce or 28.35 grams for CSF. Currently the criteria weights and associated penalty changes are set at 0.5 ounces for both forms of the drug. The laboratory has chosen to continue the practice of distinguishing the two forms of the drug.

B. **SAFETY**:

The FTIR utilizes a laser which should not be directly viewed. As with any electrical device there is a chance of electrical shock if not handled properly, do not perform maintenance on this instrument unless trained to do so. A laboratory coat should be worn while working with samples on the FTIR

C. **RESPONSIBILITY**:

All analysts (however titled) assigned to the CS section are responsible to follow the guidance of this SOP when utilizing the FTIR for case analysis.

D. **DEFINITIONS**:

FTIR: Fourier Transform Infrared spectrometry

HATR: Horizontal Attenuated Total Reflectance

E. **PROCEDURE**:

1. Instrumentation: the CS laboratory utilizes a Perkin-Elmer Spectrum BX FTIR with an Pike HATR attachment. This attachment allows for samples to be run directly without sample preparation. Other FTIR may be used if it has been shown to be able to distinguish CFS from CFB.
2. Controls;
    a. Monthly (see below for instructions)

i. A validation is performed on the FTIR on a monthly basis. For this the following are run: the instrument validation program, a polystyrene standard, a Schott's glass standard, a blank, a background and a cocaine free base standard. The monthly check is recorded as being acceptable or not in the instrument log book.

ii. If any component of the validation fails document the failure in the instrument log book and contact the section Supervisor to follow-up on the issue.

b. Day of Use: The maintenance log (CS-8.1) is completed on the day of use indicating the following was run and acceptable.

i. Prior to each batch of samples the energy of the light source is monitored to verify that there has not been a significant energy decrease; which may indicate a loss of sensitivity.

ii. A control powder consisting of benzocaine is run each day of instrument use to demonstrate that the instrument is running properly. Benzocaine is used since it absorbs IR energy in areas similar to those of primary interest for cocaine. The benzocaine standard is then printed and filed. The lot number of the Benzocaine standard will be included on the instrument printout. Note if the monthly check has been run on the day of use, the Benzocaine standard need not be run since the CFB standard run demonstrates proper working condition of the device.
Prior to running a batch of samples a background correction is run; this aids in removing any ambient room conditions (such as moisture in the air) and instrument noise from the spectra.

iii. Prior to each run the sampling platform is cleaned with ethanol or other appropriate solvent to remove any materials from previous samples.

iv. Prior to each sample a blank is run. The blank assures that the sampling platform is clean and void of materials from a previous sample.

3. Analysis: Using Pike HATR

a. Open the Spectrum program.

i. Prior to the first sample in the batch (or as needed) run a background scan. Humidity in the room may cause a need for more frequent background scans, this will be evidence in the noise seen in a blank run.

(a) Under Instrument, pick scan background -type in the required information.

(b) Run the scan, this background scan is subtracts out the room/instrument conditions for each sample or blank run after.

ii. Run a blank scan. The blank is the platform on the HATR with the piston in the down position covering the diamond crystal, with nothing on the diamond crystal.

(a) Under Instrument pick Scan Sample - a prompt will be given for the file name and description.

(i) The file name must be distinct or the file will be overwritten.

(ii) The description should allow for the understanding of what the sample is. Example: blank for TX-11-XXXX and analysts initials.

(iii) Run the blank scan, the spectra will appear on the screen. This should be a flat line; there may be some baseline noise, however there should not be any noise in the areas of interest for cocaine (see interpretation of data for areas of interest). If there are peaks in the areas of interest for cocaine, the platform should be re-cleaned and the blank re-run.

iii. Run the sample scan - Under Instrument pick Scan Sample a prompt will be given for the file name and description.

(a) The file name must be distinct or the file will be overwritten. Putting the case number in the file name can be helpful.

(b) The description should be the case number followed by the sample identifier, followed by the analysts initials. Example: TX-11-XXXX 1A1 jmr.

(c) Place a small portion (enough to just cover the crystal) of the sample on the diamond crystal and dial the piston to the closed position. Run the scan, the scan will appear on the screen.

(i) Click on sample spectra. Then click on the compare function from the Process menu; this will compare the scan of the unknown to the library standards.

(ii) To see the differences of the CFB and CSF clearer it may be helpful to open the cocaine salt form standard spectra from the instrument library so that it overlays the unknown spectra, print this to show the presence or absence of the CSF peak. If a significant "cut" substance was identified by GC/MS, it may be helpful to also open that particular standard spectra to show the substance would/would not interfere with identification of CFB.

(iii) Print the sample spectra with the blank and the sample spectra with the comparative standard. The comparative value of the sample to the standard is written on one of these pages along with the analysts conclusion.

b. Interpretation of Data:

i. Peaks of Interest:

(a) Cocaine freebase (CFB), ~2945cm-1, ~1734cm-1 (doublet peak) ~1706cm-1 ~712cm-1.

(b) Cocaine salt form (CSF), same as for CFB however there will be an additional peak at ~730cm-1. Additionally for CSF slight shifts can occur at the 2945, 1734 and 1706cm-1 ranges.

ii. The quality match is a tool to aid the analyst; however it is the analyst's responsibility to visually compare the spectra of the unknown to the standard to verify the presence of the peaks of interest and absence of the peak seen in salt form samples. The following are general guidelines for the spectrum quality match:

(a) 100-95% : scan is acceptable, the sample can be identified as cocaine freebase

(b) 90-95% :scan is acceptable, however report must show documentation that no CSF present (zoom in on peak at ~730cm-1).

(i) If a peak is present at 730cm-1, then the sample is identified as a mixture of cocaine freebase and salt form (the CSF standard should be opened and printed with the sample and the CFB standard to show this comparison).

(ii) If no peak is seen at 730cm-1 then the sample is identified as cocaine freebase.

(c) <90%: Cocaine freebase can be identified only if no peak is seen at 730cm-1 and the GC/MS identifies the unknown as cocaine.

(i) Cocaine freebase and salt form mixture is reported if there is a peak at 730cm-1 and the GC/MS identifies the cocaine.

(ii) <75% The identification of the unknown is from the GC/MS data. In these cases it is likely that there is a cut interfering with the identification, if the cut is known (by GC/MS or as identified by the IR) and the cut occurs in the IR library the analyst should open this standard along with the sample and CFB or CSF. If the GC/MS demonstrates that Cocaine is present the analyst will need to use their experience to determine if the form can be distinguished by the IR. Refer to the section supervisor if needed.

1. The analyst should view the ~730cm-1 region is there is no interfering peaks in this area and no peak is seen the sample can be identified as the free base from of the drug.

4. Analysis: Using Gas Cell

a. The gas cell is a chamber which allows a sample that is in a gaseous state to be sampled and analyzed on the FTIR. This laboratory utilizes this cell for the analysis of evidence submitted that is believed to be nitrous oxide. This is a rarely run analysis within the CS section.

i. Remove the HATR and attach the gas cell to the FTIR

(a) The gas cell is a closed glass cylinder with 2 stop cocks. These are utilized to fill and purge the cell.

ii. Utilizing the FT-IR spectrum software run a background scan; this scan will have a "zeroing" effect correcting for any interference by the glass cell itself or the room air.

(a) Purging the gas cell (cleaning) open both stop cocks so that it is an open system; attach a CO_2 to the gas dispenser and gently allow the gas to be sent into the cell.

(b) Run a blank; the blank spectra should be a straight line; note that small peaks corresponding to moisture do to humidity are acceptable. The blank spectra will correspond to a purged cell.

(c) If the blank spectra is a straight line (possibly with the above mentioned exceptions) the sample can run

(d) If the blank is unacceptable re-purge the cell until an acceptable blank is produced.

(e) Close one of the stop cocks and dispense the sample into the cell via the open 2nd stop cock once sample is added close the 2nd stop cock. This is a closed gas cell system containing the sample.

(f) In the Spectrum program run the sample. The spectra produced is then compared to the library standards, the compare program will give the percentage to the top matches in the library.

(g) The sample should be run in duplicate if there is only one item or 2 individual items run if there are 2 or more identical items (i.e. 2 or more like gas cylinders)

(h) Run a nitrous oxide standard in the same manner starting with step (iv).

F. **SOURCES OF ERROR**:

1. For Cocaine Free base/Salt form differentiation:

a. Interfering agents in the sample; these are street drugs which can be mixed with most anything therefore it is important to realize that the spectra can be skewed.

b. Not covering the surface of the crystal; poor readings can occur if the sampling crystal is not sufficiently covered, if this is found to be true simply repeat the sample run.

c. Not cleaning the sample platform sufficiently between samples. This will be seen in "dirty" blank spectra.

d. Misinterpretation of data;

i. If a peak is present at 730cm-1 and the presence of the salt is not indicated.

ii. Improper input of data into the software so that the sample is misidentified.

2. For Nitrous Oxide Analysis:

a. Not performing a background scan to correct for the glass cell and environmental conditions

b. Not performing a blank to verify that there is no carry over, i.e. the cell is clean.

G. **INSTRUMENT MAINTENANCE**:

1. The FTIR is a very stable instrument and requires very little maintenance. The maintenance performed includes; daily (as used), monthly, Bi-yearly and as needed items.

a. Day of use: see checks under control section.

b. Monthly a check is performed as described under the Controls section. The instrument's validation program in conjunction with running a polystyrene standard, Schott's glass standard, background scan and a Cocaine free base standard are used to verify that the instrument is working consistently from month to month.

i. Validation Program; this is a program that is part of the instrument software which allows the instrument to perform a self-check.

(a) The instrument validation program is used to provide a test of wave number precision, %T repeatability and noise against previously stored results.

(b) To run and interoperate the results of the validations see the instrument operator's manual.

(c) The printed validation report is signed and stored in the instrument's maintenance log book.

(d) Report validation failures directly to the section supervisor or designee. The supervisor will contact Perkin-Elmer if required.

ii. Polystyrene and Schott NG11 Glass Reference Standards; these two standards are used to show the consistent working condition of the instrument.

(a) Run the standards per the instruments operator's manual.

(b) Interpretation; Compare the spectra of the reference standard to previously run standards to confirm that there are no shifts in the spectra. Major differences indicate

problems with the instrument. Consult with your supervisor or designee and they will contact Perkin-Elmer if required.

(c) Print the spectrum, initial the report and store the spectra in the instrument's maintenance logbook.

iii. Background Scan;

(a) A background scan is run under the Spectrum program like any other sample.

(b) Interpretation: Compare the spectra to previously run backgrounds; if there are major shifts in the spectra consult your supervisor or designee. The supervisor will contact Perkin-Elmer if required.

(c) Print the spectrum, initial the report and store the spectra in the instrument's maintenance logbook.

iv. Cocaine Standard Scan;

(a) A cocaine free base standard is run to verify that the spectrum matches the library standard. This is run as any typical sample

(b) The spectra is printed, initialed and stored in the IR Maintenance logbook. The lot number of the Cocaine standard run is documented on the spectra.

v. Interpretation; Compare the spectra to previously run standards; if there are major shifts in the spectra consult your supervisor or designee. The supervisor will contact Perkin-Elmer, or other appropriate service representative if required.

c. Bi-annually: the desiccant is changed. This is performed by opening the cover of the instrument removing the cage which contains the desiccant and placing fresh desiccant bags in the cage. The cage is then returned to the instrument and the software is updated to show the change. See below.

d. As Needed such as due to a drop in the energy level or after replacing the Pike: the Pike alignment can be adjusted to maximize the energy levels of the light source.

i. While in the Spectrum program monitor the Energy level, adjust the two adjustment screws on the Pike HATR to obtain the maximum energy level. Record this level in the maintenance log.

H. REFERENCES;

a. Operating the Perkin-Elmer Spectrum 1000 FT-IR Spectrophotometer, operator's manual.

b. Spectrum BX FT-IR User's Guide; Perkin-Elmer, 1998

c. Spectrum BX FT-IR Help Topics; Instrument Validation.

d. Spectrum BX FT-IR Help Topics; Performing an instrument validation

e. Clark's Isolation and Identification of Drugs in pharmaceuticals, body fluids, and post-mortem materials, The Pharmaceutical Society of Great Britain.

Introduction:

The instrument validation is used to provide a test of wavenumber precision, %T repeatability and noise against previously stored results.[1] Along with the validation a polystyrene film reference standard, a Schott NG11 glass reference standard and a background will be run. A cocaine freebase standard will be run to compare to the library standard in the shortcut to CFB program and the Spectrum program. The intent of these tests is to assure that the instrument is working consistently. These tests will be performed monthly.

Running the Validation:

1. Remove the Pike attachment from the sampling compartment. This compartment is left completely empty for the validation.
2. Open the Spectrum software.
3. Click on the Instrument menu – **Validate**.
4. The software will remind you to make sure that the sample compartment is empty.
5. Click on **OK**.
6. The validation will start and messages will be displayed showing the progress of the validation. When complete a dialog box will appear saying that the instrument has passed or failed the test click **OK** and the report will print automatically.
7. Review the report and sign in the appropriate spot.

Interpretation:

The instrument validation report prints automatically. This includes an abscissa test, ordinate test and a noise test. The report lists the observed result and the upper and lower limits for the test. The final column lists whether the instrument passed of failed the particular test. The abscissa test represents the x-axis of the graphed spectrum this checks the wavelength (or wavenumber). The Ordinate test represents the y-axis of the graphed spectra, this checks the percent transmittance. The noise test checks the level of random signals detected by the instrument. The following are the limits set by Perkin Elmer:

	Nominal	Upper limit	Lower Limit
Abscissa test:			
Peak 1 (cm-1)	3060.00	3062.00	3058.00
Peak 2 (cm-1)	1601.00	1604.00	1598.00
Peak 3 (cm-1)	1028.00	1030.00	1025.00
Ordinate test:			
3990.00 cm-1 (%T)	70.00	80.00	60.00

3030.00 cm-1 (%T)	30.00	40.00	20.00
2000.00 cm-1 (%T)	0.00	5.00	-5.00

Noise test:

RMS (%T)	0.00	1.00	-
Peak to peak (%T)	0.00	1.00	-
Trend (%T)	0.00	1.00	-

If the instrument passes the validation sign the report and file the report in the maintenance log book. If the instrument fails the test, turn the instrument off, restart the instrument allow it the equilibrate and perform the test again. If the instrument fails again inform your supervisor so that a Perkin-Elmer representative can be contacted. File the copies of all validation reports (pass or fail) in the maintenance log book. [1] Spectrum™BX FT-IR Help Topics; Instrument Validation.

Running the Polystyrene and Schott NG11 Glass Reference standards:

1. Place the sample holder in sampling compartment (not the Pike attachment.)
2. Place in the reference slide (Polystyrene traceable PE02691 or PE NG11 Glass 1mm) in the sample holder.
3. Open the Spectrum program.
4. Click on Instrument and **Scan Sample.**
5. A message box will appear requesting a file name and sample information.
   Under file name for the Polystyrene standard use PS and the date (i.e. PS4902) and for the Schott glass standard use GL and date (i.e. GL4902.) Under sample information a description such as Polystyrene traceable PE02691 or PE GN11 Glass 1MM is acceptable.
   Note: The instrument will save the data in an .sp file, to keep a running record of this information use the file manager and place the spectrum in the file name instrument validation.
6. Click on **OK** and the standard will run.
7. Print the spectra. Sign the page and file in the instrument maintenance logbook.
8. Put the HATR back in the instrument and align.

Interpretation:

Compare the spectra of the reference standard to previously run standards to confirm that there is no shift in the spectra. Major differences indicate problems with the instrument. Consult with your supervisor and they will contact Perkin Elmer if needed.

Running a background check:

1. Open the Spectrum program.
2. Click on Instrument and Scan background.
3. A dialog box will appear requesting a file name and sample information.
   For the file name use BG and date (i.e. BG4902) for the sample information a description such as instrument background check is acceptable.
   Note: The instrument will save the data in an .sp file, to keep a running record of this information use the file manager and place the spectrum in the file name instrument validation.
4. Click **OK** and the background will run.
5. Print the spectra.
6. Sign the report and place this in the maintenance logbook.

Interpretation:

Compare this to previously run backgrounds, if there is a major shift in the spectra consult your supervisor. The supervisor will contact Perkin Elmer if necessary.

Running a CFB standard:

1. Run a blank like any sample blank
2. Add Cocaine Free base standard to the platform and close the piston.
   a. Type in FB and the date for the file name (i.e. FB052110)
3. Under Description type in CFB standard and the lot number.
4. Run the standard.
5. Go to Compare.
6. Print the spectrum with the comparison standards and the comparison percentages.

Interpretation:

The standard should compare at 95% or above to the library standards. If the result is <95% repeat the standard, if it remains below 95% consult the supervisor or a Deputy Director.