MultiScope System Microscope User's Reference

Release History

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Any comments about the documentation for this product should be addressed to:

User Assistance
PerkinElmer Ltd
Chalfont Road
Seer Green
Beaconsfield
Bucks HP9 2FX
United Kingdom

Or emailed to: info@perkinelmer.com

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Contents

Introduction

Introduction to this User's Reference

This introduction gives you information about this *User's Reference*, so you can use it effectively when learning to operate the microscope.

We recommend that you use it in the following order:

- Read the rest of this *Introduction* and become aware of requirements and conventions.
- Read the *Overview of the MultiScope System Microscope*.
- Do the setup procedures in *Getting Ready to Use the Microscope*.
- Try the tutorial in *Tutorial: Using the Microscope*. This gives you experience of using the microscope to collect both transmittance and reflectance spectra.
- The procedures you use for all your work with the microscope are in *Getting Ready to Use the Microscope*, *Preparing Samples, Collecting Spectra with the Microscope* and *Operating the Optional Equipment.*
- *Getting Ready to Use the Microscope* contains the procedures you follow at the beginning of the day's work to make sure the microscope is set up properly.
- *Preparing Samples* describes techniques for preparing many types of microscopic samples. It includes descriptions of the sample preparation tools provided with, or available for, the PerkinElmer MultiScope System Microscope.
- *Collecting Spectra with the Microscope* contains general procedures for collecting spectra with the microscope.
- *Operating the Optional Equipment* describes how to use the optional equipment that is available for your microscope.
- Further reference information on the microscope is given in *Applications* and *Maintenance*.
- *Applications* gives information on several applications for which the microscope is particularly useful.
- *Maintenance* contains maintenance information. Read through it to see how to care for the microscope and to learn about the performance checks that we recommend that you do routinely.

If you find an error in this manual or have any comments or suggestions, please contact User Assistance at PerkinElmer. Our address is on page 2.

Related Documents

Manuals for PerkinElmer FT-IRs

Information on using your FT-IR is found in the manuals that were supplied with it.

Optional equipment

Certain items of the optional equipment for the microscope have their own manuals. Consult the appropriate manual before attempting to use the equipment.

What am I Expected to Know?

We assume that you have some familiarity with operating the FT-IR. Specifically, you must be able to:

- Set the FT-IR to use the external beam;
- Monitor the signal;
- Set the gain;
- Collect background and sample spectra.

This *User's Reference* does not give detailed procedures for these operations. Consult the manual for the FT-IR.

Conventions Used in this Manual

Normal text is used to provide information and instructions.

Bold text refers to text that is displayed on the screen.

UPPERCASE text, for example ENTER or ALT, refers to keys on the PC keyboard. '+' is used to show that you have to press two keys at the same time, for example, ALT+F.

All eight digit numbers are PerkinElmer part numbers unless stated otherwise.

Notes, cautions and warnings

Three terms, in the following standard formats, are also used to highlight special circumstances and warnings.

NOTE: A note indicates additional, significant information that is provided with some procedures.

CAUTION	*We use the term CAUTION to inform you about situations that could result in **serious damage to the instrument** or other equipment. Details about these circumstances are in a box like this one.*
D	***Caution (Achtung)*** *Bedeutet, daß die genannte Anleitung genau befolgt werden muß, um einen **Geräteschaden** zu vermeiden.*
DK	***Caution (Bemærk)*** *Dette betyder, at den nævnte vejledning skal overholdes nøje for at undgå en **beskadigelse af apparatet**.*
E	***Caution (Advertencia)*** *Utilizamos el término **CAUTION** (ADVERTENCIA) para advertir sobre situaciones que pueden provocar **averías graves en este equipo** o en otros. En recuadros éste se proporciona información sobre este tipo de circunstancias.*
F	***Caution (Attention)*** *Nous utilisons le terme **CAUTION** (ATTENTION) pour signaler les situations susceptibles de provoquer de **graves détériorations de l'instrument** ou d'autre matériel. Les détails sur ces circonstances figurent dans un encadré semblable à celui-ci.*
I	***Caution (Attenzione)*** *Con il termine **CAUTION** (ATTENZIONE) vengono segnalate situazioni che potrebbero arrecare **gravi danni allo strumento** o ad altra apparecchiatura. Troverete informazioni su tali circostanze in un riquadro come questo.*
NL	***Caution (Opgelet)*** *Betekent dat de genoemde handleiding nauwkeurig moet worden opgevolgd, om **beschadiging van het instrument** te voorkomen.*
P	***Caution (Atenção)*** *Significa que a instrução referida tem de ser respeitada para evitar a **danificação do aparelho**.*

 **WARNING**	*We use the term WARNING to inform you about situations that could result in **personal injury** to yourself or other persons. Details about these circumstances are in a box like this one.*
D	***Warning (Warnung)*** *Bedeutet, daß es bei Nichtbeachten der genannten Anweisung zu einer **Verletzung** des Benutzers kommen kann.*
DK	***Warning (Advarsel)*** *Betyder, at brugeren kan blive **kvæstet**, hvis anvisningen ikke overholdes.*
E	***Warning (Peligro)*** *Utilizamos el término **WARNING** (PELIGRO) para informarle sobre situaciones que pueden provocar **daños personales** a usted o a otras personas. En los recuadros como éste se proporciona información sobre este tipo de circunstancias.*
F	***Warning (Danger)*** *Nous utilisons la formule **WARNING** (DANGER) pour avertir des situations pouvant occasionner des **dommages corporels** à l'utilisateur ou à d'autres personnes. Les détails sur ces circonstances sont données dans un encadré semblable à celui-ci.*
I	***Warning (Pericolo)*** *Con il termine **WARNING** (PERICOLO) vengono segnalate situazioni che potrebbero provocare **incidenti alle persone**. Troverete informazioni su tali circostanze in un riquadro come questo.*
NL	***Warning (Waarschuwing)*** *Betekent dat, wanneer de genoemde aanwijzing niet in acht wordt genomen, dit kan leiden tot **verwondingen** van de gebruiker.*
P	***Warning (Aviso)*** *Significa que a não observância da instrução referida poderá causar um **ferimento** ao usuário.*

Warning Signs on the Instrument

Caution, hot surface.

Caution, risk of electric shock.

Caution, laser radiation hazard.

Caution risk of danger.

Refer to accompanying documents in all cases where this symbol is used to find out the nature of the potential HAZARD and any actions which have to be taken.

FT-IR

The MultiScope System Microscope can be used with several different PerkinElmer FT-IR spectrometers. In this *User's Reference*, we have used the term FT-IR to mean the FT-IR spectrometer that you have connected to the microscope.

Spectrum 100 Series

The MultiScope System Microscope can be used with Spectrum 100 Series spectrometers.

Spectrum One

The MultiScope System Microscope can be used with Spectrum One spectrometers.

Spectrum GX

The MultiScope System Microscope can be used with Spectrum GX, System 2000, Spectrum 2000, System 2000R NIR FT-Raman, Spectrum 2000R NIR FT-Raman FT-IR, and Spectrum GXR FT-IRs. In this *User's Reference* we have used Spectrum GX to indicate Spectrum GX, System 2000, Spectrum 2000, System 2000R NIR FT-Raman, Spectrum 2000R NIR FT-Raman FT-IR, and Spectrum GXR spectrometers.

Spectrum BX

The MultiScope System Microscope can be used with Spectrum BX, Paragon 1000PC, and Spectrum 1000 FT-IRs. In this *User's Reference* we have used Spectrum BX to indicate Spectrum BX, Paragon 1000PC, and Spectrum 1000 spectrometers.

Spectrum RX

The MultiScope System Microscope can be used with Spectrum RX, or Paragon 1000 FT-IRs. In this *User's Reference* we have used Spectrum RX to indicate Spectrum RX or Paragon 1000 spectrometers.

Spectrum 400 Series

The MultiScope System Microscope can be used with Spectrum 400 Series spectrometers.

Frontier

The MultiScope System Microscope can be used with Frontier FT-IR spectrometers.

Warnings and Safety Information

Summary

The PerkinElmer MultiScope System Microscope has been designed to comply with a wide variety of international standards governing the safety of laboratory equipment. In routine use, the microscope poses virtually no risk to you. If you take some simple, common-sense precautions, you can make sure that you maintain the continued safe operation of the microscope.

DO make sure that the microscope is properly connected to the electrical supply; in particular, make sure that the ground (earth) is securely connected.

DO disconnect the electrical power supply before opening the main cover of the microscope.

DO keep the microscope dry. Avoid spilling liquid into the microscope. Clean all external spills immediately. If anything that is spilled enters the main body of the microscope, switch off the power and call a PerkinElmer Service Engineer.

DO wear safety glasses and protective gloves when you are filling the detector dewar with liquid nitrogen. Slowly pour the liquid nitrogen into the dewar. Stand back from the detector during filling since liquid nitrogen may be expelled from the dewar flask. Use **only** liquid nitrogen.

DO NOT stare into the laser beam. Laser radiation will be present when the microscope is connected to an FT-IR spectrometer. Typically these FT-IRs use low-power visible (red) lasers and momentary exposure to the beam is not dangerous, but deliberate, direct viewing of the beam along its axis could damage your eyes.

DO NOT use a flammable gas to purge the MultiScope System Microscope. The microscope contains a hot lamp, and fire or explosion may result. Use only clean, dry oil-free nitrogen or air to purge the instrument.

DO read the more detailed information on safety in the following pages.

WARNING

Removal of covers and fixtures not specified in this manual may result in hazardous radiation exposure.

Use of controls or adjustments or performance of procedures other than those specified herein may result in hazardous radiation exposure.

General Operating Conditions

The MultiScope System Microscope has been designed and tested in accordance with PerkinElmer specifications and in accordance with the safety requirements of the International Electrotechnical Commission (IEC). The MultiScope System Microscope conforms to IEC 61010-1 (Safety requirements for electrical equipment for measurement, control, and laboratory use) as it applies to IEC Class 1 (earthed) appliances and therefore meets the requirements of EC Directive 2006/95/EC.

If the microscope is used in a manner not specified by the manufacturer, the protection provided by the microscope may be impaired.

Only use the MultiScope System Microscope indoors under the following conditions:

Temperature	15 °C to 35 °C
Relative humidity	75% maximum (non-condensing)

If possible, avoid any adjustment, maintenance and repair of the opened, operating instrument. If any adjustment, maintenance and repair of the opened instrument is necessary, this must be done by a skilled person who is aware of the hazard involved.

Whenever it is likely that the microscope is unsafe, make it inoperative. The microscope may be unsafe if it:

- Shows visible damage;
- Fails to perform the intended measurement;
- Has been subjected to prolonged storage in unfavorable conditions;
- Has been subjected to severe transport stresses.

Environmental Conditions

The microscope has been designed to be safe under the following conditions:

- Indoor use;
- Altitude up to 2000 m;
- Ambient temperatures of 5 °C to 40 °C;
- A maximum ambient relative humidity of 80% for temperatures up to 31 °C, decreasing linearly to 50% relative humidity at 40 °C;
- Mains fluctuations not exceeding ± 10% of the nominal voltage.

Electrical Safety

Connect the MultiScope System Microscope to a power supply line that includes a switch or other adequate means of disconnection from the electricity supply.

Plug the microscope only into an electricity-supply socket that is provided with a protective ground (earth) connection.

If fuses need replacing, use only those with the required current rating and of the specified type. Do not use makeshift fuses and do not short-circuit fuse holders.

When replacing the fuse drawer on the mains inlet connector make sure that the drawer is in the correct orientation for your mains supply. The drawer is also the voltage selector.

When the MultiScope System Microscope is connected to its electricity supply, terminals may be live. Removing covers other than those which can be removed by hand is likely to expose live parts.

Capacitors inside the microscope may still be charged even if the microscope has been disconnected from all voltage sources.

Disconnect the microscope from all voltage sources before it is opened for any adjustment, replacement, maintenance or repair.

When replacing lamps, remember that the surface may be hot. The lamp and surrounding area may exceed 30 °C above ambient.

WARNING

Any interruption of the protective ground (earth) conductor inside or outside the microscope or disconnection of the protective earth terminal can make the microscope dangerous.

The MultiScope System Microscope has:

- An IEC Pollution Degree 2 classification - usually only non-conductive atmospheric pollution of the equipment occurs; occasionally, however, a temporary conductivity caused by condensation must be expected.
- An IEC Installation Category (Overvoltage Category) II classification - suitable for connection to local level power supplies.

The instrument is designed to be safe under transient overvoltages typically present on the MAINS supply.

NOTE: The normal level of transient overvoltages is impulse withstand (Overvoltage) category II of IEC 60364-4-443.

Location and Ventilation

The microscope is installed by a PerkinElmer Service Representative, who will be able to advise on the siting of the system. To allow for adequate cooling, the system should not be sited near to room heating equipment, for example, central-heating radiators. There should be a minimum gap of at least 15 cm (6 inches) from the top and side surfaces of the microscope to permit adequate cooling.

WARNING

Make sure that the switch at the electrical supply inlet at the rear of the microscope is not obstructed.

Laser Safety Regulations

When connected to a PerkinElmer FT-IR spectrometer, the MultiScope System Microscope complies with IEC Publication 60825-1: 2007, “Safety of laser products. Equipment classification and requirements.”

Laser Product Classification

The MultiScope System Microscope is not a laser product because it does not contain a laser system. However, the microscope forms part of a laser product when it is assembled to an FT-IR spectrometer (which does contain a laser system). The appropriate laser product classification will be described in the FT-IR spectrometer user manual.

Accessible Levels of Laser Radiation

Laser radiation may be accessed in the following areas of the microscope:

- Between the lower cassegrain input mirror (mounted beneath the lower cassegrain mirror) and the upper cassegrain mirror when the microscope purge covers are displaced or removed. In Transmittance mode, the laser radiation propagates upwards from the lower cassegrain input mirror. In Reflectance mode, the laser radiation propagates downwards from the upper cassegrain mirror; a sample reflects the radiation upwards, along the same path (see Figure 2 on page 25).
- At the Remote Aperture, between the upper cassegrain mirror housing and the detector mirror housing, when the aperture disc is removed. The laser radiation propagates upwards, through the Remote Aperture (see Figure 2 on page 25).

The laser radiation levels that may be accessed on the MultiScope System Microscope are within Class 1 limits (IEC 60825-1:2007). Class 1 levels of laser radiation are not considered to be hazardous.

Laser Labels and Laser Apertures

This manual contains information and warnings that you must follow to make sure that you use the microscope safely. Warning labels are fixed to the microscope in the locations shown on the following pages (Figure 1 and Figure 2). Class II/Class 2 laser warning labels are attached to the Model 1600/Spectrum BX/Spectrum RX version of the microscope. No laser warning labels are required on the Class I / Class 1 Spectrum GX, Spectrum One and Spectrum 100 Series versions of the microscope. The laser apertures are shown in Figure 2 and described in *Accessible Levels of Laser Radiation* on page 23.

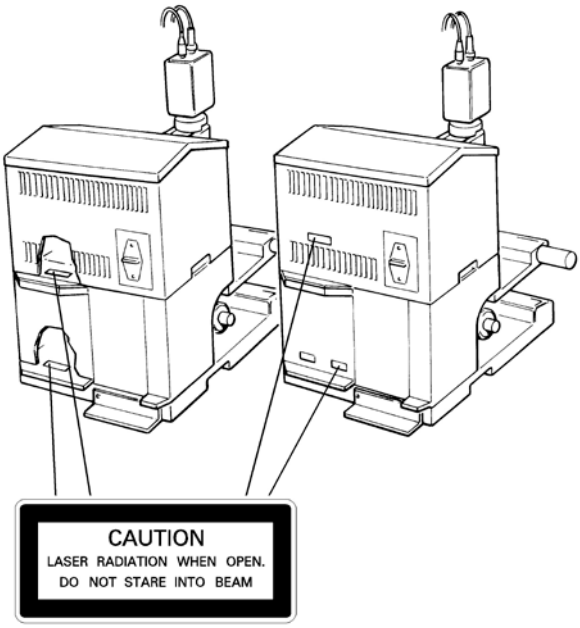

Figure 1 Position of laser radiation warning labels

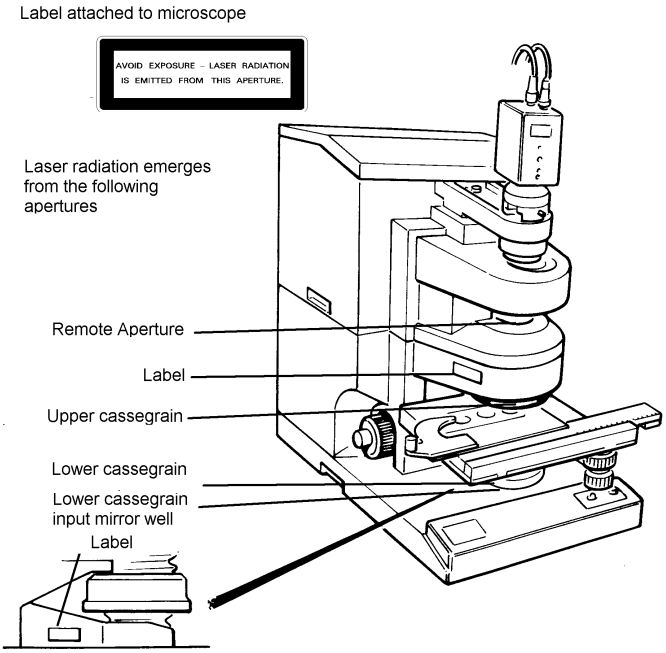

Figure 2 Position of laser radiation warning labels and laser apertures

To maintain compliance with laser safety regulations, at least once a year, or whenever the unit has been subjected to adverse environmental conditions, verify that all features of the unit are functioning properly.

Visually inspect the housing periodically to verify that no panels are loose or distorted so as to allow access to laser radiation in the interior. Verify that operation of the external beam lever on a Model 1600, Spectrum BX or Spectrum RX blocks laser radiation from entering the microscope. Verify that all required labels are firmly in place.

Warning Label Near the Lamp Housing

Caution, hot surface.

Lifting the Microscope

The MultiScope System Microscope weighs approximately 27.5 kg (approximately 33 kg with packaging).

Lift the microscope only by the base. Do not attempt to lift it by the knobs, cassegrain assembly or other attachments.

We recommend that the microscope is lifted by 2 adults.

Consult local codes of practice issued by safety advisors before attempting to lift the microscope.

Do not move the microscope after it has been installed without consulting your local PerkinElmer service department.

EMC Compliance

EC Directive

The MultiScope System Microscope has been designed and tested to meet the requirements of the EMC Directive 2004/108/EC. The microscope complies with the EMC standards EN 55011 (ISM) class A (rf emissions), and the generic immunity standard EN 50082-1 for residential, commercial and light industrial environments.

FCC rules and regulations

This product is classified as a digital device used exclusively as industrial, commercial, or medical test equipment. It is exempt from the technical standards specified in Part 15 of the FCC rules and Regulations, based on Section 15.103(c).

System Requirements

Give attention to the following points before installing the MultiScope System Microscope.

Electrical Requirements

The power consumption of the microscope does not exceed 80 VA.

The line supply must be within 10% of the nominal voltage.

If possible, do not connect the microscope to circuits that have heavy duty equipment, such as large motors, connected.

If possible, do not use photocopiers, discharge lamps, radio transmitters, and other equipment with large or frequent transient loads on the same supply circuit.

The microscope can operate on electricity supplies of 50 or 60 Hz and on voltage ranges of either 100 to 120 V or 220 to 240 V.

The primary fuses (2 A time delay, 250 V, UL/CSA) are in the voltage selector drawer on the mains inlet panel. One fuse is connected in the live line of each voltage range.

The voltage selector for the microscope is located on the mains inlet panel on the back of the top cover.

The voltage you require must be at the bottom of the panel with the label the correct way up. The white arrow on the fuse drawer must point down to the white bar.

If the setting is incorrect, remove the voltage selector/fuse drawer, turn it over, and refit the drawer.

Site Requirements

Minimum bench dimensions of 55 x 36 cm to accommodate the microscope.

To get the best performance from your microscope:

- Place the microscope in an environment that is relatively dust-free.
- Make sure that the bench top is free from vibrations or mechanical shocks.
- Do not place the microscope near to room heating equipment, for example, central heating radiators.

Leave at least 15 cm (6 inches) from any vertical obstacle to the sides of the microscope, to permit an adequate flow of cooling air.

Overview of the MultiScope System Microscope

Features of the MultiScope System Microscope

This chapter introduces the PerkinElmer MultiScope System Microscope. It describes the features of the microscope and the available optional equipment, lists special applications for which you can use the microscope, describes the optical system, and gives specifications.

The PerkinElmer MultiScope System Microscope (Figure 3) can be used with any of the following PerkinElmer FT-IRs: Spectrum 100 Series, Spectrum One, Model 1600, Model 16PC, Spectrum RX, Spectrum BX or Spectrum GX.

The microscope enables you to collect the spectra of extremely small samples. It is designed around PerkinElmer cassegrain collection optics for high-performance infrared microspectroscopy. The microscope is combined with a viewer, which magnifies the visible-light image of the sample so that you can see, position, and isolate a point of interest.

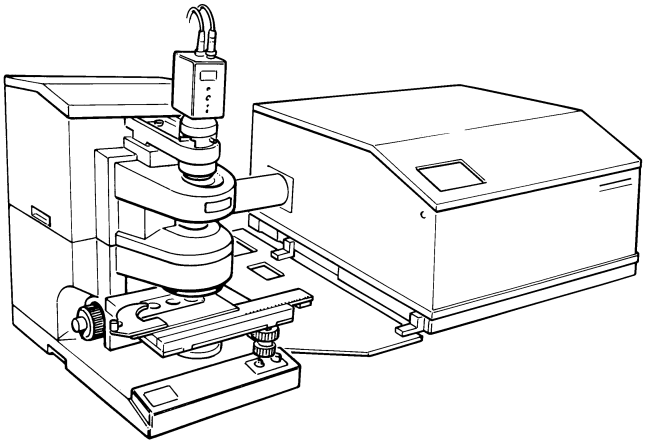

Figure 3 The MultiScope System Microscope connected to an FT-IR

List of Features

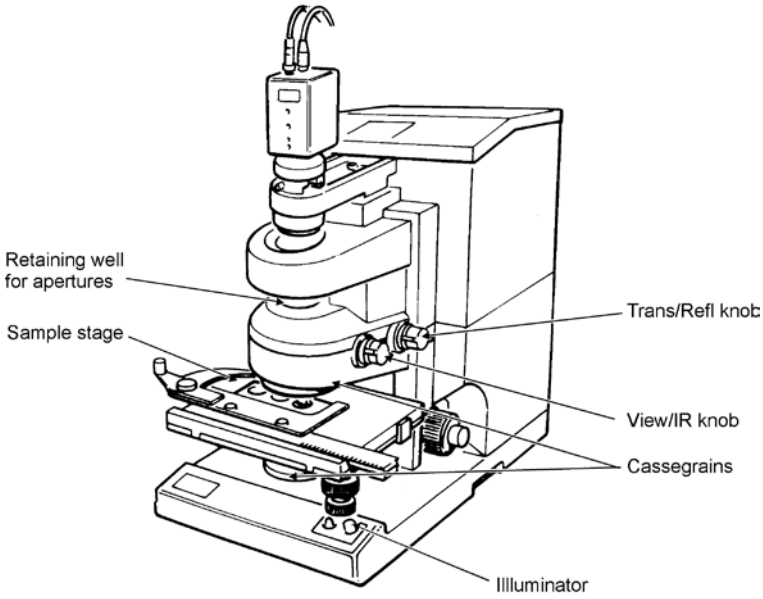

Figure 4 The MultiScope System Microscope

The MultiScope System Microscope (Figure 4) includes the following features:

- High-performance PerkinElmer collection optics for infrared microspectroscopy. The cassegrain mirror systems used in the MultiScope System Microscope have a wide collection angle (high numerical aperture) for efficient collection of radiation.
- Video viewer linked to a framegrabber for optical microscopy.
- MCT detector. Gives spectra with high signal-to-noise ratio.
- No mechanical errors in switching from viewing to infrared. The same cassegrain is used to send both infrared radiation and visible light to the remote aperture. The location of the image does not change when you switch from viewing to infrared.
- High signal-to-noise ratio. Noise is reduced by having a small target (0.25 mm) detector to which the infrared image from the cassegrains is very well matched.

- Larger working distance for thick samples. The lower cassegrain can be removed to provide space for thick samples to be studied in reflectance.
- Spectra can be collected in either reflectance or transmittance modes. A micro-ATR accessory is available as an optional extra.
- Internal coaxial illumination, with variable intensity.
- Whole-field aperture kit with independent variable knife-edge apertures. The apertures are transparent to visible radiation, but opaque to infrared, making it easier to locate the point of interest on the sample.

Applications

Because the microscope enables you to collect spectra of very small samples, and can collect both reflectance and transmittance spectra, it is useful in a wide variety of applications. Some examples are listed below, and several of these are described fully in *Applications* on page 109.

- Reflectance FT-IR microscopy, including
  - diffuse reflectance spectra of powders
  - specular reflectance spectra of coated and uncoated materials
  - reflection/absorption spectra
- Polymers
- Textiles
- Criminalistics (forensic work), including
  - transmittance spectra of fibers
  - paint chips
  - drugs
- Biological materials
- Product contaminants, including
  - microcontaminants on silicon wafers
  - contaminants on electrical contacts
  - spots in paper
  - imperfections in polymer films

The Optical System

The MultiScope System Microscope combines two optical systems: PerkinElmer optics for infrared microspectroscopy and a video viewer. The two systems intersect at the remote aperture (Figure 5). When a sample on the sample stage is in focus, its conjugate image is focused at the remote aperture. In visible light, you see this image through the video viewer; in infrared, this image is sent to the detector.

You select between visible light (viewing the sample) and infrared (collecting a spectrum) by moving only one control, the View/IR knob. The selection between transmittance and reflectance is equally simple. This section describes what happens within the optical system when you make these selections.

Viewing

When you view a sample with the video viewer, you are really looking at the conjugate image of the sample, located at the remote aperture.

You focus with the Focus knob of the microscope. This moves the sample stage up and down until the conjugate image is focused at the remote aperture.

You then position the area of the sample that is of interest and isolate it by masking off unwanted areas, using the variable knife-edge aperture.

Viewing in transmittance

When the microscope is set for viewing in transmittance (Figure 5):

- Mirror M3, beneath the cassegrains, receives light from the illuminator and directs it up through cassegrain C1.
- Cassegrain C1 condenses the beam to an appropriate size for a microscopic sample and focuses it at the sample position.
- Cassegrain C2 collects light from the sample and sends it upward through the remote aperture. The light has an unobstructed path from C2 to the optical microscope.

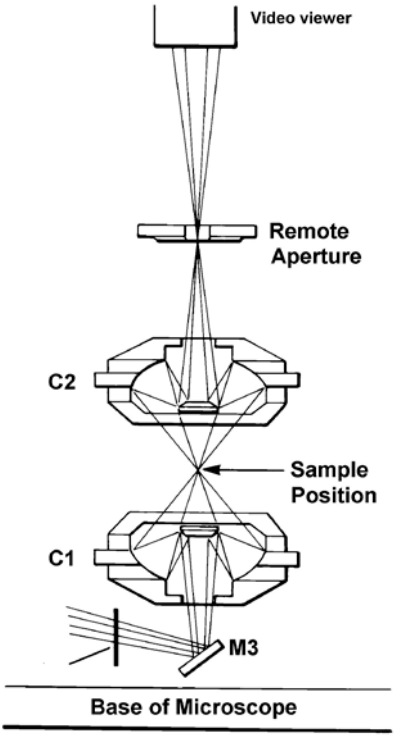

Figure 5 Path of the Visible Beam for Viewing a Sample in Transmittance

- While viewing, laser and infrared radiation are blocked from reaching the sample.

Viewing in reflectance

When the microscope is set for viewing in reflectance (Figure 6):

- Mirror M4, above the cassegrain, receives light from the illuminator and directs it down through the cassegrain C2. M4 is a relay mirror that directs the beam down one side of cassegrain C2 to the sample.
- Cassegrain C2 collects light from the sample and sends it upward through the remote aperture.
- While viewing, laser and infrared radiation are blocked from reaching the sample.

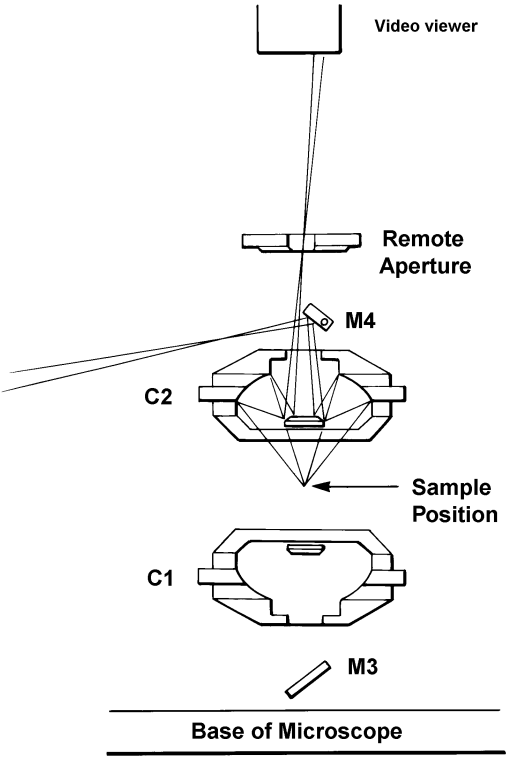

Figure 6 Path of the Visible Beam for Viewing a Sample in Reflectance

Collecting a Spectrum

The same cassegrain is used for both visible light and infrared radiation. For this reason, when you adjust the sample position so that the visible image of the sample is in focus, the sample is also correctly positioned for collecting the infrared spectrum. Similarly, when you adjust the aperture so that the required area of the sample is isolated visually, you have also isolated the area of the sample of which the spectrum is to be collected.

To collect a spectrum, you use the View/IR knob to move the detector mirror into the beam (Figure 7). It blocks the beam from reaching the optical microscope and reflects it toward the detector, then the MCT cassegrain focuses the beam on to the detector.

Collecting a spectrum in transmittance

Collecting a spectrum in transmittance differs from viewing in transmittance as follows (Figure 7):

- Instead of receiving light from the illuminator, mirror M3 receives the infrared beam from toroid M2, which is set to direct the beam downward.
- The detector mirror is moved into the beam above the remote aperture.
- The MCT cassegrain focuses the beam on to the MCT detector.
- In transmittance mode, the reflectance mirror, M4, is completely removed from the beam.

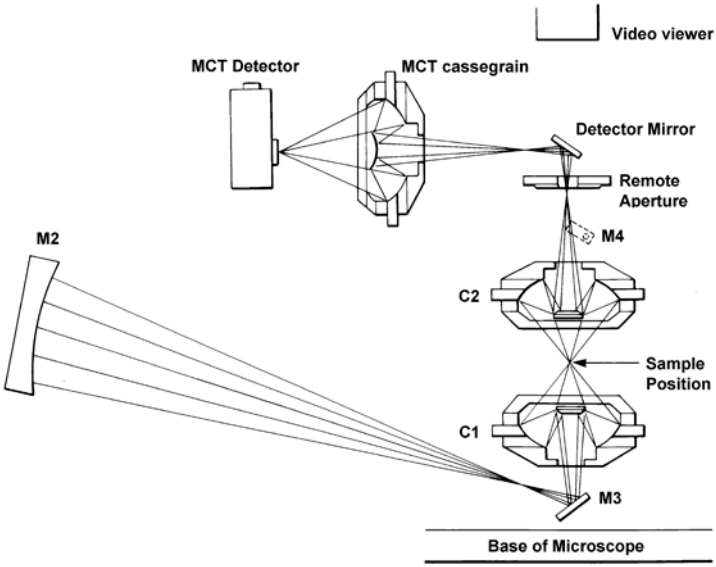

Figure 7 Path of the Infrared Beam for Collecting a Spectrum in Transmittance

Collecting a Spectrum in Reflectance

Collecting a spectrum in reflectance differs from viewing in reflectance as follows (Figure 8):

- The sample does not receive light from the illuminator. Instead, the infrared beam from M2 is directed to M4.
- The beam is reflected off the sample and back through the other side of the cassegrain, toward the remote aperture.
- The detector mirror is moved into the beam above the remote aperture.
- The MCT cassegrain focuses the beam on to the MCT detector.

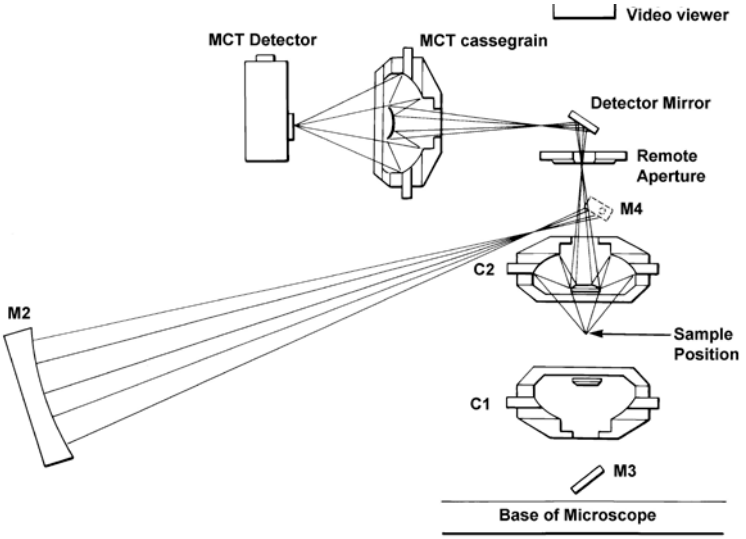

Figure 8 Path of the Infrared Beam for Collecting a Spectrum in Reflectance

Specifications

Sample stage

Manual stage: Range of travel = 75 mm × 50 mm
Working distance: 25 mm
Working distance for thick samples: 30 mm (with lower cassegrain removed)
The manual stage is equipped with a sample clip and is calibrated in 1 mm increments.

Microspectroscopy modes

Transmittance and Reflectance (Standard)
Micro-ATR objective (Optional)

Objective and condenser cassegrains

Specifications for cassegrain collection optics: magnification 6×, 0.60 numerical aperture, permanently aligned, factory-set focal separation. The substage condenser cassegrain has a variable focusing control.

MCT detector cassegrain

On-axis design, permanently aligned, factory-set focal separation.

Apertures provided

Whole-field aperture kit with variable independent knife-edge aperture.

Illuminator

Mounted in microscope.
Continuously variable intensity.
Coaxial illumination for transmittance and reflectance.
Power 35 W.

Optical microscope specifications

Video viewer fitted with video camera linked to a framegrabber board for display of visible image on PC screen.

Detector

Choice of three 0.25 mm MCT detectors cooled by liquid nitrogen: wide band, medium band and narrow band spectral ranges:

Narrow Band	10 000 to 700 cm^{-1}
Medium Band	10 000 to 580 cm^{-1}
Wide Band	10 000 to 450 cm^{-1}

Getting Ready to Use the Microscope

Getting Ready to Use the Microscope

This chapter gives the routine procedures for getting ready to collect spectra with the MultiScope System Microscope. It includes procedures for:

- Setting up the FT-IR for use with the microscope;
- Cooling the MCT detector;
- Viewing the sample.

Use these procedures at the beginning of the day's work, or any time the microscope has not been in use or has been used by others.

At the end of the chapter is a list of continuing adjustments that you can make at any time that you are using the microscope.

Setting Up the FT-IR

1. If necessary switch on the FT-IR.
2. Leave the source to warm up as directed in the manual for the FT-IR.

   The power supply to the detector of the microscope is also switched on through the FT-IR.

NOTE: It can take an FT-IR up to 2 hours to equilibrate after it has been switched off overnight. To save time, we suggest that you leave the FT-IR switched on at all times.

3. Select the external beam of the FT-IR:
   - Spectrum 100 Series and Spectrum One: Select the beampath to the microscope using the Beam tab of the Scan and Instrument Setup dialog.
   - Model 1600, Model 16PC, Spectrum BX or Spectrum RX:
     Move the external beam lever, at the front lower left of the FT-IR, as far to the left as possible.
   - Spectrum GX: Select the beam path to the microscope using the Spectrum GX Instrument Setup tabs in the Spectrum software.

Cooling the MCT Detector

An MCT (Mercury Cadmium Telluride) detector is standard in the MultiScope System Microscope. The MCT detector must be cooled to 77 K before you can operate it. It is mounted in a dewar that can be filled with liquid nitrogen. As you fill the dewar, the temperature of the detector drops, and the preamp supplies power to the detector. Use the following procedure to cool the MCT detector.

WARNING

The extremely low temperature of liquid nitrogen can burn skin and eyes. Avoid exposure by wearing heavy gloves and safety goggles whenever you work with it.

WARNING

When liquid nitrogen warms to room temperature, nitrogen gas vaporizes so rapidly that resulting pressures can send a funnel or detector cap suddenly, and forcefully shooting upward from the top of the microscope.

WARNING

Be sure to wait the specified time when filling the funnel and before replacing the detector cap. This enables the bubbling nitrogen to settle down and the pressure to dissipate. In addition to wearing safety goggles at all times, stand back from the microscope after each time you fill the funnel.

Cooling the MCT detector

1. Press on the front edge of the lid of the dewar, which then opens.
2. Lift off the cap of the dewar.
3. Place the small funnel supplied with the microscope in the opening in the red detector dewar (Figure 9).

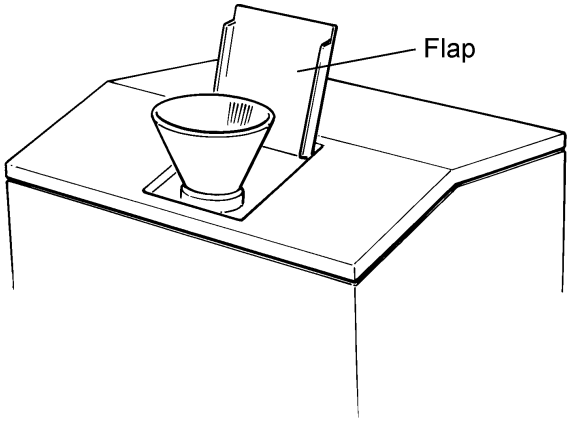

Figure 9 The Dewar Opening with the Funnel Inserted

WARNING

Stand where you can see the inside of the funnel as you pour the nitrogen in. Pour slowly, so that neither the funnel nor the dewar overflows. If liquid nitrogen runs down the outside of the dewar, it can damage the optics of the microscope.

4. Carefully fill the funnel with liquid nitrogen. Stand back and let the funnel empty completely.

   The liquid nitrogen bubbles rapidly as it drains into the dewar. This initial quantity of liquid nitrogen vaporizes completely as it cools the dewar.

5. Add another one and a half funnels of liquid nitrogen. Stand back and wait two minutes.

   This nitrogen also vaporizes as the dewar continues to cool. The two-minute wait enables the bubbling to settle down and the pressure of the vaporizing nitrogen to dissipate.

6. Continue to pour liquid nitrogen into the funnel, adding a little more each time the funnel empties.

   The funnel takes longer to empty as the dewar fills. This happens after two to three more funnels of liquid nitrogen.

   Because the dewar has now cooled, the liquid nitrogen does not vaporize, but instead fills the dewar.

7. Remove the funnel and wait two minutes.

   The liquid nitrogen settles down and bubbling slows.

8. When the nitrogen stops bubbling, refit the detector cap.

   The filled dewar cools the MCT to the correct operating temperature for at least eight hours. After that, the dewar begins to return to room temperature, and the preamp switches off power to the MCT.

Setting Up the Microscope

1. Switch on the microscope at the switch on the rear.

   The Power On light at the front is lit.

2. Make sure that the video camera power supply unit is connected to a mains electrical supply, and the video camera is connected to the power supply unit.

3. If you are using a monitor, switch on the monitor; if you are using a PC, switch on the PC.

Viewing the Sample

You can view the visible image of a sample in a window on a PC screen by using the video viewer, which comprises a video camera that is linked to a framegrabber board in the PC. For full details of the use of the video display software see the Users Manual which is provided. The video viewer, framegrabber card and software are installed and set up by your PerkinElmer Service Engineer.

The video display software enables you to:

- View the sample on the PC screen in Windows;
- Resize the visible image window;
- Store visible images as bitmaps and in other file formats. This enables you to archive visible images along with IR data;
- Copy and paste visible images to other Windows packages.

The field of view of the microscope that is displayed on the PC has dimensions of 600 µm (X-axis) and 400 µm (Y-axis) and the microscope is set-up so that the infrared beam is aligned with the center of the field of view. For all infrared measurements, use the stage controls' position, the sample (or the area of interest on a larger sample) in the center of the field of view.

Tutorial: Using the Microscope

Introduction to the Tutorial

This tutorial describes how to prepare two frequently-studied types of samples (fibers and thin films) and collect their spectra. In this tutorial you will:

- Learn some techniques for preparing samples;
- Use several of the tools in the sample preparation kit, if you purchased the kit with your microscope;
- Use the microscope to collect both transmittance and reflectance spectra.

Getting Ready

Before you begin this tutorial:

- Switch on the FT-IR and the microscope;
- Switch on the power supply of the video camera;
- Switch on the monitor if used;
- Make sure that the software that controls the FT-IR and the Win-TV framegrabber software are open on the PC;
- Select the external beam of the FT-IR;
- Fill the dewar of the MCT detector with liquid nitrogen;
- Familiarize yourself with the positions of the controls on the microscope;

All of these procedures are in *Getting Ready to Use the Microscope*.

Using Optional Equipment

This tutorial does not include special instructions for any optional equipment you may have with your microscope.

Transmittance Spectrum of a Flattened Fiber

In this section you prepare a flattened fiber sample and collect its transmittance spectrum. You can use any convenient organic fiber as the sample in these procedures, provided it is small enough in diameter to give a transmittance spectrum. Wool or nylon from a sweater is ideal; polyester fibers are suitable; hair may be too thick.

Flattening a fiber sample before you collect its spectrum decreases its optical density. Flattening also increases the area, so that you need fewer scans to obtain an acceptable signal-to-noise ratio.

Other Equipment Needed

Scissors
Roller knife (or other tools for cutting and flattening) or Scalpel
Forceps
Sample holder (3x1)
Glass microscope slide
Adhesive tape (Sellotape or Scotch tape)

The roller knife and forceps are included in the microscope sample preparation kit (N187 0151); for more information contact your local PerkinElmer office or agent. The sample holder is provided with the Microscope.

Preparing the flattened fiber sample

1. With forceps, place the fiber on the microscope slide.
2. Hold the fiber with the forceps and use the knife end of the roller knife to cut the fiber so that it is just long enough to stretch across one of the holes on the 3 x 1 sample holder.
3. Flatten a short section of the fiber against the glass with the roller end of the roller knife or the handle of the scissors.
4. With forceps or a probe, transfer a flattened piece of fiber to the 3 x 1 sample holder.
5. Stretch the sample across the hole and, using small pieces of adhesive tape, attach the fiber to the sample holder.

   The fiber should be under tension and the flattened part should be roughly in the middle of the hole.

   The sample is ready to use.

Position and focusing the sample

1. Set the microscope for viewing a sample in transmitted light: switch the Trans/Refl knob to Trans and the View/IR knob to View.
2. To position the sample stage correctly, look from the front with your naked eye and adjust the stage in the x and y directions until you see light striking the sample.
3. As you move the stage in the x and y directions, look at the visible image to center the fiber in the field.
4. Looking at the visible image, bring the fiber into the sharpest focus possible by turning the Focus knob.
5. Place the variable knife-edge aperture in the retaining well.
6. The aperture has blades and knife edges that are transparent to visible radiation, but opaque to infrared.
7. Looking at the visible image, select a flat, transparent portion of the fiber. Adjust the knife edges until this portion fills the aperture.

Collecting the Background Spectrum

You are ready to collect a background spectrum for this sample.

1. Using only one of the stage controls (x or y), move the sample out of the aperture.

   The background spectrum is collected through air using the same aperture.

   Two metal scales along the front and left sides of the stage give the x and y coordinates of the center. Each scale has a vernier with 100 µm resolution. If you note the location of the sample before you move it, you can return it very near to the same location later.
2. Switch the View/IR knob to IR.

Correcting the microscope for focal shifts

When the *sample* for a transmittance spectrum is mounted on a window of KBr (or another substance), you must correct the microscope optics for focal shifts that occur as the infrared beam comes through the window. Although the sample that you are studying is suspended in air, it is good practice to make sure that the correction control of the microscope is correctly set before you collect a background or sample spectrum. This is essential when the sample or its support have a different refractive index (RI) from air.

1. Display the energy bar produced by the Monitor command of the FT-IR.
2. Adjust the Correction knob on the right near the microscope base until the energy is at a maximum. Do not leave Monitor yet.
3. Adjust the gain to give maximum signal:
   - With the Spectrum 100 Series or Spectrum One, the number must be as near as possible to, but not more than, 6000.
   - With the Model 1600 or Spectrum RX, the number must be as near as possible to, but not more than, 100. If the signal does not meet this requirement, return to Ready For Next Command and use SETUP SCAN OTHERS GAIN to adjust the gain.
   - With the Model 16PC or Spectrum BX, the number must be as near as possible to, but not more than, 10000. If the signal does not meet this requirement, display the Setup menu and choose Instrument, then click on Gain to increase the gain.
   - With the Spectrum GX, the number must be as near as possible to, but not more than, 6000. If the signal does not meet this requirement, display the Setup menu and choose Instrument, then click on Gain to increase the gain.
4. Return to Monitor to make sure the number is not greater than numbers given in step 3.
5. When the gain is correct, collect a 16-scan background spectrum.

Collecting the Sample Spectrum

Before you can collect the sample spectrum, you must once again center the sample in the field visually.

1. Switch the View/IR knob to View.
2. Return the sample to the center of the field. Do not change the size of the aperture.
3. Switch the View/IR knob to IR.
4. Use the SCAN command to collect the spectrum of the sample; collecting the same number of scans as you collected for the background.
5. Remove the variable knife-edge aperture and the sample from the microscope.

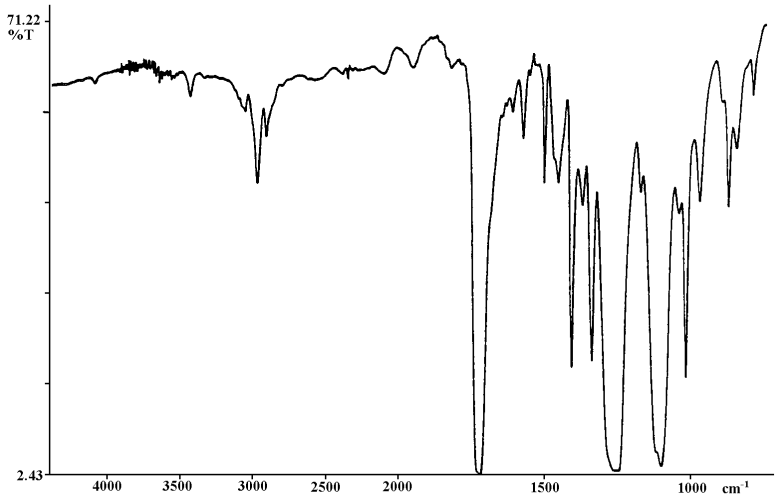

Figure 10 Spectrum of a Flattened Poly(Ethylene Terephthalate) Fiber

Reflectance Spectrum of a Thin Film

Now you are going to collect the reflectance spectrum of a thin polymer coating on a reflective surface. A convenient sample to use for these procedures is an aluminum soda (drinks) can. The surfaces of such cans are coated with a polymer film.

Other equipment needed:

Scissors
Reference mirror

Preparing and positioning the sample

1. With scissors, cut a piece about 7 mm square from the side of a soda (drink) can.

   If it is noticeably curved, flatten it.

WARNING

Be careful. The edges of the cut piece can be sharp.

2. Place the 3 x 1 sample holder on the sample stage of the microscope, reflective side up, as in Figure 11.

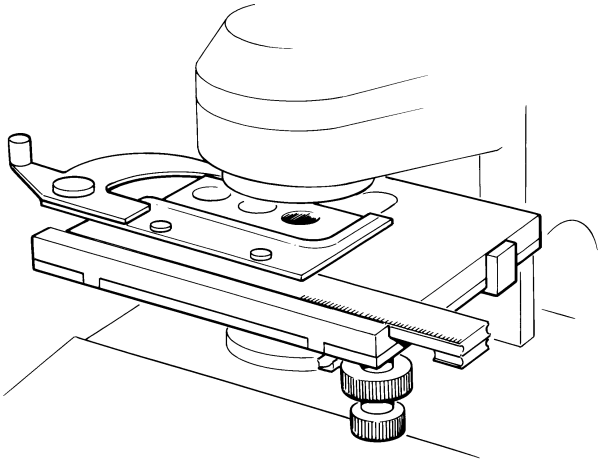

Figure 11 Reference Mirror on the Sample Stage

3. With forceps, set the sample over one of the unused holes on the 3 x 1 sample stage, outer (painted) side up.
4. Switch the View/IR knob to View.
5. Switch the Trans/Refl knob to Refl.
6. Looking at the visible image, focus on the surface of the mirror with the Focus knob.

   There are almost always dust particles or scratches on the surface that you can use to focus on.
7. Move the stage in the x and y directions with the stage controls until the sample is in the center of the field of view.
8. Focus on the sample with the Focus knob.
9. Place the variable knife-edge aperture in the retaining well.

Because the sample is large and has uniform areas, you do not need to use it all or to isolate the best portion; just adjust the knife edges to give a convenient aperture size (50 μm).

1. Looking at the visible image and using only one of the stage controls (x or y), slowly move the sample until it is out of the aperture and the reference mirror is seen.
2. Using the Focus knob, focus on the mirror surface.

NOTE: Do NOT adjust the *Correction* knob or *Focus* knob again. The *Correction* knob adjusts the cassegrain below the sample, and in reflectance mode the infrared beam does not pass through this. In reflectance mode, the infrared beam and the visible image are both focused using the *Focus* knob.

3. Turn the View/IR knob to IR.
4. Adjust the gain to give maximum signal (see *Transmittance* Spectrum *of a* Flattened Fiber on page 54).

NOTE: If, with the gain at its lowest setting, the instrument reports ***overload*** or ***gain too high***, you must adjust the knife edges to make the aperture smaller.

5. When the gain is correct, collect a background spectrum (16 scans).

Collecting the Sample Spectrum

1. Switch the illuminator on again.
2. Switch the View/IR knob to View.
3. Return the sample to the center of the field by moving the same stage control you used to move it out of the field. Do not change the size of the aperture.
4. Even though the microscope may not be focused on the sample, you can see a change in the light intensity as the sample moves into the aperture.
5. Use the Focus knob to focus on the sample.
6. Switch the View/IR knob to IR.
7. Collect the spectrum of the sample. (16 scans)

   Figure 12 shows a typical result.
8. Remove the variable knife-edge aperture, the reference mirror and the sample from the microscope.

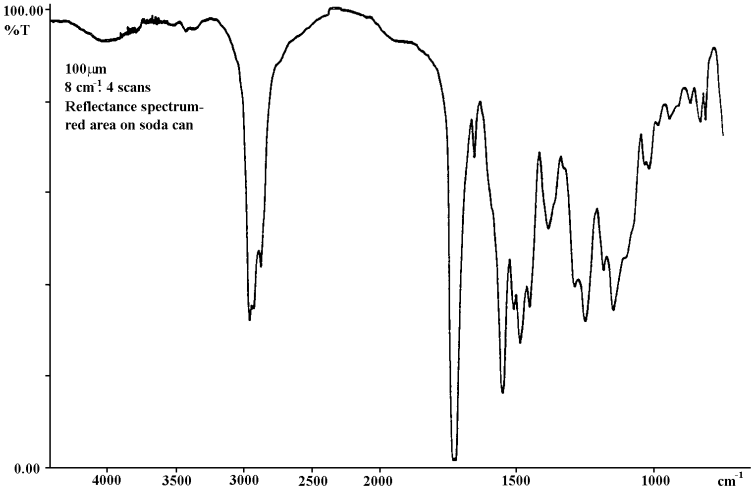

Figure 12 Reflectance Spectrum of the Coating on a Soda Can

Preparing Samples

Preparing Samples

Using a microscope for infrared spectroscopy can simplify sample preparation, because you can examine the sample visually and select the best part to use. Proper sample preparation is still important, however, if you are to collect good quality spectra.

- The sample must be thin enough (approximately 5 to 20 µm) to give good detail and undistorted absorption bands.
- The area of the sample must be large enough to give an adequate signal; otherwise, the scan time must be increased.

Preparing a sample, therefore, often involves flattening it; this both thins it and increases its area. Flattening is accomplished by rolling, squeezing, or pressing the sample.

This chapter tells you how to prepare samples for spectroscopy with the microscope. It includes:

- A list of useful tools;
- A list of window materials commonly used for mounting samples;
- Descriptions of special techniques used to prepare particular types of samples.

For hands-on experience in preparing samples of fibers and of coatings on reflective surfaces, we suggest that you try the tutorial in *Tutorial: Using the Microscope*.

Tools for Sample Preparation

This section lists the tools you need for preparing samples:

- Sampling accessories provided with the microscope;
- Sampling accessories available as options;
- Tools in the microsampling toolkit;
- Materials to have available;
- Specialized accessories you may want to purchase;
- Tools you can make yourself (with procedures for making them).

Tools Provided with the Microscope

The following items for use in sampling are provided supplied with the microscope:

Holder for 13 mm disks with gold mirror - supports 13 mm disks on the sample stage and provides a reference mirror for reflectance measurements.

Sampling Accessories Available as Options

The following items for use in sampling are available in accessories kit, part number L186 0250:

Item	**Use**
Holder for 13 mm disks	Supports 13 mm disks on the sample stage
Gold mirror in holder	Reflection measurements
Slides, glass (box)	Support samples for sample preparation
Rotating 13 mm disk holder	Supports samples and allows rotation
Support for large samples	Clips on to the sample stage. Supports bulky samples so that the stage clip does not interfere with them
KBr windows	Support samples
100 µm aperture	fixed-size circular aperture
Low magnification screen	Viewing samples
Variable knife-edge aperture	Isolating sample areas

The following tools are in the sample preparation kit for infrared microscopy N187 0151, see Figure 13.

Tool	**Use**
Forceps, 4½ inch, Cd plated	Picking up small objects
Roller knife	Cutting (knife end) and flattening (roller end)
Steel tweezers	Picking up extremely small objects
Tungsten alloy needle	Transferring particles
Steel probe	Pulling samples apart, separating fibers
Interchangeable handle for micro tools	Handle for tungsten needle or steel probe
Pin vise	Holds needles for sharpening or for flattening samples

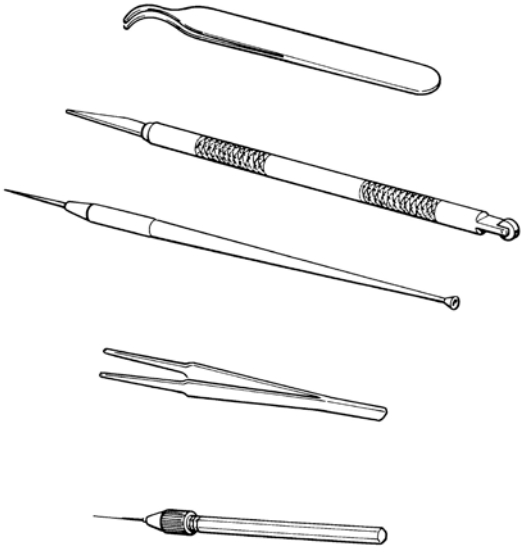

Figure 13 Tools in the Microsampling Toolkit

Other Useful Tools

Depending on the type of samples that you usually work with, it may be helpful to have some of the following tools:

Tool	Part Number
Wide-tipped forceps, hooked	0990 8138
Wide-tipped forceps, flat	0990 8400
1.5 mm microdisk. Fits in 13 mm disk holder to support very small samples.	0186 1043
Fiber slit, 18 µm	L116 0581
Fiber slit, 33 µm	L116 0582
Pinhole, 500 µm	L116 0586
Pinhole, 1000 µm	L116 0587

The items listed below are available from PerkinElmer. For more information, contact your local office or agent.

Tool	Part Number
Microprobe with right angle bend	N930 2606
Forceps, round tips	N930 2607
Forceps, narrow needle points	N930 2608
Windows: All 13 mm diameter	
BaF_2 (1 mm thick)	N930 2611
BaF_2 (2 mm thick)	N930 2612
ZnSe (1 mm thick)	N930 2613
NaCl (2 mm thick)	N930 2614
KBr (2 mm thick)	N930 2615

Specialized Accessories

The following accessories are extremely useful in preparing certain types of samples (as described in *Techniques for Preparing Samples*, on page 72):

- Miniature Diamond Anvil Cell (N930 2618)
- Microtome

Items to Have Available

In addition to the items provided with the microscope, we recommend that you have the following available:

- Tape with adhesive on both sides ("double-sided tape") for holding long or large samples on the sample stage;
- Single-edge razor blades for cutting samples.

Tools You Can Make Yourself

This section gives procedures for making the following tools and equipment used in microspectroscopy:

- Micropipettes, used for transferring liquids;
- Salt plates, used as mounts for a wide variety of samples;
- Tungsten needles, used for transferring particles;
- Microbrushes, used in pyrolysis and in transferring minute amounts of liquid.

Micropipettes

You can purchase micropipettes, or you can make them yourself in one of the following ways:

- Heat glass tubing, draw it to the desired diameter, and break it.
- Slowly heat polyethylene tubing that has a narrow melting range, draw it to the desired diameter, and cut it with a razor blade.

Salt windows

If you have a pellet press, you can use it to make KBr windows.

Tungsten Needles

You can make tungsten needles by sharpening 24 to 26 gauge (0.5 mm) tungsten wire. This is done chemically, by etching the wire with sodium nitrite (or ammonium nitrite) to produce a smooth, fine, stiff-pointed needle.

Use the following procedure to sharpen a tungsten wire:

WARNING

Wear eye protection during this procedure. The reaction between tungsten and sodium nitrite is strongly exothermic; hot material may spatter. Also, these needles are much sharper than conventional needles and must be handled with great care.

1. Place powdered sodium nitrite in a vial.
2. Insert the tungsten wire or needle in the pin vise provided in the Microsampling Toolkit.
3. Heat one end of the needle until it is red.
4. Quickly insert the hot needle into the sodium nitrite.
5. Wash the salt residue from the needle with water.
6. For convenience, sharpen the other end of the needle also, so that you can reverse the needle in its mount when the first tip becomes damaged.

Microbrushes

To make a microbrush for transferring small amounts of liquid or for pyrolyzing samples:

1. Use a thin wire to push fibers of borosilicate glass wool about 3 cm into the end of a 200 µm by 10 cm capillary tube. Leave the fibers protruding from the tube.
2. With a hobbyist's microtorch, fuse the glass wool to the sides of the capillary.
3. Trim the protruding fibers to about 1 cm with scissors.

The use of the microbrush is described in the section *Techniques for Preparing Samples* on page 72.

➢ To clean the brush, ultrasonicate it in a suitable solvent.

Common Window Materials

Both liquid and solid samples are often mounted on salt windows. Very thin windows, 1 to 2 mm thick, give the best spectra. The following materials are commonly used in windows:

- KBr: Potassium bromide is inexpensive, and it transmits infrared radiation to below 400 cm^{-1}. The major disadvantage of this material is that it is hygroscopic, so that the windows fog easily.
- BaF2: Barium fluoride is not hygroscopic. Its transmittance cut-off is 750 cm^{-1}, which means that it is best suited for use with a narrow band MCT detector, although not with a wide band MCT. It can break or crack easily.
- NaCl: Sodium chloride transmits infrared down to 600 cm^{-1}. Otherwise, its properties are similar to KBr.
- ZnSe: Zinc selenide is not hygroscopic. Its transmittance cut-off is 650 cm^{-1}. ZnSe is more durable than the other windows but is yellow, so that the field of the optical microscope appears yellow.

Techniques for Preparing Samples

This section describes some useful techniques for preparing various types of samples.

Flattening Solids

Samples that are too thick to give good infrared spectra can often be flattened by pressing or squeezing. Because the samples are usually quite small, only moderate force is necessary.

Rolling with the roller knife

The roller end of the roller knife provided in the microsampling tool is one of the simplest and most effective devices for flattening samples. It is especially useful for flattening fibers or particles.

Different types of sample may be treated in different ways:

- If the sample is soft, roll it on a small salt window.
- If the sample is hard, roll it on a hard surface, such as glass or metal. A flat, black cap from a jar makes a good surface for rolling a light-colored sample.
- If the sample is rolled on a small, flat piece of metal it can be viewed and spectra measured in reflection mode. Samples rolled on windows transparent to infrared can be examined in transmittance.
- Fibers can be flattened on a glass microscope slide, then peeled off and mounted either on a window or over the aperture for the microscope slide.

Squeezing with a pellet press

Samples can be squeezed between the polished anvils of a KBr pellet press without KBr. To collect the spectrum use either of the following methods:

- Peel the flattened sample off the anvil with a probe or knife and place it on a sample mount. Collect the transmission spectrum.
- Leave the sample on the polished anvil and collect the reflection spectrum. Use a clean area of the anvil as the reference.

Using a diamond anvil cell

See the procedure under *Polymers* (page 76) for flattening samples with a miniature diamond anvil cell.

Compressing between infrared transmitting windows

Pressing two windows together, with the sample between them, compresses the sample. This also provides optical contact between the windows and the sample, reducing surface scattering.

Windows made of NaCl or KBr are relatively soft. If your sample is hard, or if it is wet, use BaF_2 or ZnSe.

Pressing with the heel of a probe

Press on small samples with the flat end of the probe handle. Even moderate pressure usually produces considerable thinning.

Pressing with a needle

Pressing with the point of a needle or probe applies a high force per unit area, because the area of contact is small.

Rolling a hard sample with the side of a sewing needle held in a pin vise presses it into a flake.

Slicing Samples from Solids

Cutting a wedge of sample

If a solid sample is too thick to give good spectra, cutting a wedge-shaped piece from its edge produces a thin sample while destroying very little of the original. This technique can be used with laminates, plastics, films, paint chips, paper.

To cut a wedge-shaped piece from a relatively thick sample:

- Hold the sample in tweezers as you slice a thin wedge from it with a razor blade. Taper the wedge to as thin a slice as possible.

To cut a wedge-shaped piece from a relatively thin sample:

1. Place the sample between two offset glass slides.

   Allow a triangular portion of the sample to protrude as shown on the left in Figure 14.

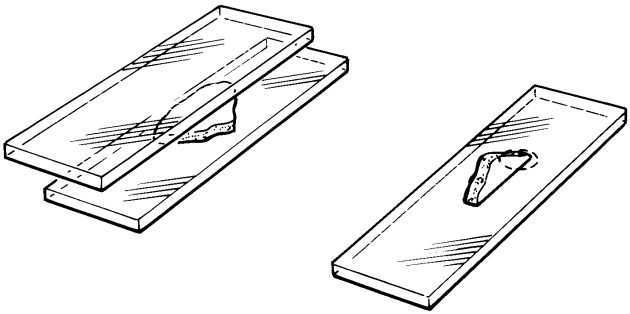

Figure 14 Cutting a Wedge-Shaped Sample

2. Run a razor blade or the roller knife along the edge of the upper slide.

   The triangular piece of the sample is sliced off, giving a wedge-shaped sample.

3. To mount the sample, rotate it as shown on the right in Figure 14. Position it under the microscope so that the infrared beam goes through the thin end of the wedge (circled in Figure 14).

Microtoming

A microtome is a device for slicing a sample into thin cross-sections, 0.5 to 20 µm thick. It is commonly used to prepare samples for light microscopy; the same range of thicknesses is also appropriate for infrared microscopy.

If you are trying to identify the individual components of a laminate, microtomed samples give the best results.

Samples are often embedded in a supporting medium before they are microtomed. If you must use an embedding material, choose it carefully, so that it does not alter the sample by reacting with it, dissolving it, or contaminating it. Some commonly used materials are:

- paraffin wax: This is the preferred medium for infrared spectroscopy. It produces few spectral interferences, and it can usually be easily removed from the sample with warm xylene.
- β-pinene wax: This material is similar to paraffin.
- plastic embedding materials: These can be used depending on the size and porosity of the sample.
- acrylic and epoxy resins: Although these are commonly used in light microscopy, they are not recommended for infrared, because they are hard to remove and can cause spectral interferences.

Polymers

Diamond anvil cell

Polymer samples such as paint chips, thick films, elastomers, or fibers can be pressed or squeezed to reduce their pathlength.

A convenient device for pressing polymers (or other compressible samples) is the miniature diamond anvil cell, shown in Figure 15. It enables you to both thin the sample and collect its spectrum in the same device, an advantage when you have limited material available.

It is small enough to be easy to manipulate, and fits in the recessed retainer in the support for large samples. By collecting a background spectrum of an empty area of the cell, you can completely compensate for the absorption bands of the diamonds.

To thin a sample in the miniature diamond anvil cell:

1. Loosen and remove the three screws that hold the cell together.
2. Lift off the top half of the cell and set it aside.
3. Place the sample on the bottom half of the cell. (The sample must be small.)
4. Put the top half back on the cell, lining up the red dots on the top and bottom halves.

   Do not tighten the screws yet; applying uneven sheer forces may damage the diamonds.

CAUTION *One diamond can damage another.*

5. Press down on the cell with your thumb to thin the sample. Replace the three screws and tighten them finger tight.

NOTE: If the spectrum collected with the diamond anvil cell shows interference fringes, place some KBr in the cell and collect a background spectrum through it.

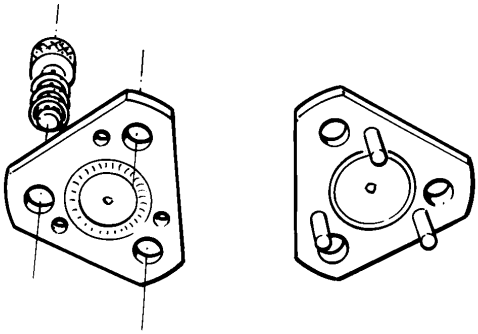

Figure 15 The Miniature Diamond Anvil Cell

Pressing elastomers between windows

If your sample is elastic and you are compressing it between windows, you need a way to apply pressure continuously. Use the following procedure:

1. Press on the windows with a probe, flattening the sample.
2. While maintaining the pressure, apply small amounts of quick-setting nitrocellulose cement to the edges of the salt plates.

   When the cement is dry, the sample remains compressed.

The compression cell (see *Special Cells*, on page 81) is designed to compress this kind of sample.

Filled Polymers

When a polymer contains a high concentration of fillers, you have to prepare a sample for analysis that is free of filler.

Often you can obtain a suitable sample by cutting a thin wedge of the material with a sharp blade. If the filler is not uniformly dispersed, you can find clear regions of polymer for analysis.

Pyrolysis can be used to remove the fillers. As you heat the sample, the polymer volatilizes, and the fillers are reduced to ash. The pyrolysis can be accomplished in any of the following ways:

- Place the polymer in a disposable pipette and seal the large end. When this end cools, tap the polymer into it, then heat the sample gently.
- The pyrolyzed polymer condenses on the walls of the pipette. The filler is left behind as ash.
- Score and break the pipette between the ash and the pyrolysate. Add a drop of solvent to the pyrolysate to wash it on to a salt plate.
- If the amount of sample is small, use a capillary tube instead of a disposable pipette for the above procedure.
- Use a microbrush to pyrolyze micro amounts. Seal the end that is away from the brush fibers and tap the sample particle into this end then heat it gently with a microtorch. After pyrolysis, break off and discard the end of the tube that contains the ash. While holding the fibers of the brush against the salt plate, add a drop of solvent to the broken end. Allow the solvent containing the pyrolysate to flow into the fibers. When it evaporates, the pyrolysate is left on the salt plate for analysis.

Particles

Crushing

Samples such as large particles that cannot be sliced can often be crushed to thin them. This can be done in various ways:

- If the sample is small, crush it with the roller end of the roller knife.
- If the sample is larger, use a pestle and mortar.

Separating by aperturing

Powders and other particulate solids may contain several different components. Instead of separating them, use the variable knife-edge aperture to isolate the component you want to sample:

1. Spread the sample out with a probe so that you can visually distinguish the components.
2. Looking through the microscope, find a particle of the component you want to sample.
3. Center this particle in the field of view.
4. Adjust the variable knife-edge aperture until only the particle that is of interest is visible.

Extremely small particles are easy to pick up and transfer with a very fine-pointed tungsten needle. Scoring the surface of the salt plate with the needle makes a simple map to help you positively identify the particles under the microscope.

Transferring with a tungsten needle

When necessary, sharpen the tungsten needle as described in *Tools You Can Make Yourself* (see page 69).

Nujol or fluorolube mulls

Suspending fine particles of a solid sample in nujol or fluorolube reduces or eliminates the surface reflections that can distort absorption measurements. These oils also reduce the amount of radiation lost to reflection or scattering.

If the film is thin enough, you can correct the spectrum for the presence of the oil by subtracting a spectrum of the pure liquid. It is difficult, however, to obtain the correct thickness for a good subtraction.

Fibers

Fibers can be rolled to flatten them (see *Flattening* Solids on page 72), or they can be pressed in a diamond anvil cell (see *Polymers* on page 76). The following is another approach.

Fibrous Solids

If a fibrous sample, such as paper, is too thick, tear it and examine the torn edges. The edges contain single fibers and thin clumps of fibers.

Coatings on Substrates

If the sample is coated on a substrate, the method for collecting its spectrum depends on the nature of the substrate:

- If the substrate is reflective, you can analyze the sample in reflectance.
- If the substrate is opaque, scrape off a sample of the coating; use the roller knife to scrape a small piece on to a KBr or BaF_2 disk.

Liquids

Solutions of samples

Although liquids are seldom analyzed with the microscope, sometimes the sample of interest is in solution.

1. Transfer the solution on to a salt plate.
2. Allow the solvent to evaporate, leaving the sample on the plate.

Micropipettes

You can use a micropipette to apply liquid to the surface of a salt plate, or to the edge of the junction between two salt plates. In the latter case, the liquid flows between the plates by capillary action.

Preventing liquids from spreading

If the amount of liquid being transferred to the salt plate is very small, restrict it to a small area of the plate. There are several ways to do this:

- Use a microbrush to transfer solutions. The bristles of the microbrush hold the liquid in a small region of the salt plate until the solvent evaporates (see *Tools You Can Make Yourself* on page 69 for a procedure for making microbrushes).
- Repeatedly jab a small area of the salt plate with a tungsten probe. Leave the resulting small salt particles in the well that is produced.

  The capillary spaces between the salt particles retain the liquid and minimize spreading.
- Place the salt plate on a small metal washer that is being gently heated.

  Because there is more heat at the outside of the salt plate than near the center (over the hole in the washer), the droplet of liquid is forced toward the center.

Special Cells

For discussion of the diamond anvil cell, see *Polymers* on page 76.

Compression Cell

The compression cell (N187 0185, Figure 16) is a device for flattening soft materials and holding specimens flat and in optical contact with salt windows. The cell consists of an aluminum block, machined to accept salt windows, with window retainers and a special wrench to apply pressure across the windows. The sample is held between the two windows. The compression cell fits into the sample slide holder on the stage of the microscope. 1 mm and 2 mm thick windows of 13 mm outer diameter can be used with the cell; two KBr windows (2 mm thickness) are included. The cell can apply pressure without rotating the windows, and therefore avoids scratching them.

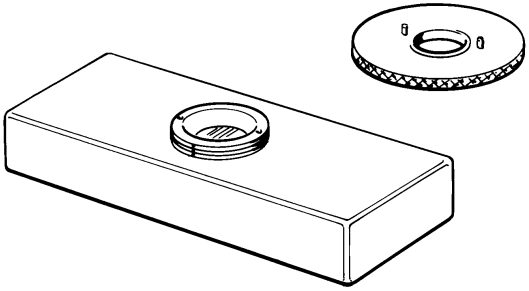

Figure 16 The Compression Cell

Although thinning can be accomplished with the compression cell, it does not replace the miniature diamond anvil cell as a sample-thinning device. The primary application of the compression cell is for keeping specimens flat over the entire visual field of view. The sample area to be isolated is then more accurately determined and apertured, and you do not have to refocus the microscope when viewing different parts of the sample.

Hot Stage

The hot stage for the MultiScope System Microscope (N187 0184, Figure 17) is used to study temperature-dependent phenomena in microsamples. The hot stage consists of a temperature controller and a heating block that accepts infrared windows. The heating block contains an integral thermocouple, and the temperature is digitally displayed in degrees Celsius on the controller. The hot stage can heat samples up to 250 °C in 1-degree increments. A target temperature can be selected and maintained. The hot stage is held in the slide clip on the sample stage of microscope. The microscope requires no modifications to accept the hot stage.

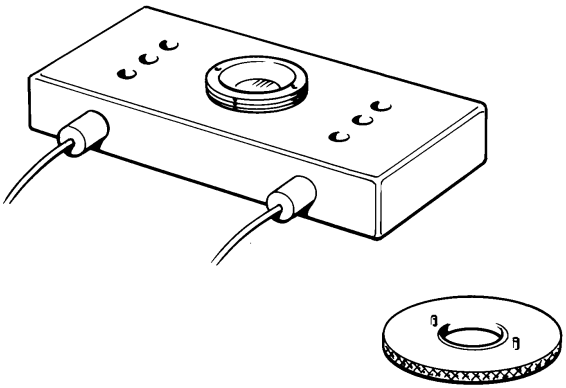

Figure 17 The Hot Stage

Applications of the hot stage include the study of phase transitions and temperature-dependent chemical reactions. Infrared microscopy can provide detailed molecular structural information for systems undergoing phase transitions; this information is not available from thermal data only. Polymers, pharmaceuticals and liquid crystals are examples of materials where investigations of phase transition are important.

Collecting Spectra with the Microscope

Collecting Spectra with the Microscope

Usually, the MultiScope System Microscope is used for collecting the spectra of microscopic samples. This chapter contains the necessary procedures for:

- Positioning the sample correctly on the sample stage;
- Collecting a background spectrum;
- Collecting the spectrum of the sample.

Also, there are procedures that can be used in special situations:

- When the spectrum of the sample must be collected before the background;
- When the sample is too thick to fit on the stage in the usual arrangement;

These procedures assume that the microscope and the FT-IR are ready to use (*Getting Ready to Use the Microscope* on page 45), and that the microscopic sample has been correctly prepared (*Tutorial: Using the Microscope* on page 53).

Positioning the Sample

When you use the microscope to collect spectra, you must center the sample on the sample stage before you collect the background spectrum. This enables you to set the correct aperture for the sample and then use that aperture when collecting the background spectrum.

1. Take out any aperture that is in the retaining well.
2. Place the sample on the sample stage.
   If necessary, secure it with the slide clip:

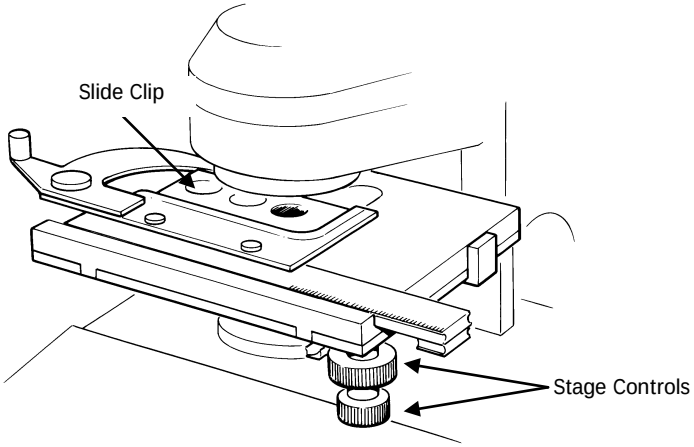

Figure 18 Parts of the Sample Stage

3. To set the microscope for viewing a sample, turn the View/IR knob to View. The detector mirror moves out of the optical path, and the illuminator is switched on.
4. Set the Trans/Refl knob to the required position for viewing.
5. Turn the Illuminator knob to adjust the light to an adequate level for viewing.

NOTE: If you need to increase the illumination during the procedures that follow, move the knob to a higher setting.

6. Looking from the front, adjust the stage in the x and y directions with the stage controls (under the stage on the right, Figure 18) until the sample is illuminated.

    - Move the sample in the x direction (left and right) with the lower, smaller knob.
    - Move the sample in the y direction (forward and back) with the upper, larger knob.

7. Use the Focus knob on the left of the microscope (Figure 19) to bring the sample into focus.

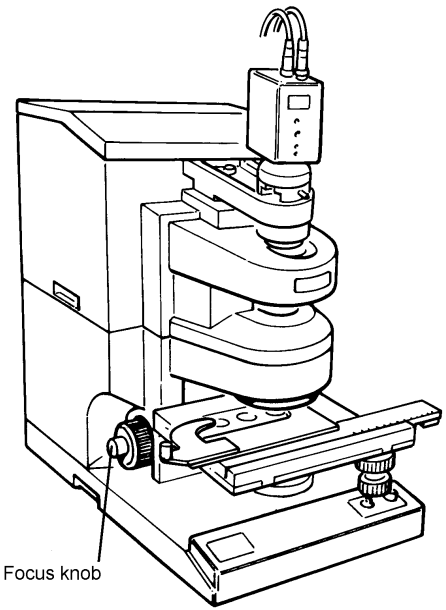

Figure 19 The Focus Knob

8. Look at the visible image as you use the stage control knobs to place the sample as required in the center of the field.
9. Adjust the Focus knob again until the sample is in focus.
10. Place the whole field aperture in the retaining well.

    All four knobs on the aperture must be in front of the retaining spring.

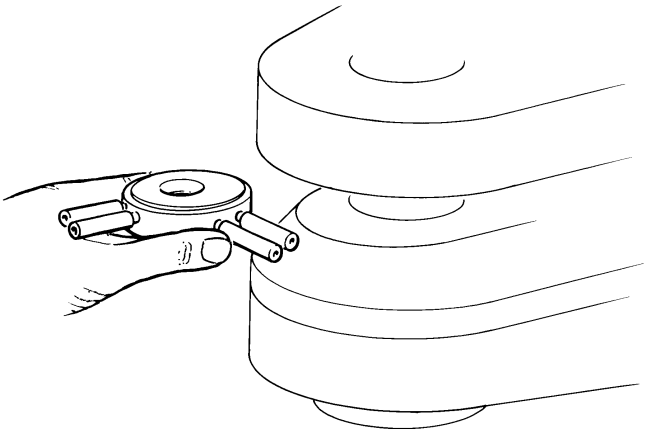

Figure 20 Inserting the Variable Knife-edge Aperture in the Retaining Well

11. Look at the visible image as you turn each of the four knobs of the knife-edge. Reduce the size of the aperture until it just covers the area of interest in the sample.

    The material of the aperture is transparent, so that you can see the surrounding area of the sample. This helps in positioning the sample correctly.

    Move the sample out of the field of view using either the x or y stage controls.

    You are now ready to collect a background spectrum for this sample.

Collecting a Background Spectrum

We recommend that you collect a new background spectrum for each sample, because the aperture size and FT-IR settings can vary from sample to sample.

Collect the background spectrum using the same conditions that you used to collect the sample spectrum. For example, if the sample rests on a KBr window or a reference mirror, include the window or mirror in the background.

NOTE: Sometimes it is inconvenient to collect the background spectrum first. If this is so, read the section *If You Must Collect the Sample Spectrum First* on page 92.

1. Using only one of the stage controls (x or y), move the sample out of the aperture.

   Two metal scales along the front and left sides of the stage give the x and y coordinates of the center. Each scale has a vernier with 100 µm resolution.

   If you note the location of the sample before you move it on the vernier scale, you can re-position it very near to the same location later.

2. To select the type of spectrum to be collected (transmittance or reflectance), set the Trans/Refl knob to Trans or to Refl as required.

Correcting the microscope for focal shifts

1. Display the energy bar produced by the **Monitor** command of the FT-IR.

   If you are collecting a reflectance spectrum, go to step 5. Otherwise, you may need to correct for focal shifts that occur as the infrared beam comes through the window on which the sample is mounted

2. Adjust the Correction knob on the right of the microscope base until the energy is at a maximum. Do not leave **Monitor** yet.

NOTE: If the reading on the energy bar is very low, make sure that there is liquid nitrogen in the dewar flask. (See *Cooling the MCT Detector* on page 48).

3. Adjust the gain to give maximum signal:

   - With the Spectrum 100 Series or Spectrum One, the number must be as near as possible to, but not more than, 6000.

   - With the Model 1600 or Spectrum RX, the number must be as near as possible to, but not more than, 100. If the signal does not meet this requirement, return to Ready For Next Command and use SETUP SCAN OTHERS GAIN to adjust the gain.

   - With the Model 16PC or Spectrum BX, the number must be as near as possible to, but not more than, 10000. If the signal does not meet this requirement, display the Setup menu and choose **Instrument**, then click **Gain** to increase the gain.

   - With the Spectrum GX, the number must be as near as possible to, but not more than, 6000. If the signal does not meet this requirement, display the Setup menu and choose **Instrument**, then click **Gain** to increase the gain.

4. Return to **Monitor** to be sure the number has not exceeded the figures given on above.

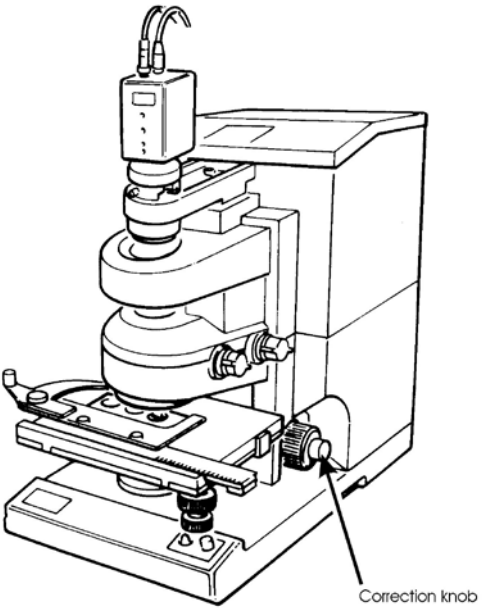

Figure 21 Location of the Correction Knob

5. When the gain is set, collect a background spectrum:

    - With the Spectrum 100 Series or Spectrum One, display the Instrument menu and choose **Scan**. Select the Scan tab and choose **Scan type: Background**.
    - With the Model 1600 or Spectrum RX, use **SCAN BACKG**.
    - With the Model 16PC or Spectrum BX, display the Instrument menu and choose **Scan Background**.
    - With Spectrum GX, display the Instrument menu and choose **Scan Background**.

Collecting the Spectrum of the Sample

Before you can collect the sample spectrum, you must once again center the sample in the field visually. The following instructions describe how to center the sample and collect its spectrum.

1. Return the sample to the center of the field. Do not change the size of the aperture.
2. Switch the View/IR knob to IR.
3. Use the SCAN command of the FT-IR to collect the spectrum of the sample.

If You Must Collect the Sample Spectrum First

To collect the background spectrum with the same aperture as the sample spectrum, you usually set the aperture with the sample in place, move the sample before you collect the background spectrum, and finally return the sample and collect its spectrum.

With some samples, it is inconvenient or even impossible to move the sample and then replace it precisely, for example, if the sample is an electrical contact on a circuit board. The following procedure enables you to collect the sample spectrum first, before you move the sample and collect the background spectrum. This example shows how to perform the procedure in reflectance mode.

1. With the sample under the microscope, set the aperture.
2. Collect a spectrum of the sample in single-beam mode.
3. Remove the sample without changing the aperture.
4. Put the reference mirror on the sample stage.
5. Still without changing the aperture, focus on the mirror.
6. Collect a single-beam spectrum of the mirror into the background region of the FT-IR.
7. Divide the single-beam sample spectrum by the background spectrum to obtain the ratioed spectrum, using the difference command in IRDM or GRAMS, the spectral calculator in Spectrum, or the diff command on the Spectrum BX or Model 1600.

Collecting the Spectrum of a Thick Sample

You can lower the stage in order to focus on a thick sample. For very thick samples, this requires the removal of the lower cassegrain assembly and then the reflectance method can be used to view the sample and collect its spectrum.

To collect the spectrum of a thick sample

1. Release the locking lever, at the back of the lower cassegrain assembly on the right (Figure 22).

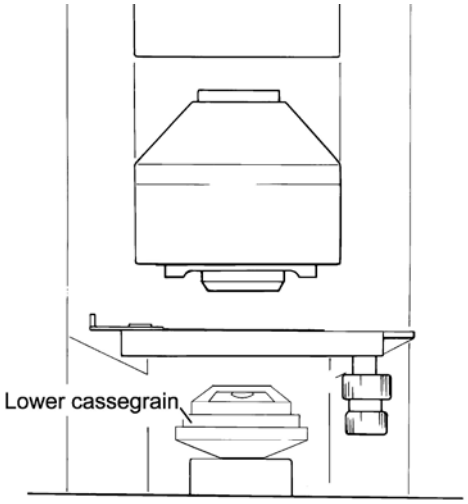

Figure 22 The Lower Cassegrain

2. Gently slide the cassegrain assembly forward and out of the dovetail connector.

NOTE: To make withdrawal easier, pull the locking lever gently.

3. Turn the Correction knob to lower the dovetail connector to the bottom of its range of travel.

4. Lower the stage using the Focus knob.

Continue as in *Positioning the Sample* on page 85 except that you must set the Trans/Refl knob to Refl.

To refit the lower cassegrain after use

1. Raise the stage using the Focus knob.
2. Turn the Correction knob to raise the dovetail connector.
3. Make sure that the cassegrain is correctly seated.
4. Slide the cassegrain assembly back into the dovetail connector, as far as it goes.

NOTE: To make withdrawal easier, pull the locking lever gently.

5. Tighten the locking lever.

Operating the Optional Equipment

Operating the Optional Equipment

This chapter contains the special instructions you need to operate any optional equipment that you purchased with the MultiScope System Microscope, for example:

- The Visible Polarizer option;
- The Infrared Polarizer option.

The following options for sample preparation are described in *Preparing Samples* starting on page 63.

- The hot stage;
- The compression cell;
- The microsampling tool kit;
- The miniature diamond anvil cell.

Visible Polarizer Option

This section describes how polarization of visible light can be useful in identifying areas or structures that differ chemically, and how this technique can be used in solving problems commonly found in infrared microspectroscopy applications.

Ordinary light and infrared radiation consists of waves vibrating in all possible planes perpendicular to the direction of propagation.

The Theory of Light Polarization

NOTE: Conventionally the plane of the light is taken to be the plane of the continuously varying electric vector.

This is represented in the left-hand side of Figure 23. If the light passes through a *polarizer*, the polarizer allows the passage of only those waves that have their plane of vibration in one particular direction. The light that emerges is said to be *polarized*, and is represented on the right-hand side of Figure 23.

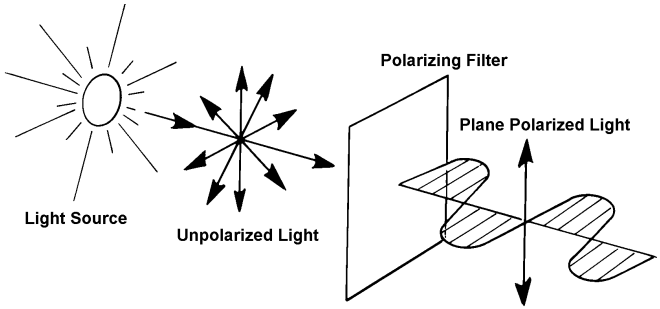

Figure 23 Representation of Unpolarized Light (left) and Polarized Light (right)

Because all components of the wave in the plane of polarization are transmitted, the ideal polarizer allows 50% of the light through.

If a second polarizer is placed in the path of the polarized light two things may result:

- If the second polarizer is placed in the same direction as the first (as at the top of Figure 24), the polarized light can pass straight through.
- However, if the second is placed at a right angle to the first, a situation which is referred to as *crossed polarizers*, the passage of the polarized light is blocked, that is, *extinction* occurs (as at the bottom of Figure 24).

  The second occurs because the light transmitted by the first polarizer oscillates in exactly the plane that is blocked by the second polarizer.

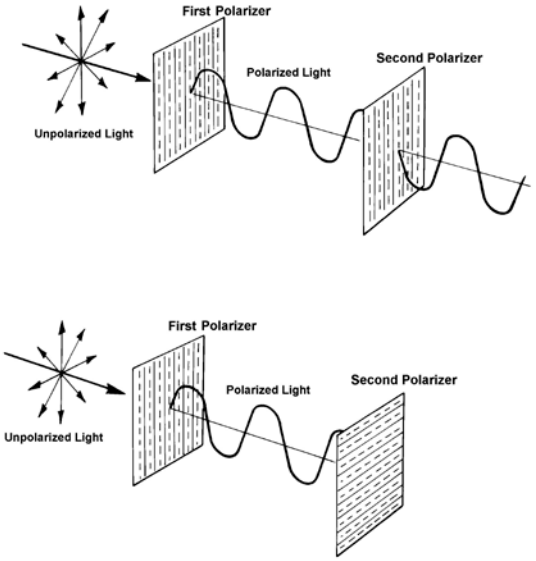

Figure 24 Polarizers Parallel (top) and Polarizers Crossed (bottom)

Some materials are *anisotropic* (or *birefringent*): their refractive index depends on their orientation. These materials can alter the polarization of light passing through them; this is dependent on the wavelength of the transmitted light.

When you look at an anisotropic sample using polarized light, the change in polarization caused by the sample means that some light leaks through the second polarizer. Because the change in polarization is dependent on the wavelength, the color of the light emerging changes with the distance traveled through the sample and the amount of birefringence encountered.

Applications

Differences in the birefringence of an object or area may be an indication of chemical disparity. This can be useful in visibly separating or identifying an object or area of interest before collecting an infrared spectrum. Some examples are given below:

Laminates

Many polymer structures consist of different layers of material and adhesives of varying thicknesses bonded together in order to meet physical requirements. By viewing a cross section of the structure using polarized light, the individual layers can be identified, apertured, and a separate infrared spectrum of each can be collected. This is useful for identifying the materials used to create the structure.

Polymer films

Although they may appear identical when using normal light, the film and inclusion present in a polymer film may exhibit different degrees of birefringence when using polarized light. If this is so, the inclusion can be visibly identified and an infrared spectrum can be collected to determine its composition.

Rocks minerals and crystals

Most crystals are characteristically birefringent and thus are ideally suited to this technique. Various components of a mixture consisting of crystals, such as an artificial sweetener, pharmaceutical powder, or an illicit substance can be viewed and then visibly separated by their relative size, shape and birefringence. The crystals that appear different can be apertured separately, and infrared spectra can be collected.

Fibers

Polarization can be used to identify and separate fibers in a clump or to view a bicomponent fiber. Most fibers in their natural state are optically thick and their cylindrical shape can cause lensing effects. For these reasons, fibers are typically flattened in preparation for infrared microspectroscopy. This flattening affects the birefringence of the structure and may degrade the usefulness of this technique.

Biological substances

Birefringence can occur in some biological substances. Infrared spectra of thin sections of these materials can be collected. In some cases, polarized light can reveal structures or chemical disparities in these structures, and infrared spectra can be collected of the different regions of interest.

Equipment

The equipment for visible polarization studies consists of two parts: the polarizer (Figure 25) and the analyzer (Figure 26).

The polarizer polarizes the incoming beam from the illuminator and the analyzer contains a polarizing element that can be rotated into any orientation.

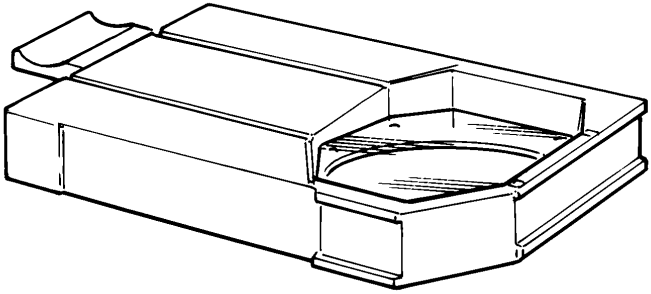

Figure 25 The Polarizer

Figure 26 The Analyzer

Operation

To use the analyzer and polarizer:

1. Insert the polarizer into the horizontal slot below the illuminator on the left of the microscope (Figure 27), with the angled corner facing towards the front of the microscope so that it pushes in the ball-bearing inside the slot. Push the polarizer fully in until it stops.

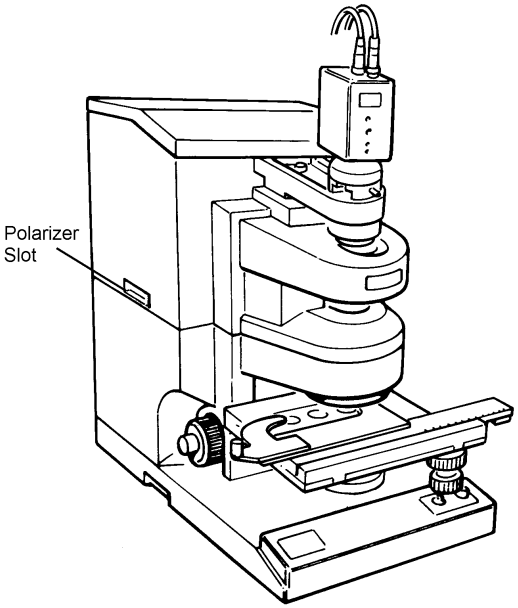

Figure 27 Polarizer Position

2. Insert the analyzer into the slot under the video camera (Figure 28).

   Push the analyzer in with the wheel up and facing towards you. It has two positions:

   - The first position allows the full beam to pass through.
   - When the analyzer is inserted fully, the polarizing element is in the beam.

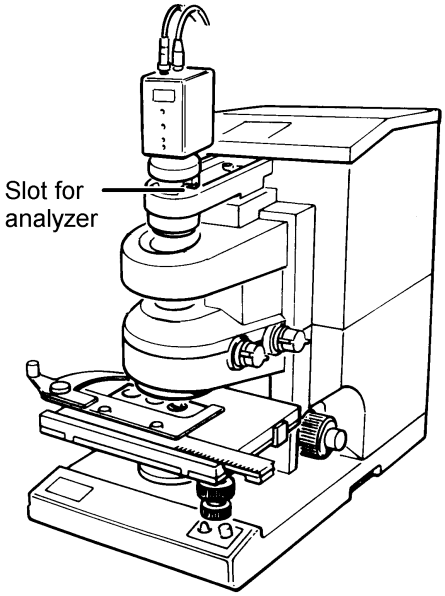

Figure 28 Analyzer Position

3. When the polarizing element is in the beam, rotate the wheel while observing the sample on the monitor.

Infrared Polarizer Option

An absorption band in the infrared range occurs when a vibration is accompanied by a change in dipole moment. The electric vector of the incident radiation must have a component in the direction of the dipole moment change.

In polarization spectroscopy, the absorption bands of greatest interest are those in which the direction of dipole moment change is related to a bond direction, for example, the nitrile stretching vibration. If, in a particular sample, all the bonds of a particular type are aligned in a specific direction, the strength of the absorption depends on the polarization of the incident radiation, that is, whether the electric vector is parallel to or perpendicular to the bond direction.

For example, stretching an acrylic fiber aligns the molecules with the general direction of the polymer chains parallel to the fiber axis, and the nitrile groups tend to be oriented perpendicular to the axis. If the spectrum is collected with the infrared radiation polarized perpendicular to the axis, the nitrile absorption peak is much stronger than if the spectrum is collected with radiation polarized parallel to the axis. The ratio of the two intensities (called the *dichroic ratio*) is a measure of the extent of alignment of the nitrile groups and thus of the polymer chains.

By the use of the microscope, polarization spectra can be collected on very small samples. These include:

- Single filaments (typically 14 x 70 µm);
- Films;
- Single crystals;
- Liquid crystal polymers.

Equipment

The polarizer has a rotatable silver bromide element in an aluminum mount (Figure 29)

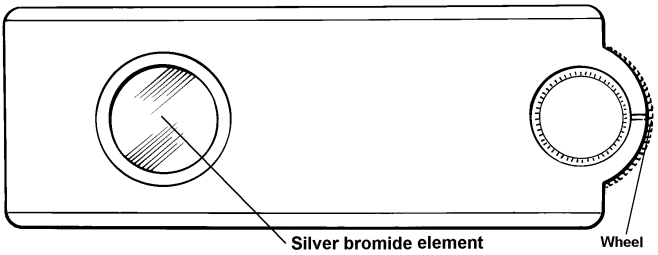

Figure 29 The Infrared Polarizer

Using the Polarizer

1. Remove the metal cover that masks the aperture for the Infrared polarizer. The metal cover is above the View/IR knob.
2. Slide the analyzer into the vertical slot on the sample holder that can be seen through this aperture.

NOTE: The flat side of the analyzer must be towards the rear of the microscope, and the wheel facing outwards.

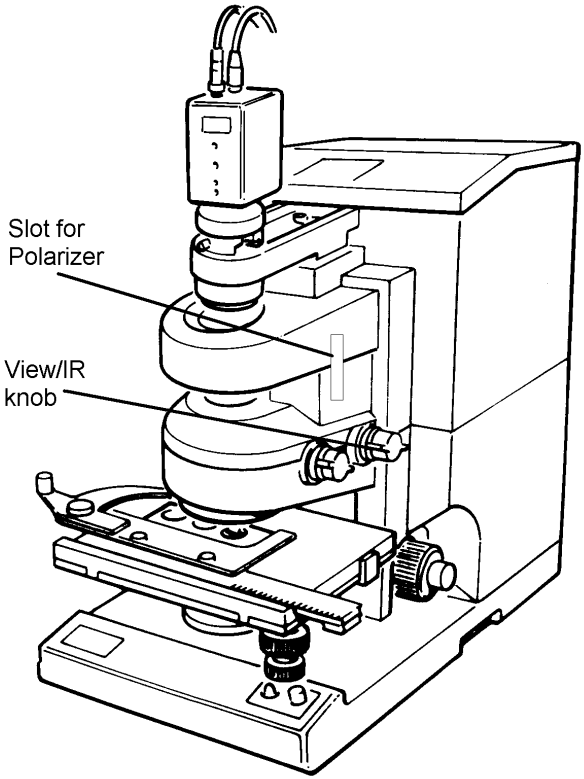

Figure 30 Infrared Polarizer Position

3. Push the analyzer fully until it stops.

4. Turn the wheel in order to orient the polarizing element.

WARNING

The polarizer element is extremely fragile. Do not touch it with anything. It cannot be washed, dusted, or blown upon by air. If damaged, it cannot be repaired. When it is not in use, protect it in the case supplied.

NOTE: Both the scribed line and the uneven coloration are normal, and do not affect the performance of the element.

Collecting a Spectrum in an Inert Atmosphere

The spaces around the sample and the infrared beam can be purged (typically with nitrogen or dry air) to provide an inert atmosphere.

Parts of the purging system

Some parts of the purging system are shown in Figure 31.

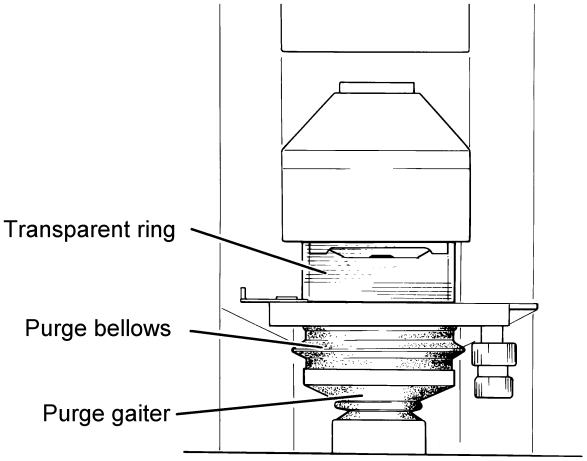

Figure 31 Parts of the Purge System

The purge system consists of:

- The gas inlet connector on the metal plate at the rear of the microscope;
- The transfer-optics housing connecting the FT-IR to the microscope;
- The lower purge gaiter under the lower cassegrain;
- The purge bellows under the sample stage, which enables you to move the stage up and down without breaking the seal;
- The metal ring (PTFE coated) fitting with an O-ring under the nose casing;
- The transparent ring that goes around the sample, and can be raised around the metal ring to enable you to access to the sample;
- The aperture in the retaining well that closes off the end of the purgeable volume.

When all of these parts are in place, the gas entering through the inlet displaces air from the path of the infrared beam and the sample area.

Purging the system

1. Make sure that all parts of the purge system, as listed above, are in place.
2. Set up the microscope.
3. Place the sample in position.
4. Purge the system for 15 to 20 minutes at a rate of approximately 10 L min^{-1}.
5. Collect the background spectrum and the spectrum of the sample.

Applications

Applications

This chapter contains examples of several applications for which the MultiScope System Microscope is particularly useful:

- Reflectance FT-IR microscopy;
- Polymers;
- Textiles;
- Criminalistics;
- Biological materials;
- Product contaminants.

Reflectance FT-IR Microspectroscopy

The MultiScope System Microscope can be used with samples that display any of the three types of reflectance: diffuse reflectance, specular reflectance, or reflection-absorption.

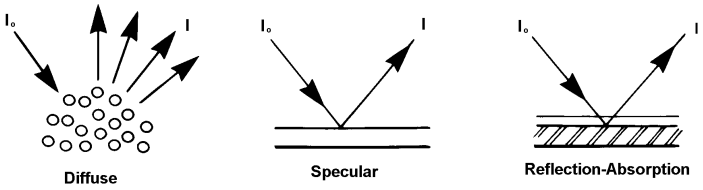

Figure 32 Three Types of Reflectance

Figure 32 shows how the incident radiation (I_0) is reflected in each type of reflectance. It is not uncommon for two or more of these processes to occur simultaneously, depending on the structure of the sample.

Diffuse Reflectance

In diffuse reflectance, the incident radiation is reflected in all directions from the surface of the sample. This type of reflectance is seen in samples with matt surfaces, such as paper. The broad collection angle of the microscope enables it to capture a large proportion of the diffusely reflected radiation and send it to the detector.

A problem often encountered when using the diffuse reflectance technique is that there is a large specular component in the reflected radiation. Figure 33 shows three diffuse reflectance spectra of polymethyl methacrylate (PMMA) shavings.

In the top spectrum, the shavings were neat. In the next two spectra, the PMMA was diluted with successively larger amounts of KBr.

In the neat sample, the presence of interfering specular reflectance introduces nonlinearities to the spectral data. For example, the relative intensities of the strong C=O and C-O stretching absorptions are not as expected (arrows on the top spectrum).

As the sample is diluted in a non-absorbing matrix (KBr), specular reflectance is minimized. The bottom spectrum in more closely matches an absorption spectrum.

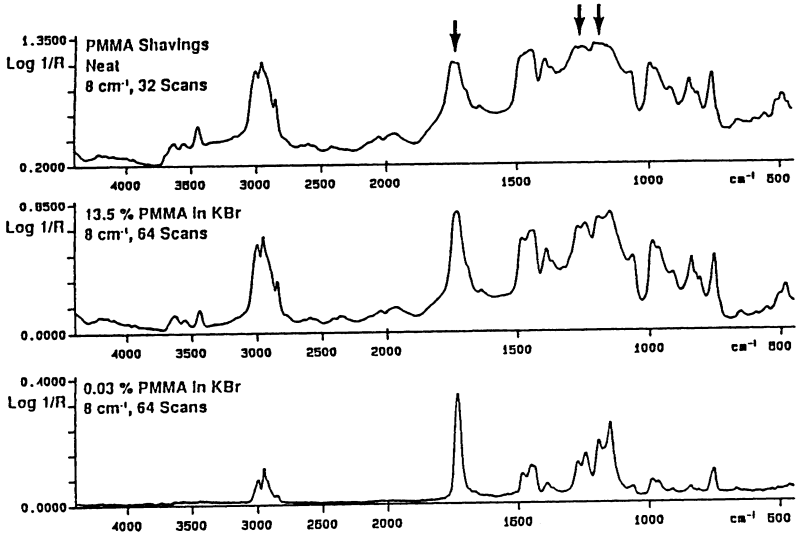

Figure 33 Diffuse Reflectance Spectra of PMMA collected with the MultiScope System Microscope and a Spectrum RX with MCT detector

Specular Reflectance

Specular reflectance is reflection in one direction (Figure 34). This is the type of reflection that occurs from a smooth, polished surface.

Absorption information cannot be obtained directly from a specular reflectance spectrum of a dielectric material, because the reflectance spectrum is governed by dispersion in the refractive index. However, you can use the Kramers-Kronig integration to calculate the absorbance spectrum from the specular reflectance spectrum. This integration is performed by the KK command in the Model 1600 and Spectrum BX software and the Spectrum software.

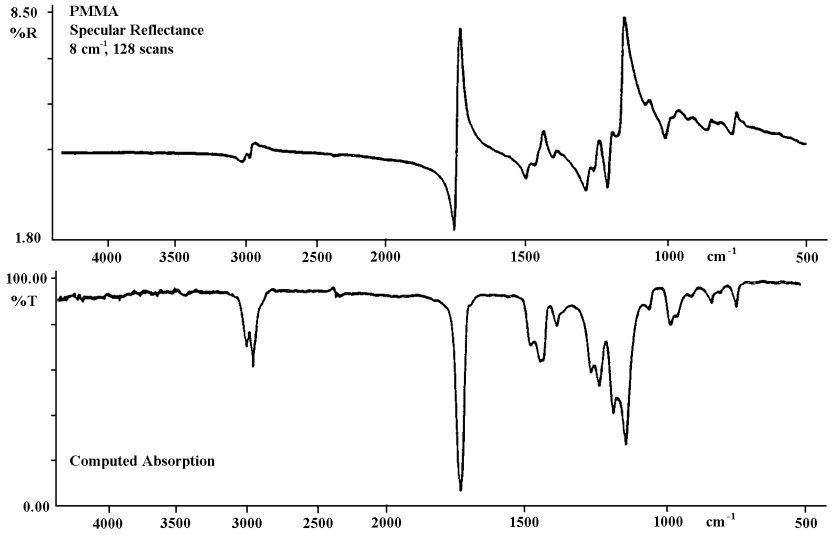

Figure 34 shows the specular reflectance spectrum of a PMMA plate and the absorption spectrum computed from it with the KK command. Collected with an MultiScope System Microscope and a Spectrum RX with MCT detector.

The greatest limitation to this method is the presence of an interfering diffuse reflection signal. This occurs when the sample surface is not perfectly smooth. The Kramers-Kronig integration is not appropriate for spectra from such samples.

Reflection-Absorption

Reflection-absorption occurs when the incident radiation passes through a thin, absorbent film that is on a reflective surface (typically a metal) and is then reflected back through the film (Figure 32). The absorbance spectrum of the film can be collected directly. Figure 35 shows the spectrum of residual oil on an electronic contact. The sample diameter was 100 µm.

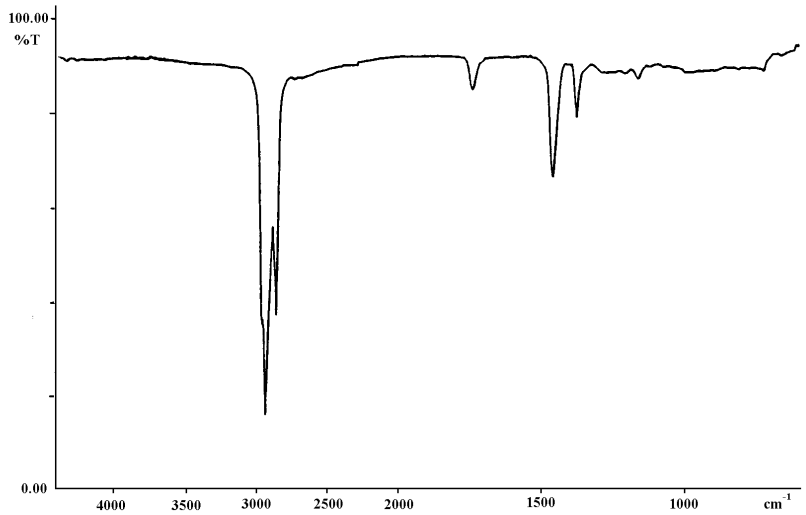

Figure 35 Reflection-Absorption Spectrum of Residual Oil on an Electronic Contact. Collected with an MultiScope System Microscope and a Spectrum RX with a medium-band MCT detector.

Polymers

In multicomponent polymeric structures such as laminates, or when one material is embedded in another, you do not need to separate the various components in order to identify them. If you obtain a microscopic cross-section, you can visually locate the component that is of interest, isolate it with the variable knife-edge aperture, and collect its spectrum. For example, Figure 36 shows spectra of the three layers of a laminated food packaging material.

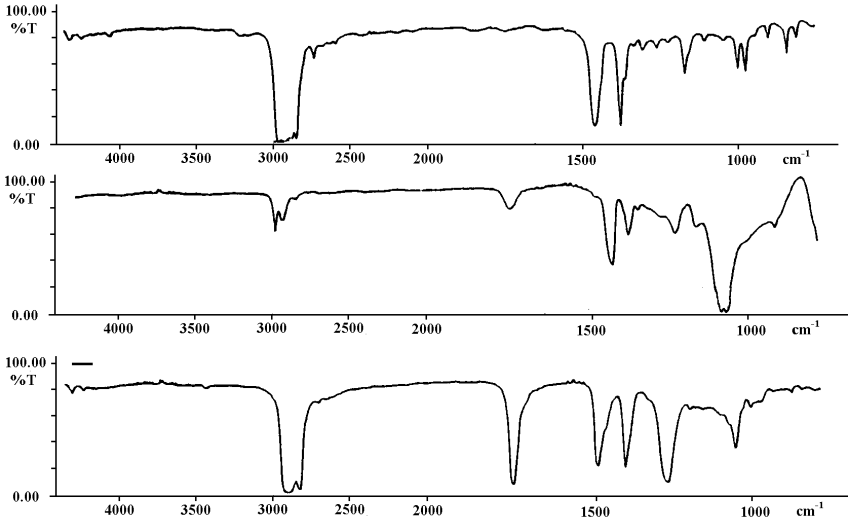

Figure 36 Cross Sectional Analysis of a Laminated Food Packaging Material.
Top: 200 µm layer of poly(propylene).
Middle: 15 µm layer of poly(vinylidine chloride).
Bottom: 10 µm layer of poly(ethylene vinyl acetate).
Collected with an FT-IR Microscope and a Spectrum RX with a narrow-band MCT detector.

Textiles

Textile scientists have used the MultiScope System Microscope to identify contaminant areas on fabrics or to study single fibers. Figure 37 is the transmittance spectrum of a flattened 20 µm poly(hexamethylene adipamide) fiber.

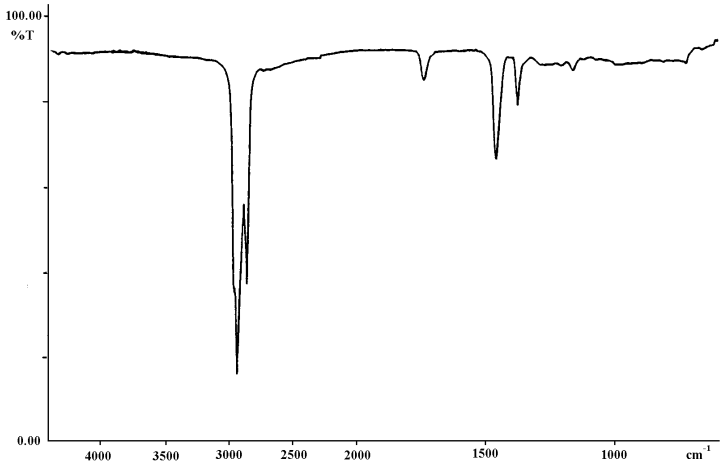

Figure 37 Transmittance Spectrum of a Flattened 20 µm Poly(Hexamethylene Adipamide) Fiber. Collected in a single scan with an MultiScope System Microscope and a Spectrum RX with a narrow-band MCT detector.

Criminalistics

The physical evidence to be analyzed in a criminal case is often microscopic; for example, fibers, paint chips, and traces of drugs or explosives. Figure 38 is the spectrum of a flattened paint chip from a green 1979 automobile. The spectrum indicates that the chip was a modified acrylic.

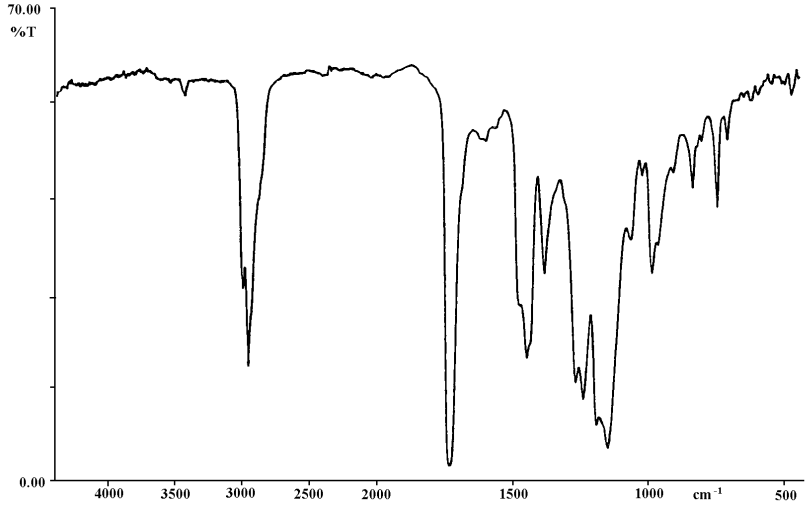

Figure 38 Transmittance Spectrum of a Flattened Paint Chip. Collected with an MultiScope System Microscope and a Spectrum RX with an MCT detector.

Biological Materials

Microscopic samples of plant and animal tissues can be analyzed spectroscopically. Figure 39 is a reflectance spectrum of an extracted human tooth, from which the absorbance spectrum was calculated with the Kramers-Kronig integration. The protein band at 1650 cm^{-1} is from blood; the apatite band at 1030 cm^{-1} is from the tooth enamel.

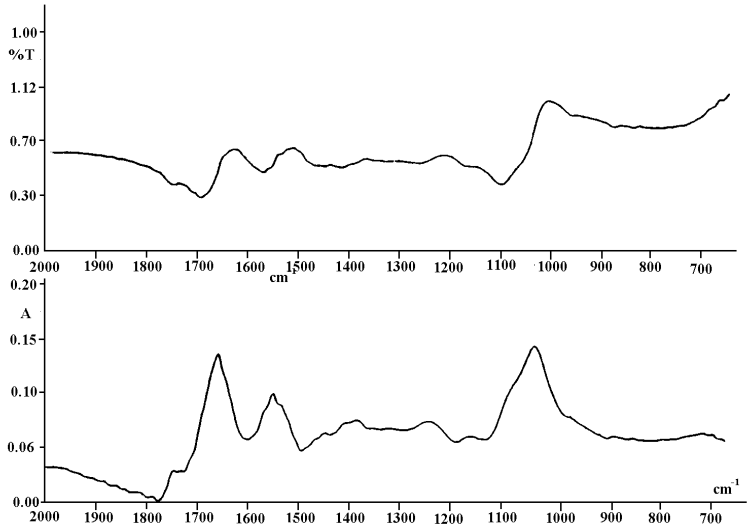

Figure 39 Top: Reflectance Spectrum of an Extracted Decayed Human Tooth.
Bottom: Absorbance Spectrum Calculated with the KramersKronig Integration.
Obtained with an MultiScope System Microscope and a Spectrum RX with a medium-band MCT detector.

Product Contaminants

When you have visual or other evidence that a contaminant is present that may affect the quality of your product, you can use the microscope to identify the contaminant spectroscopically.

For example, a manufacturer of a black paper product found it was contaminated with white spots. One of the spots was picked from the paper with a probe and placed under the microscope. The spectrum in Figure 40 was collected. The contaminant was identified as a mixture of glass (SiO_2) and wax (ester of a fatty acid).

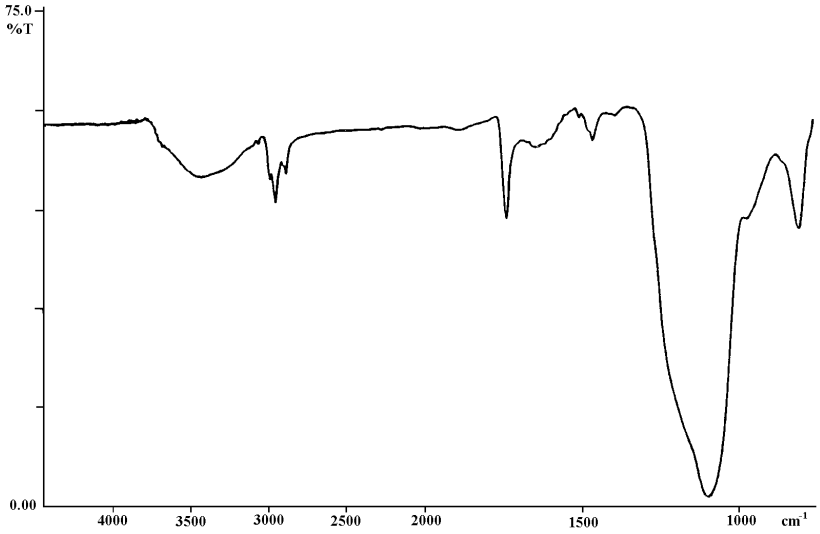

Figure 40 White Spot of Contaminant from the Surface of Black Paper. Collected with an MultiScope System Microscope and a Spectrum RX with a narrow-band MCT detector.

Figure 41 shows the spectra used to identify another paper contaminant. At the top is the spectrum of the contaminated paper. Below it, there is a spectrum of a cellulose fiber such as would be found in uncontaminated paper. The difference between the two spectra (bottom spectrum) corresponded to carbonate anion (CO_3^{2-}); the contaminant was probably either $MgCO_3$ or $CaCO_3$.

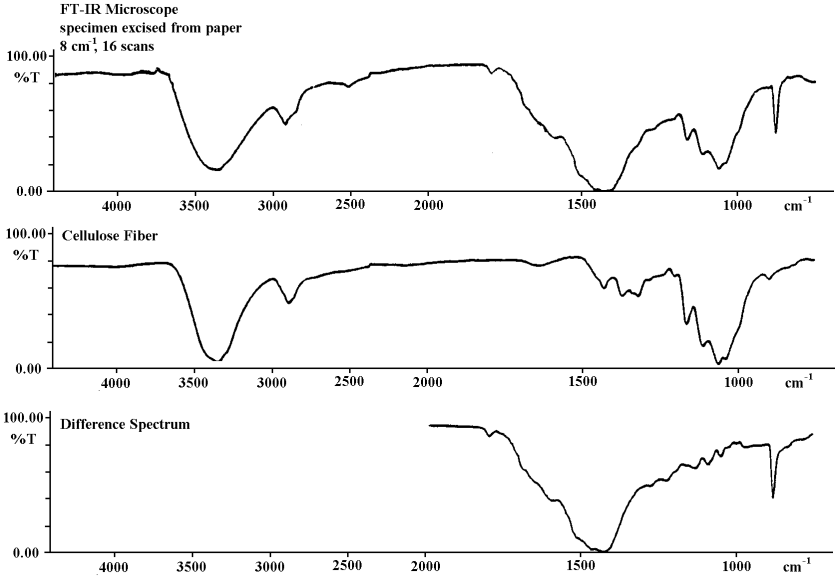

Figure 41 Identifying a Contaminant.
Top: Spectrum of Contaminated Paper.
Middle: Spectrum of Cellulose Fiber.
Bottom: Difference Spectrum Obtained by Subtracting the two Collected with an MultiScope System Microscope and a Spectrum RX with a narrow-band MCT detector.

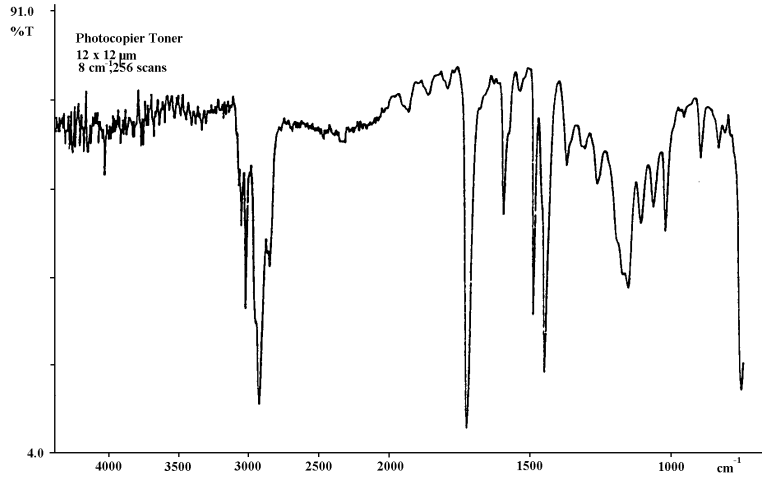

Figure 42 Photocopier Toner. Collected with an MultiScope System Microscope and a Spectrum RX with a narrow-band MCT detector.

Figure 42 shows the spectrum of a spot of photocopier toner, which is often encountered as an office or laboratory contaminant.

Maintenance

Maintenance

WARNING

Switch off the mains voltage and remove the mains cord before cleaning.

This chapter gives you the routine procedures you must follow to keep your microscope performing well:

- Inspecting the microscope;
- Protecting the microscope;
- Cleaning the optics;
- Aligning the MCT detector;
- Changing the lamp;
- Electrical connections;
- Service.

At the end of the chapter is a list of the available spare parts and optional equipment for the microscope.

If you need to replace a part, only use PerkinElmer approved spare parts.

NOTE: PerkinElmer FT-IRs contain diagnostic software. If you use the diagnostics with the microscope, be aware that it is impossible to fulfill the requirement to clear the beam. This is because the microscope is a throughput-limiting device. As a result, both the scan tests and the match adjustment generate errors that do not indicate true problems with the performance of the microscope.

Inspecting the Microscope

CAUTION	*The covers of the MultiScope must only be removed by a PerkinElmer Service Engineer.*

At least once a year, or whenever the microscope has been subjected to adverse environmental conditions, visually inspect the housing to verify that no covers are loose or distorted so as to allow access to the laser radiation in the interior.

At the same time, verify that all required labels are firmly in place (see Figure 1 on page 24 and Figure 2 on page 25).

Protecting the Microscope

The most important rule in caring for the microscope is to keep it as free from dust and dirt as possible. Dust, fingerprints, and smears on the optics reduce the quality of the images it produces.

Whenever the microscope is not in use, cover it with a plastic cover.

CAUTION	*Ensure the power is switched off when the Microscope is covered.*

Cleaning the Optics

The microscope was aligned, cleaned, and sealed at the factory. Do not attempt to take it apart.

If dust settles on the cassegrain mirrors and other optics of the microscope remove the dust using a stream of dry, clean nitrogen gas.

CAUTION	*Avoid the excessive use of solvents. Flowing solvents dissolve the cement on cemented optics; dissolved cement can damage mirror surfaces.*

Cleaning the Microscope Cover

CAUTION	*Ensure the power is switched off and the supply lead is disconnected before cleaning the cover.*

You can clean the outside of the microscope using a damp cloth. Mild detergent may be used, if necessary. Always perform a patch test on an inconspicuous area of the microscope, before you clean the entire instrument.

Avoid spilling liquid into the microscope. Clean all external spills immediately. If anything that is spilled enters the main body of the microscope, switch off the power and call a PerkinElmer Service Engineer.

Selecting the Microscope Voltage and Renewing the Fuse

The microscope is dual-voltage and can be supplied with 100 V to 120 V or 200 V to 240 V; make sure that it is set to the correct voltage. The voltage selector is on the rear panel of the microscope, under the mains inlet and power switch. If the arrow does not point to the required voltage, change it as follows:

1. Switch off the microscope and disconnect it from the power supply.
2. Insert a screwdriver into the slot at the side of the fuse drawer, and remove the fuse drawer from the rear panel.
3. Pull back the central black prong on the side of the fuse drawer, and remove the fuse tray.

   The fuse tray contains two fuses, one for use with a 100 V to 120 V power supply, and one for use with a 200 V to 240 V power supply.
4. Fit the replacement fuse into the fuse tray.

   Make sure that you fit the fuse in the correct slot. You require a 2.0 A time-delay, 250 V fuse (0C97 3084).
5. Refit the fuse tray in the fuse drawer.
6. Refit the fuse drawer so that the arrow points to the voltage you require.

Aligning the MCT Detector

The detector must be aligned when the microscope is first installed in your system. After that, it needs realignment only when it has been removed from the system and renewed, or when you suspect that it is not receiving the maximum signal. You will need the cross-head screwdriver that was supplied with the microscope.

WARNING

Laser radiation will become accessible if the MCT detector is removed.
If the MCT detector is to be removed from a Spectrum BX or Model 1600, first block off the laser radiation by selecting the FT-IR's internal beam path.

1. Switch off the microscope and disconnect the mains supply.
2. Remove the two screws that secure the microscope top cover; one is on the right at the rear, and the other at the front underneath the MCT detector cover. Lift off the cover vertically.

   The red MCT dewar and the metal frame and springs that support it become visible (Figure 43).

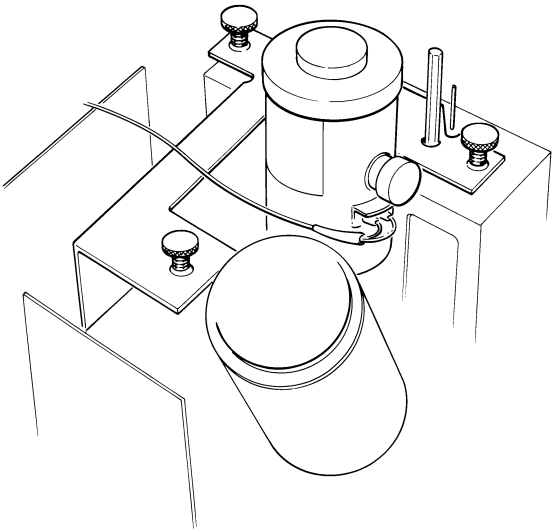

Figure 43 Interior of the Detector Area of the MultiScope System Microscope

3. If the dewar is not already filled with liquid nitrogen, fill it as described Cooling the MCT detector on page 48.

4. Set the microscope for infrared transmittance: switch the View/IR knob to IR and the Trans/Refl knob to Trans.

NOTE: If a Model 1600 or Spectrum BX is being used, select the external beam.

5. Use the stage controls to make sure that no part of the stage is obstructing the path of the infrared beam.

6. Place the variable aperture in the retaining well and set an aperture size of approximately 100 x 100 μm.

7. Use the Monitor command to display an energy bar on the FT-IR screen.

8. Turn the Correction knob at the base of the microscope until the energy bar is at a maximum.

9. The MCT dewar is supported by three screws in the top plate of its frame (Figure 43). Make sure that the screws in the front two corners rest in the depressions provided for them.

10. Maximize the length of the energy bar by turning each of the three alignment screws on the top plate of the MCT frame. Keep adjusting each screw in turn until the energy bar is as long as possible.

    The MCT detector is now aligned with the infrared beam.

Changing the Lamp

Switch off the microscope and remove the mains cord from the microscope.

WARNING

The areas surrounding the lamp and some components of the lamp power supply board may still be hot; allow a few minutes for these to cool before proceeding.

1. Remove the two screws holding on the top cover.
   One screw is under the dewar cover, the other at the back on a flange.
2. Lift off the top cover vertically.
3. Loosen the two screws holding the lamp leads at positions 4 and 5 of terminal block PL1, remove the connectors.
   The terminal block PL1 is at the top of the power supply board at the back of the microscope, next to the mains inlet unit.
4. Remove the two screws holding the lamp baffle, which surrounds the lamp, and lift it off the lamp.
5. Loosen the screw holding the lamp retaining bracket to the illuminator tube (fixed cylinder) and lift off the lamp retaining bracket.
6. Remove the old lamp.
7. Position the new lamp in the recess in the top of the illuminator tube.
8. Refit the lamp retaining bracket and align the top of the lamp to fit into the cut-out in the bracket.
9. Gently press the lamp retaining bracket down onto the lamp and tighten the lamp retaining bracket screw.

10. Pass the lamp leads through the opening in the top of the lamp baffle.

WARNING

Make sure that the screws connecting the lamp leads are tight. Failure to do this may result in overheating and damage to the microscope.

11. Refit the lamp baffle over the lamp, refit the screws.
12. Re-connect the lamp leads to terminals 4 and 5 of terminal block PL 1.
13. Refit the top cover and retaining screws.

The microscope is now ready for use.

Electrical Connections

Fitting the Plug

The power cable for the electrical supply plugs into the back of the microscope. It has a molded connector at one end. If it is necessary to fit a plug on the power cable, use the wire color code below:

Plug Pin	Wire Color (100/110/120 V)	Wire Color (220/230/240 V)
Ground (Earth)	Green or Green/Yellow	Green/Yellow
Line	Black	Brown
Neutral	White	Blue

WARNING

To ensure safe and satisfactory operation of the microscope, it is essential that the green or green/yellow ground (earth) wire of the power cord is connected to a ground that complies with the regulations of the local electricity supply authority (or equivalent body); ground circuit continuity is essential for safe operation of the equipment.

Connecting the MultiScope System Microscope to the Electrical Supply

The microscope operates on an electrical supply with a frequency of 50 or 60 Hz and at voltages in the ranges 100 to 120 V or 220 to 240 V.

Fit the molded connector of the power cable into the inlet at the rear of the microscope.

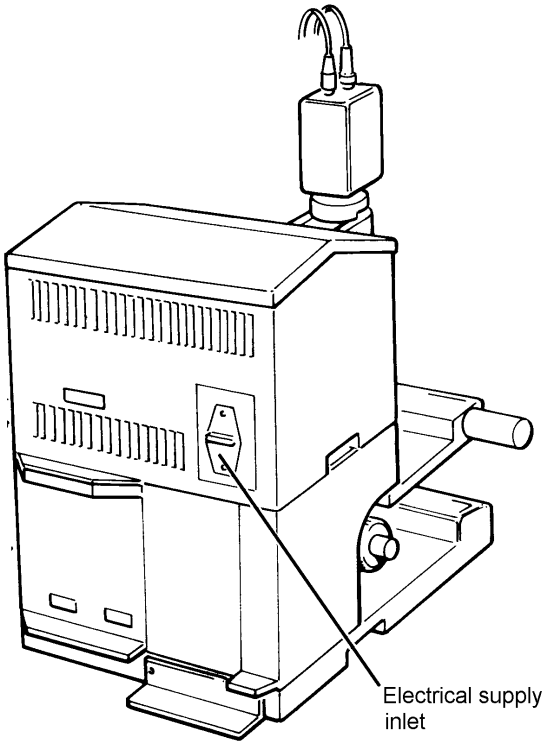

Figure 44 Location of the Electrical Supply inlet

MultiScope System Microscope Connected to a Spectrum One or Spectrum 100 Series Spectrometer

The cable from the microscope to the spectrometer comes from the preamp inside the microscope, to a connector marked on the rear of the FT-IR.

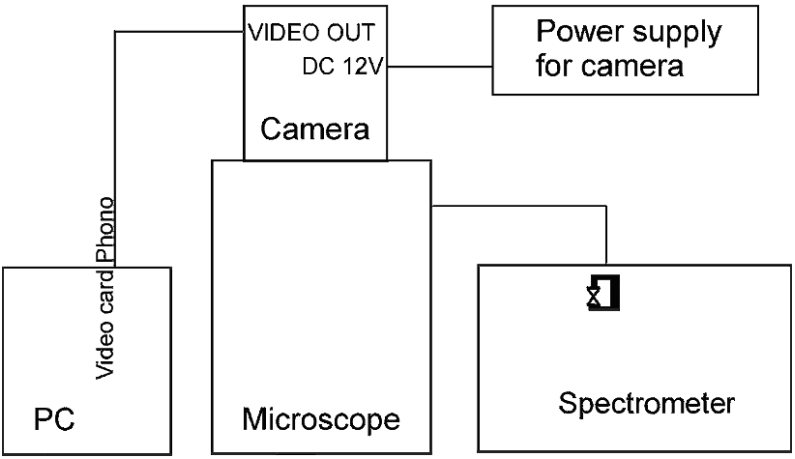

Figure 45 MultiScope System Microscope Connected to the Spectrometer

The following table shows all of the connections from the pre-amp in the microscope to the spectrometer:

PI 1 (PCB)	D-type Connector	Line	Power Requirement
1	11	Output signal	
2	10	Output signal (0 V)	
3	9	0 V	
4	12	0 V	
5	13	+12 V	250 mA
6	14	-12 V	200 mA

Connecting the MultiScope System Microscope to a Spectrum BX

The cable from the microscope to the Spectrum BX comes from the preamp inside the microscope, through a slot at the rear of the Spectrum BX to a connector inside the FT-IR (at the rear left-hand side).

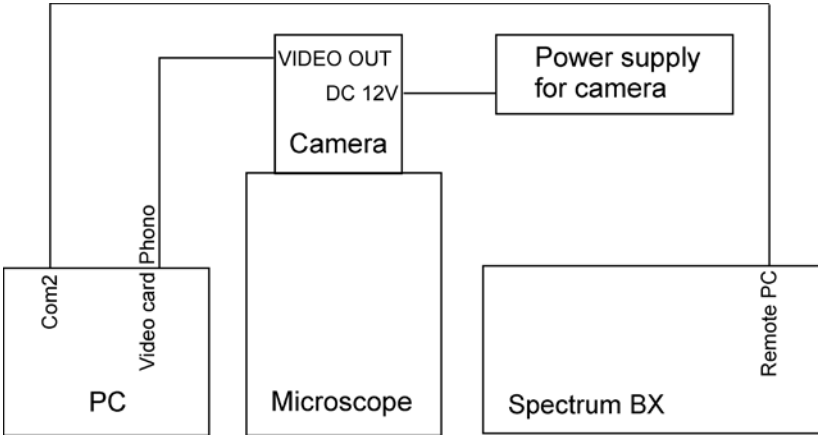

Figure 46 The MultiScope System Microscope Connected to a Spectrum BX

The following table shows all of the connections from the pre-amp in the microscope to a Spectrum BX (Paragon):

Pl 1 (PCB)	**CONN-IDC SKT 15 way**	**Line**	**Power Requirement**
1	11	Output signal	
2	10	Output signal (0 V)	
3	9	0 V	
4	12	0 V	
5	13	+12 V	250 mA
6	14	-12 V	200 mA
8	12	A1, gain selection	+5 V
9	11	A0, gain selection	+5 V

Connecting the MultiScope System Microscope to a Spectrum GX

The cable from the microscope to the Spectrum GX comes from the preamp inside the microscope, to an unlabelled 25-way connector on the lower right-hand side of the FT-IR.

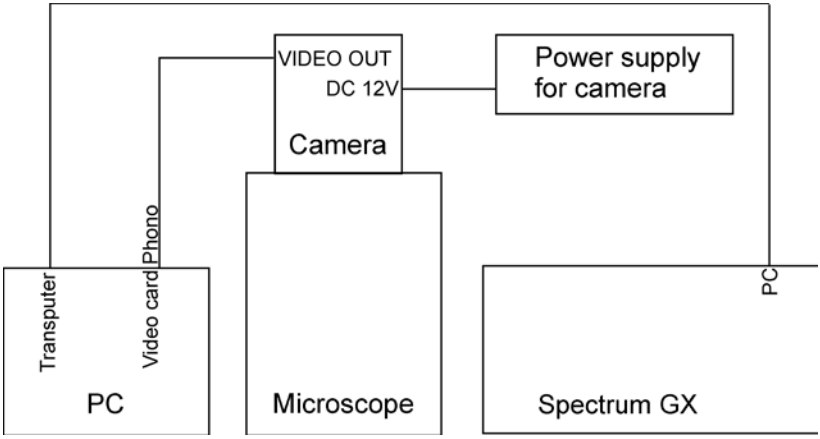

Figure 47 MultiScope System Microscope Connected to a Spectrum GX

The following table shows all of the connections from the pre-amp in the microscope to a Spectrum GX:

J1 1 (PCB), 25-way D-type	25-way D-type	Line	Power Requirement
1	1	+15 V	150 mA
2	2	+15 V	
3	3	0	
4	4	S2	
5	5	S4	
6	6	0	
7	7	-15 V	150 mA
8	8	-15 V	
9	9	OCBRES	
10	10	M0	
11	11	M1	
12	12	SO	
13	13	Output signal	
14	14	SK	
15	15	0vl	
16	16	0vl	
17	17	+5 V	150 mA
18	18	+5 V	
21	21	OSX	

Service

All optical and mechanical equipment requires periodic servicing to keep it performing properly and to compensate for wear. We recommend that the microscope is cleaned, examined, and adjusted periodically by a PerkinElmer Service Engineer.

NOTE: If you experience unexpected problems with the microscope, contact your PerkinElmer office or representative immediately.

Care for your video camera as described in the manufacturer's manuals.

Appendix: WEEE Instructions for PerkinElmer Products

or

A label with a crossed-out wheeled bin symbol and a rectangular bar indicates that the product is covered by the Waste Electrical and Electronic Equipment (WEEE) Directive and is not to be disposed of as unsorted municipal waste. Any products marked with this symbol must be collected separately, and in accordance with the regulatory guidelines in your area.

The objectives of this program are to preserve, protect and improve the quality of the environment, protect human health, and utilize natural resources prudently and rationally. Specific treatment of WEEE is indispensable in order to avoid the dispersion of pollutants into the recycled material or waste stream. Such treatment is the most effective means of protecting the customer's environment.

The requirements for waste collection, reuse, recycling, and recovery programs are set by the regulatory authority in your location. Contact your local responsible person (such as your laboratory manager) or authorized representative for information regarding applicable disposal regulations.

See the PerkinElmer web address below for information specific to PerkinElmer products, and contact details for the Customer care department in your region.

http://las.perkinelmer.com/OneSource/Environmental-directives.htm

Products from other manufacturers may also form a part of your PerkinElmer system. These other manufacturers are directly responsible for the collection and processing of their own waste products under the terms of the WEEE Directive. Please contact these manufacturers directly before discarding any of their products. Consult the PerkinElmer web site (above) for manufacturer's names and web addresses.

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