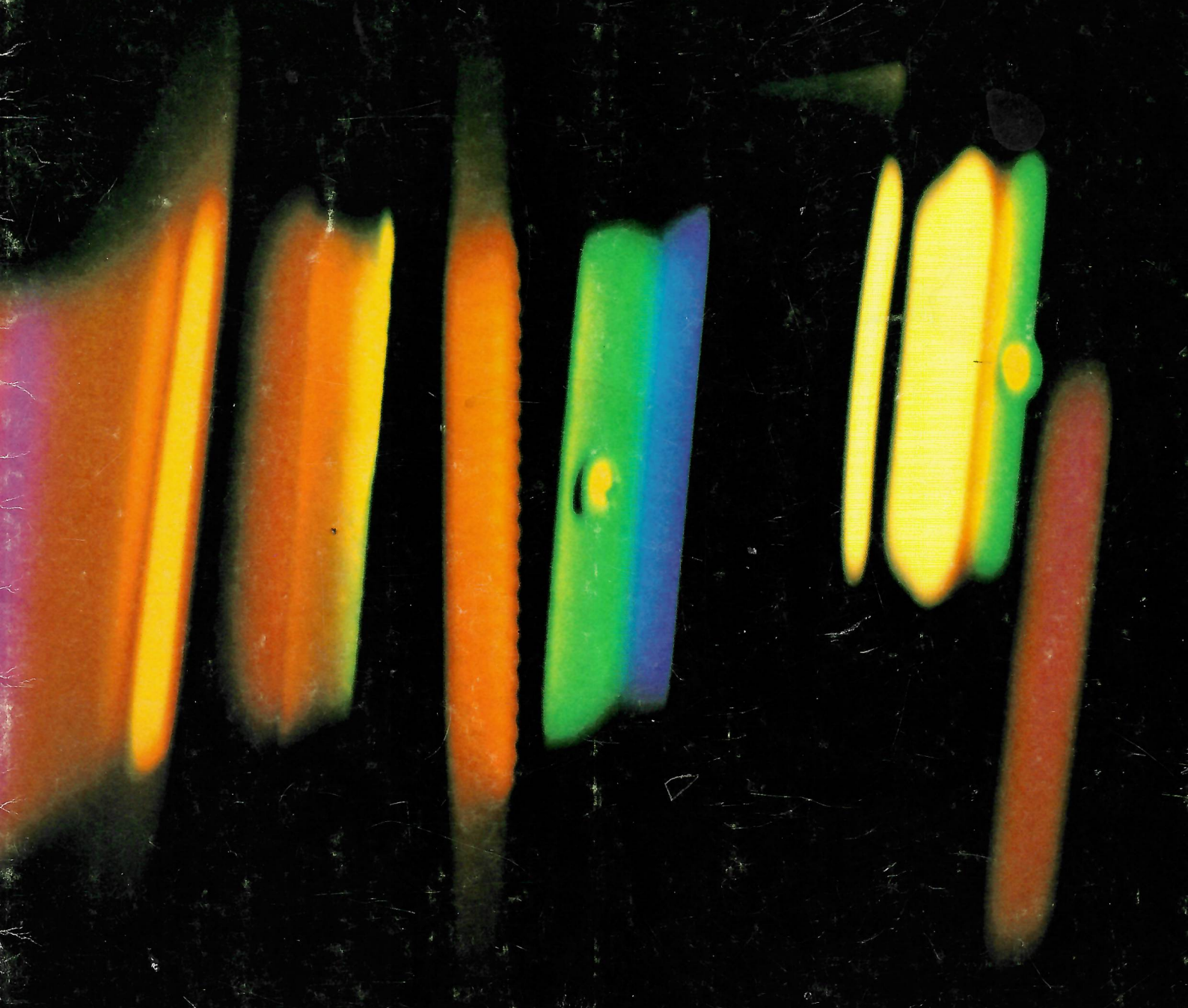


# DW-2a™ uv-vis SPECTROPHOTOMETER

Expanding the spectrum of research



AMERICAN INSTRUMENT COMPANY

DIVISION OF TRAVENOL LABORATORIES, INC.

Silver Spring, Maryland 20910 • Phone: 301-589-1727



# A very special instrument

Why? Because it has the performance capability equivalent to five separate, single mode spectrophotometers.

American Instrument Company, with its history of expertise in crafting sophisticated instruments, combined this five-instruments-in-one flexibility into a modular, versatile spectrophotometer that expands your spectrum of research.

With the large number and variety of accessories available, the DW-2a UV-VIS Spectrophotometer can fulfill the instrument requirements of your applications today and the change in direction your research studies may take in the future.

## Five Instruments in One

The five-instruments-in-one capability of the DW-2a UV-VIS Spectrophotometer provides five distinct modes of operation: Double-Beam – Dual-Wavelength – Dual-Wavelength Scanning – Rapid Kinetics and Optical Derivative. With this versatility, a variety of samples can be analyzed; clear, turbid or opaque – whether liquid or solid.

- 1. Double-Beam Mode** – is used to measure the concentration of unknown samples or small differences in concentration between a sample and a reference. It can also measure or detect differences between a sample and a reference of an identical substance after the sample is altered chemically or physically. The Double-Beam Mode uses a single, monochromatic light beam which is chopped into two beams of identical wavelength by a rotating chopper mirror. One beam passes through the sample, the other through the reference. This provides a continuous comparison between the sample and the reference while scanning with respect to time or wavelength.
- 2. Dual-Wavelength Mode** – is used to measure reaction rates or end product concentration changes in kinetic studies of complex systems. It is also used to measure concentration differences of single components in mixtures. In this mode, two different, preselected wavelengths are alternately passed through a sample in rapid succession. In the Dual-Wavelength Mode, the sample absorbance at two different wavelengths is measured and compared. This method eliminates non-specific interfering background absorption in turbid solutions caused by light scattering from particle sedimentation or size changes. It also obviates the need for a reference or standard.
- 3. Dual-Wavelength Scanning Mode** – is used to obtain spectral information from samples with changing light scattering properties such as suspensions whose particles settle or change size. Light beams of different wavelengths are alternately passed in rapid succession through a single sample. A stationary beam provides a reference, while a second beam scans a preselected wavelength interval. This eliminates light scattering errors when using turbid solutions and the need for a reference or standard.
- 4. Rapid Kinetics Mode** – is used to determine rapid kinetic reaction rates. Two light beams of different, preselected wavelengths are alternately passed through a mixing chamber in quick succession. The extremely fast electronic response of the instrument and the high speed of the rotating chopper mirror permit the determination of reaction rates in the millisecond range. The AMINCO-MORROW® Stopped-Flow Accessory is used in this procedure. The data produced by the kinetic reaction can be processed with the DASAR® System or displayed on a storage oscilloscope.
- 5. Optical Derivative Mode** – provides qualitative and quantitative

spectral information from absorption peaks which are obscured and appear merely as shoulders. These spectra are easily obtained by passing two monochromatic light beams of slightly different wavelengths through a single sample. The sample is then scanned over a selected spectral range. The use of the Dual-Wavelength Mode to obtain derivative spectra optically, eliminates the need for a reference or standard and provides  $dA/d\lambda$  spectra independent of scanning speed.

## New Measures of Precision

The DW-2a UV-VIS Spectrophotometer provides the precision in performance you expect from an instrument of its caliber. It has the highest signal-to-noise ratio of any comparable spectrophotometer. This helps to detect trace substances in nearly opaque samples, even at 0.005A full scale sensitivity. Other features that contribute to precision in performance are:

- Easy-to-read digital wavelength and slit readouts
- Bilateral curved slits for better resolution
- Excellent baseline stability
- Wavelength stability for accuracy and reproducibility
- Variable optical light beam attenuators to balance intensity of sample and reference light beams

## Many "Extras" Are Standard

The DW-2a UV-VIS Spectrophotometer comes as a complete system ready to use. Many of the features normally offered as options at additional cost are standard. To list just a few:

- X-Y-Time-base recorder for recording absorbance or transmittance as a function of wavelength or time
- Automatic repetitive scanning capability which allows you to repeatedly scan any portion of the X-axis
- Optical beam scrambler which minimizes magnetic interference and improves baseline flatness
- Derivative mode of operation without an accessory
- Calibration filter for checking wavelength and photometric accuracy
- Test curves run on your instrument
- 4 UV quartz cuvettes suitable for fluorescence studies

## Capability Expanding Accessories

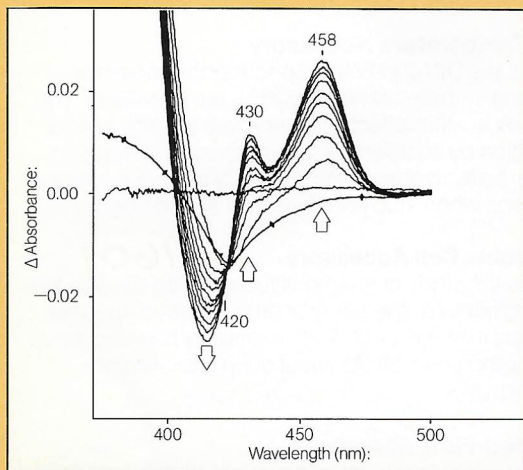
Because it has the capability of five individual, single-mode instruments, the DW-2a UV-VIS Spectrophotometer is used in a large number of research applications. Application needs change. Therefore, American Instrument Company offers an extensive number of capability expanding accessories. However, some research applications are so specific they may need a custom-built accessory. The DW-2a UV-VIS Spectrophotometer was designed with this in mind. The large, optical baseplate has ample space and mounting facilities for precise, easy installation of your special accessories.

American Instrument Company accessories greatly expand the versatility of this unique instrument. They provide the means to accurately process experimental data—to work with heavy suspensions and solid samples—to study reactions that require an anaerobic environment—to study complex plant systems—to study biological samples at cryogenic temperatures—to perform fluorescence analyses—to measure oxygen concentration changes—to study fast kinetic reactions and many other applications. The DW-2a UV-VIS Spectrophotometer—meeting your needs TODAY and TOMORROW!

# Providing special results

The sophistication and versatility of the DW-2a UV-VIS Spectrophotometer make it extremely valuable in any area of research. The following examples demonstrate the versatility of the instrument in the study of •carcinogens• toxic substances• plant physiology• human development• pollution control• pharmacology and metabolic processes. Over 500 applications for which American Instrument Company Spectrophotometers were used are listed in our bibliography of "Selected

Applications for the DW-2/DW-2a UV-VIS Spectrophotometer." Ask your American Instrument Company Representative for a copy or contact our Applications Laboratory at our headquarters. The bibliography will quickly demonstrate the wide variety of applications for this unique spectrophotometric system. The bibliography is also an invaluable reference tool in determining how the versatility of this instrument can be best applied to your particular needs.

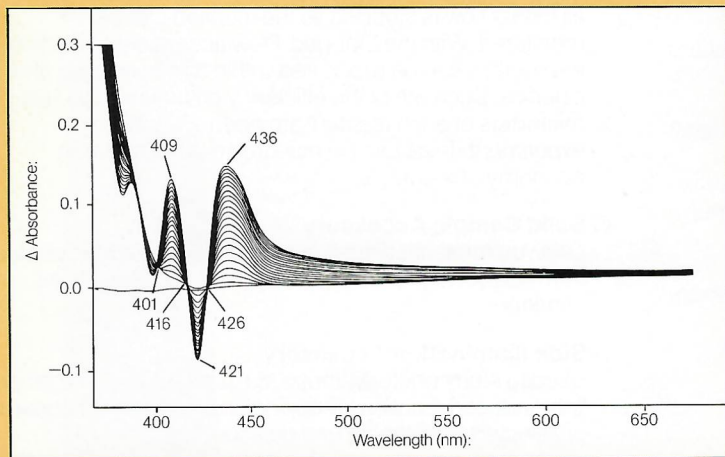


## Health Research

### Cancer Research

The oxidative metabolism of benzo ( $\alpha$ ) pyrene by 3-methylcholanthracene induced liver microsomes.  
Courtesy of R.W. Estabrook

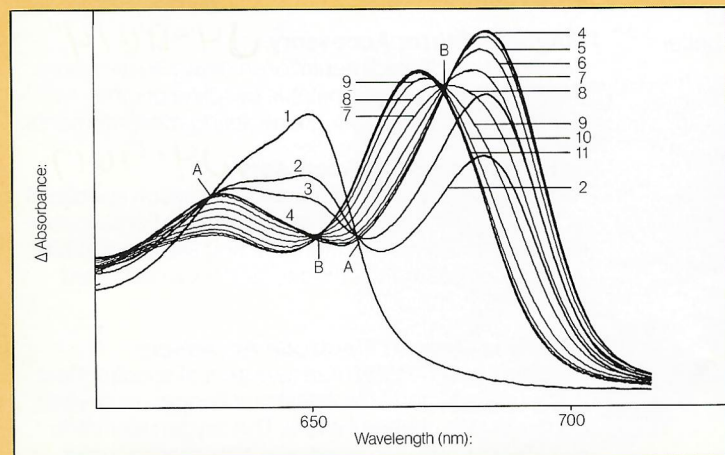
**Mode: Double-Beam, Repetitive Scan**



### Metabolic Processes

The oxidative metabolism of benzo ( $\alpha$ ) pyrene by phenobarbital induced liver microsomes.  
Courtesy of R.W. Estabrook

**Mode: Double-Beam, Repetitive Scan**



## Agriculture, Food

### Plant Physiology

1. Leaf before illumination: protochlorophyllide only.
2. After 1 flash: increasing amount of chlorophyllide a.
3. After 2 flashes: decreasing amount of protochlorophyllide.
4. After several flashes in quick succession: maximum conversion.
- 5.
- ↓ Shibata shift
- 11.
- A. Isosbestic points for conversion of protochlorophyllide to chlorophyll.
- B. Isosbestic points for the Shibata shift.

Courtesy of N.O. Björn

**Mode: Double-Beam**

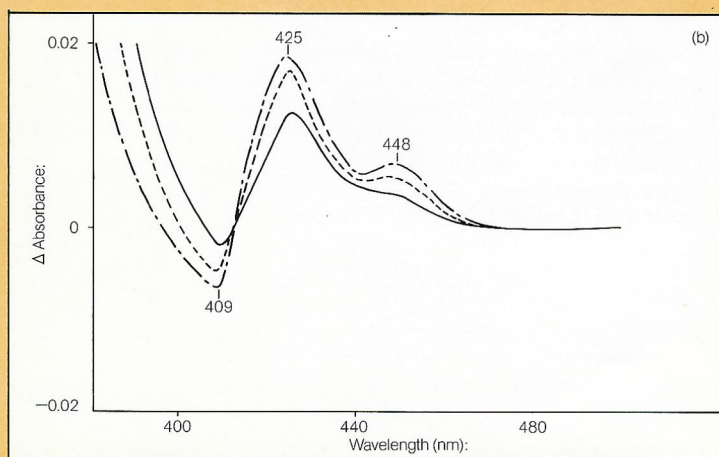
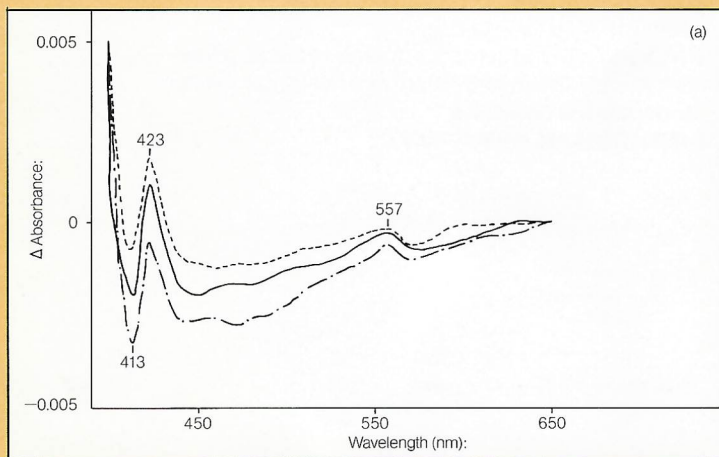
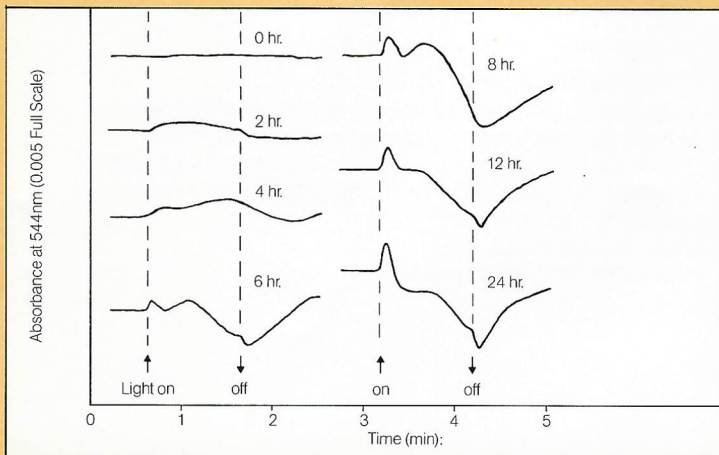
## Agriculture, Food

### Plant Growth

Development of the signal of conformational change during greening of mutant C-2A from *Scenedesmus obliquus*. Conformational changes were measured as absorbance changes (544 nm) due to light scattering under actinic light of wavelengths longer than 620 nm ( $9 \times 10^4$  erg/cm<sup>2</sup> sec). The sample contained 5  $\mu$ l packed cell volume/ml culture medium.

Courtesy of H. Senger

**Mode: Dual-Wavelength**  
**Accessory: Side Illumination**



## Toxic Substances, Pollution

Difference spectra of mouse epidermal microsomes  
Key: (—) acetone pretreated; (-----) 1,2 benzanthracene pretreated; (- · - ·) CSC fraction 20 pretreated.

(a) NADH-reduced minus oxidized microsomes, protein concentration = 0.5 mg/ml.

(b) Carbon monoxide bubbled-dithionite-reduced minus carbon monoxide-bubbled microsomes, protein concentration = 1.0 mg/ml.

Courtesy of W. Norred

**Mode: Double-Beam**

## Medical Research

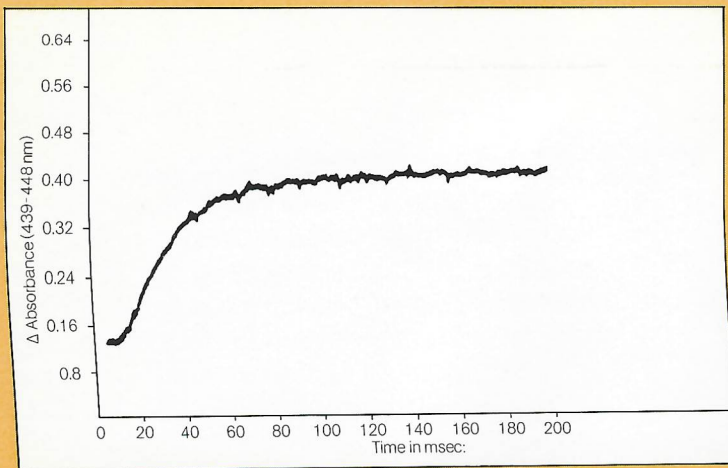
### Sickle Cell Deoxygenation

Human red blood cells containing 95% hemoglobin S were suspended in isotonic bisTris-HCL buffer, pH 7.4. The deoxygenation was achieved in the Stopped-Flow accessory of the DW-2a Spectrophotometer by mixing this suspension with isotonic bisTris-HCL buffer containing 1.1 mg per ml sodium dithionite. The red cell suspension was stabilized by an addition of 20% Ficoll Solution to prevent cell sedimentation.

Courtesy of L. Kiesow

**Mode: Dual-Wavelength**

**Accessory: AMINCO-MORROW® Stopped-Flow**



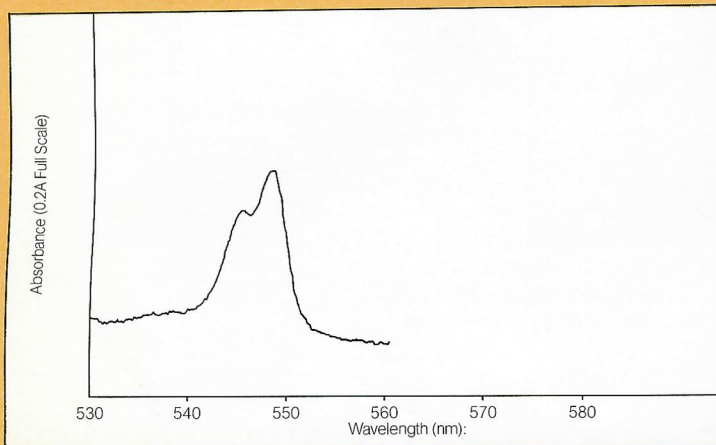
## Cytochrome Research

### Absolute Absorption Spectrum

The sample side of the 2 mm path length Low Temperature cell was filled with reduced cytochrome c solution in phosphate buffer pH 7, the reference side with phosphate buffer pH 7. The cell was then immersed into the dewar containing liquid nitrogen.

**Mode: Double-Beam**

**Accessory: Low Temperature**

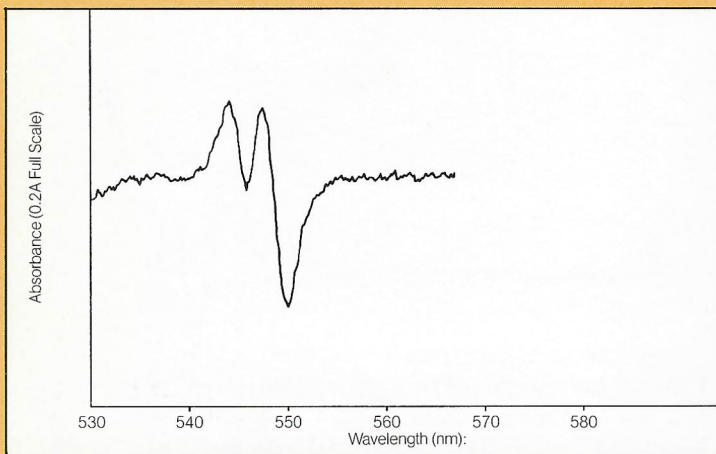


### Derivative Spectrum

Recorded with the same sample as in the absolute spectrum above, however, without reference solution.

**Mode: Optical Derivative**

**Accessory: Low Temperature**

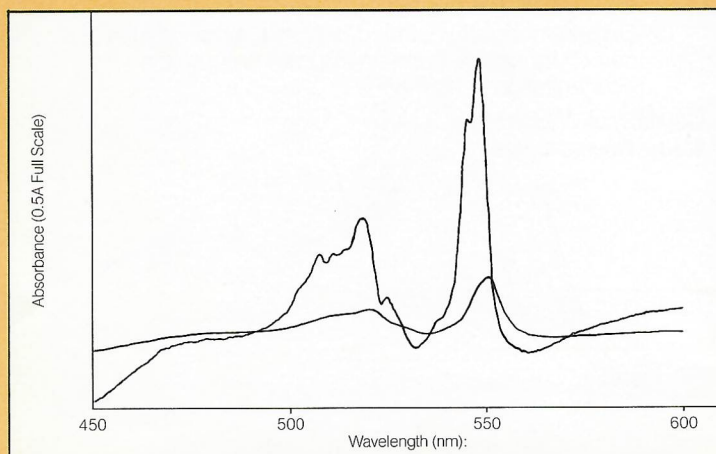


### Difference Spectrum

Recorded in the same way as the absolute spectrum above, except that the reference side was filled with oxidized cytochrome c solution in phosphate buffer pH 7. A room temperature spectrum of the same sample under identical conditions is shown for comparison.

**Mode: Double-Beam**

**Accessory: Low Temperature**



## Drug Research

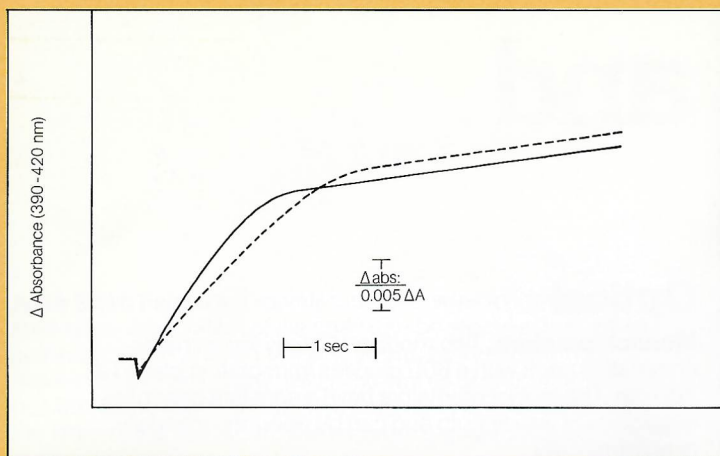
### Hexobarbital Interaction with Cytochrome P-450

Rate of formation of hexobarbital-induced type I spectral change in liver cells from control (-----) and phenobarbital-treated rats (—). Cell concentrations were adjusted to give the same cytochrome P-450 levels in both instances. The reaction was started by plunging 50  $\mu$ l of 0.1M hexobarbital into the 3 ml suspension. Binding was complete in 7-8 sec. Temperature was 20°C.

Courtesy of P. Moldeus

**Mode: Dual-Wavelength**

**Accessory: Anaerobic Cell**



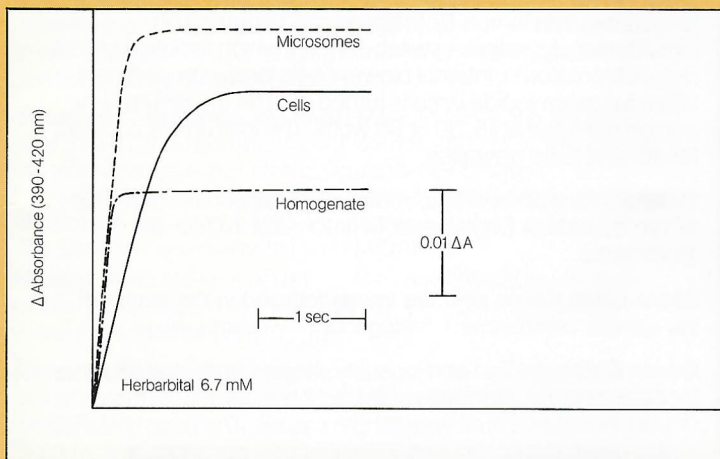
### Hexobarbital Interaction with Cytochrome P-450

Rate of formation of the type I spectral change upon addition of hexobarbital to liver homogenate, microsomes and isolated liver cells: microsomes were suspended to contain 0.8 nmoles P-450 per ml, homogenate to 0.35 nmoles per ml and liver cells were suspended to contain  $3 \times 10^6$  cells per ml, equivalent to 0.5 nmoles P-450 per ml.

Courtesy of C. von Bahr

**Mode: Dual-Wavelength**

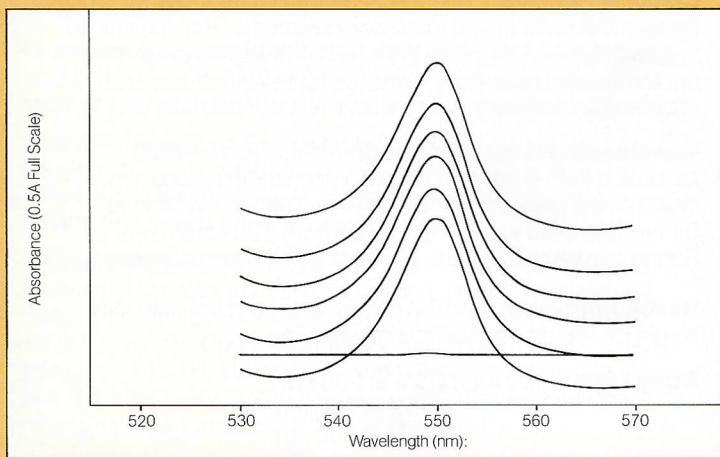
**Accessory: Anaerobic Cell**



## Pure Spectra From Turbid Samples

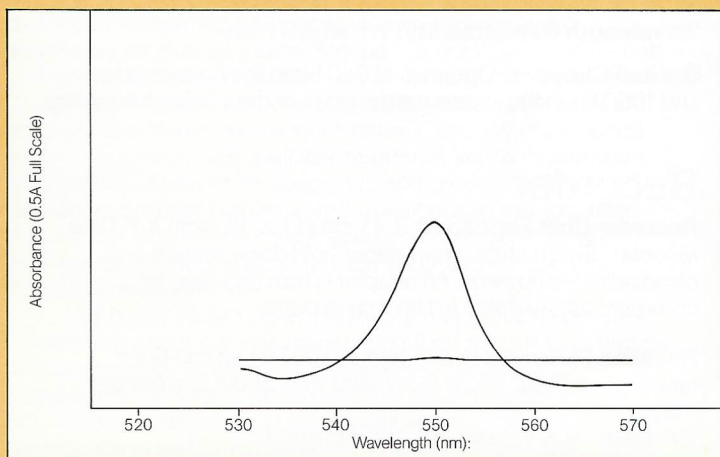
The elimination of light scattering effects in the dual-wavelength scanning mode. Fig. (a) represents absorbance changes with time in a settling medium when recorded in the double-beam mode using 2 cuvettes.

**Mode: Double-Beam**



In Fig. (b) the same scans are superimposed, showing the elimination of light scattering dependent absorbance changes when recorded in the dual-wavelength scanning mode using only 1 cuvette.

**Mode: Dual-Wavelength Scanning**



# Specifications and technical data

## System

**Sample Compartment:** 12.5 cm wide by 16 cm deep by 10.5 cm high ( $5 \times 6\frac{3}{8} \times 4\frac{9}{16}$  inches) with two sample positions for optimizing parameters when using turbid or clear solutions. An external circulator can be connected when temperature control of samples is required. Access ports for side-illumination in both sample positions. Beam geometry allows for use of long path length cells up to 10 cm, microcells, and a diffuse reflectance accessory for solid samples.

**Detector:** End-on (5 cm diameter) quartz window photomultiplier tube with an extended S-20 spectral response. This detector has the best performance for both clear and turbid samples.

**High Voltage Control:** Operates in automatic and manual modes. In automatic mode, the voltage applied to photomultiplier tube is under automatic gain control, which maintains the sensitivity of the instrument constant. In manual mode, 0 to 1400 volts can be applied to the photomultiplier tube. An audible alarm indicates when the high voltage limit is reached.

**Modes of Operation:** Double-Beam, Dual-Wavelength, Dual-Wavelength Scanning, Optical Derivative, Rapid Kinetics.

**Photometric Stability:** Drift is less than 0.0004 absorbance units/hour in the double-beam mode at 550 nm with 3 nm bandpass.

**Photometric Accuracy:** Certified Absorbance within the following limits established using NBS traceable calibration filters:  $0.302 \pm 0.002$ ,  $1.030 \pm 0.003$ ,  $1.613 \pm 0.005$ , and  $2.194 \pm 0.015$ .

**Time Constants:** The DW-2a Spectrophotometer has the following time constants:

Response Pushbutton Selected	Time Constant for Selected Sensitivity	
	Sensitivity Ranges: 1, 2.5, or 5%T; 0.005, 0.01, or 0.02A	All Other Sensitivity Ranges
slow	485 msec	440 msec
medium	185 msec	140 msec
fast	50 msec	5 msec
kinetic	50 msec	0.5 msec

**Power Requirements:** 115 VAC, 50/60 Hz, 700 watts.

**Space Requirements:** The optical unit and the recorder require a minimum table area of  $153 \times 76$  cm ( $5 \times 2.5$  feet) and minimum height clearance of 61 cm (2 feet). The power supply requires  $76 \times 61$  cm ( $2.5 \times 2$  feet) of space and a height clearance of approximately 46 cm (1.5 feet). The recorder unit is usually placed to the right of the optical unit and the power supply anywhere within reach of the interconnecting cables, all of which are 153 cm (5 feet) long.

**Net Weight:** Optical Assembly, 75 kg. (170 lbs.); Recorder Assembly, 48 kg. (110 lbs.); Power Supply, 57 kg. (130 lbs.).

**Shipping Weight:** 259 kg. (580 lbs.).

## Optical

**Monochromators:** Two modified Czerny-Turner monochromators each with a 600 grooves/mm grating blazed at 300 nm. The monochromators have a constant bandpass regardless of wavelength and can be operated manually or automatically.

**Dual-Lamp Compartment:** Contains deuterium and tungsten-iodide lamps. Both lamps can be turned on simultaneously; selector switch controls which radiation enters monochromator(s). Internal blower cools lamp compartment when tungsten-iodide lamp is turned on. The deuterium lamp can be operated at 15, 30 or 60 watts. The instrument is preset for 30-watt lamp operation.

**Slits:** Bilateral curved slits, continuously adjustable from 0 to 17 nm bandpass. Digital readout graduated in 0.02 nm increments.

**Zoom Lens:** Keeps aperture image focused in the center of the sample compartment throughout wavelength range.

**Beam Balance:** Two continuously variable optical attenuators for balancing the intensities of the light beams in both the double-beam and dual-wavelength modes. The beam balance is also used as a continuously variable zero suppression.

**Beam Scrambler:** Optically mixes light beams after they pass through the sample and reference cuvettes so that there is no spatial difference between the beams when they strike the photomultiplier tube. It improves the baseline flatness and photometric accuracy, and minimizes magnetic field effects.

**Wavelength Range and Readout:** Monochromators calibrated from 0 nm to 1200 nm. Wavelength readout on digital scale, graduated in 0.2 nm increments, readable to 0.1 nm. Standard wavelength range from 200 to 825 nm. Range can be extended to 1100 nm with Infrared Accessory.

**Resolution:** Better than 0.3 nm, measured at the half peak height of 486 nm deuterium emission line.

**Stray Light:** Less than 0.2% at 200 nm  
0.03% at 300 nm  
0.01% at 700 nm

**Wavelength Accuracy:** Within  $\pm 0.2$  nm

**Wavelength Repeatability:** Within  $\pm 0.1$  nm

**Optical Chopper:** Operates at 250 Hz in the normal mode and 1000 Hz in the kinetic mode, independent of line frequency.

## Recorder

**Recorder Unit:** Flat-bed  $28 \times 43$  cm ( $11 \times 17$  inch) X-Y-Time recorder. Electrostatic chart paper hold-down system and disposable felt tip pens. An adapter is also provided for commercially available felt tip writing pens.

**Baseline Flatness:** Better than  $\pm 0.0025A$  with 11 point correction without Beam Scrambler throughout wavelength range. The Baseline is virtually flat with MIDAN T.





# AMERICAN INSTRUMENT COMPANY

DIVISION OF TRAVENOL LABORATORIES, INC.

Silver Spring, Maryland 20910 Phone: 301-589-1727

European Headquarters: Rue Dautzenberg 36-38,  
1050 Brussels Belgium, Phone (02) 640-03-20

