



MARAMA LABS

SPECTROSCOPY. UNFILTERED.



# CloudSpec

Combined Extinction/Absorption Spectrophotometer



# UV/Vis Spectroscopy and turbidity

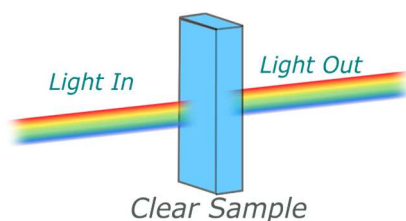
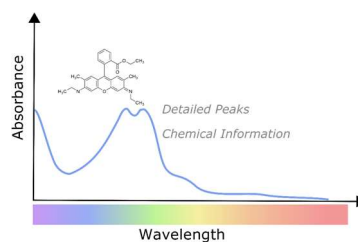


Figure 1: A typical UV-Vis measurement.



## UV-Vis Spectroscopy

Shown in Figure 1, UV-Vis spectroscopy is a simple, yet powerful, technique that uses light in the ultra-violet and visible range to obtain physical and chemical information of liquid samples. The spectrum of light transmitted by the sample can indicate the chemical make up of the sample, simply determine its optical properties (e.g. colour). Because the pathlength inside the cuvette is well-defined (commonly 1cm), it can also be used to quantify molecular concentrations (Fig. 2).

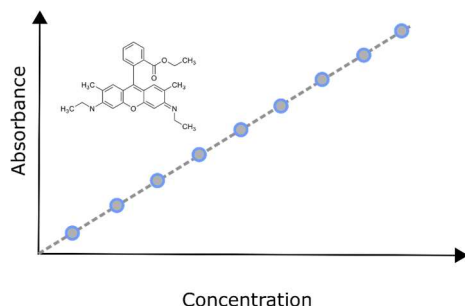


Figure 1: The Beer-Lambert Law for UV-Vis, relating concentration to absorbance.

The Beer-Lambert Law gives a linear relationship between peak intensity (absorbance) and concentration of analyte. A "concentration-curve" can be established and thus an arbitrary sample can be assessed for analyte concentration via measuring its absorption spectrum.

UV-Vis can also be used for checking sample consistency, detection of contaminants, monitoring of reaction kinetics among many other applications. The versatility and simplicity of the technique has made it an invaluable tool for laboratory analysis.

## Extinction, Absorbance, Scattering

Despite often being called *absorbance*, what is actually measured in a UV-Vis experiment is the *extinction* of the sample. Optically, light can be extinguished either through *absorption* (molecular bonds absorbing light)

or *scattering* (light scattered by the sample in many directions). The extinction of the sample is then given simply by:

$$\text{Extinction} = \text{Absorption} + \text{Scattering}$$

As a rule of thumb, molecules do not scatter and only absorb light, while particles (anything bigger than 10nm) will scatter light and may also absorb it. In many applications of UV-Vis spectroscopy, samples are brilliantly clear and contain no scattering component. The absorbance is then equal to extinction, and the Beer-Lambert Law holds.

If the sample scatters however, then the UV-Vis technique no-longer yields a simple absorbance measurement. This could be caused by unavoidable

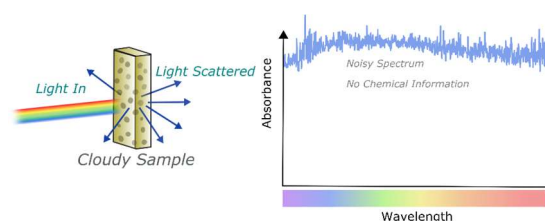


Figure 3: Light scattering in a UV-Vis measurement.

particulates such as fat particle in milk, by bubbles as in beer or fizzy drinks, or simply by contamination by dirt/dust particles. For strong scattering, the sample then appears cloudy/hazy/turbid; in other cases, small amounts of scattering may not be obvious but could still affect your measurement. In cases with high turbidity, such as milk, scattering can be so large that the absorbance of the sample is indiscernible and the spectrum measured contains little chemical information about the sample (Figure 3).

Such cloudy samples usually require extensive pre-processing (filtering, centrifugation, extraction) before a UV-Vis measurement can be taken. Unfortunately, this pre-processing is not always possible if it removes what we wanted to measure.

# The CloudSpec: How it Works

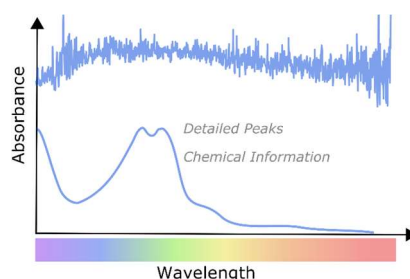
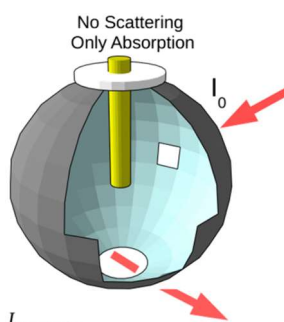


Figure 4: The CloudSpec uses integrating-sphere technology to overcome the effects of light scattering.

## The Integrating Sphere

The CloudSpec, developed by MaramaLabs, solves the problem of light-scattering from cloudy samples by placing the sample inside a device known as an *integrating sphere* (Figure 4). These devices comprise of a spherical cavity made of a highly reflecting material, into which the sample is mounted.

Light is sent into the integrating sphere via an entrance port in the walls of the sphere. The highly reflective walls allow the light scattered by the sample to be reflected many times within the cavity, finally exiting through an exit port and sent to a detector.

*Integrating spheres remove the effects of sample turbidity from UV-Vis measurements. No more filtering, no more centrifugation.*

This means that the scattered light is no longer lost as in an existing UV-Vis configuration, and the only contribution to the reduced light intensity is from the sample absorbance. As such, the “true-absorbance” of a sample can be measured directly without interference from scattering/turbidity.

The CloudSpec uses state-of-the-art, custom-designed integrating spheres to allow high quality absorption spectra to be obtained on turbid samples. It moreover incorporates a unique mechanism to insert standard UV cuvettes inside the integrating sphere: the measurement is as easy as standard UV/Vis.

## Combined Extinction-Absorbance Spectroscopy

The CloudSpec’s unique optical design has two pathways in which light interrogates the sample. The first is the standard “UV-Vis” configuration that yields the extinction spectrum of the sample, while the second pathway uses the integrating sphere to measure the absorption spectrum. We call this method *Combined Extinction Absorbance Spectroscopy*. This has a number of advantages:

CloudSpec can be used as a regular UV-Vis instrument for clear liquids

Sample turbidity can be assessed directly

Scattering spectra can be computed by subtracting absorbance from extinction.

A CloudSpec measurement, for a sample that both scatters and absorbs light, is shown below. The three

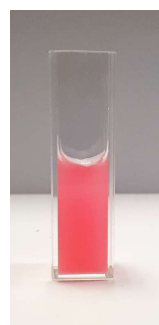
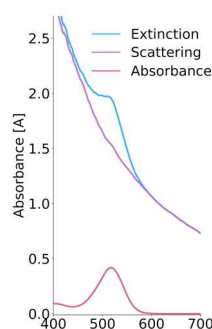


Figure 5: Extinction, Absorption and Scattering spectra obtained for a sample of 10µM Eosin dye + silica particles

spectra are generated automatically. The extinction spectrum is complicated by the presence of scattering from the silica particles, whereas the absorbance spectrum is scattering-free.

## Path Length Enhancement

Because the light in an integrating sphere passes through the sample many times, the *effective path length* can be significantly longer compared to a UV-Vis measurement. In the CloudSpec, this can translate into an increase in path length of up to 4, so effectively a 4cm cuvette is being measured.

This enhancement is shown below for a typical dye sample measured in the CloudSpec, where the CloudSpec absorbance signal is 3 times stronger than the UV-Vis measurement. Samples with absorbances as low as 0.001A can be measured with

*Measure samples of lower concentration, with a standard cuvette, with better accuracy.  
Cloudy or clear.*

ease in the CloudSpec, whereas regular UV-Vis instruments would struggle to accurately measure these signals.

Path length dependence in an integrating-sphere is a complex phenomenon and depends on a range of factors such as sample absorbance and wavelength. The CloudSpec calibration algorithm ensures that all spectra obtained are normalised to path length and thus are directly comparable to standard UV-Vis measurements (i.e. reported in units of  $\text{cm}^{-1}$ ).

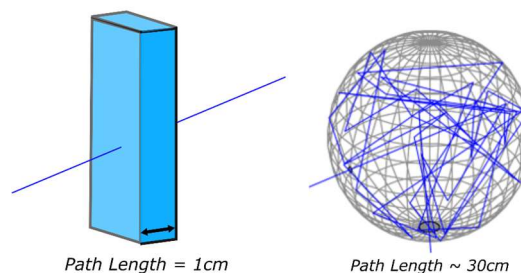


Figure 6: (Left) The optical path of light in a "standard" UV-Vis setup and (right) the path of light travelling within an integrating sphere.

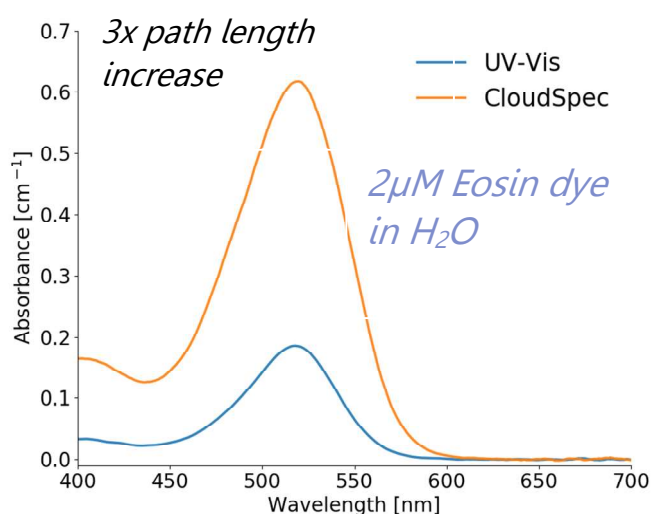
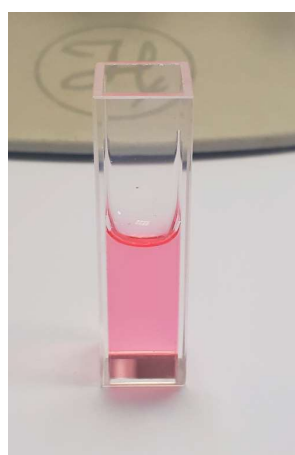
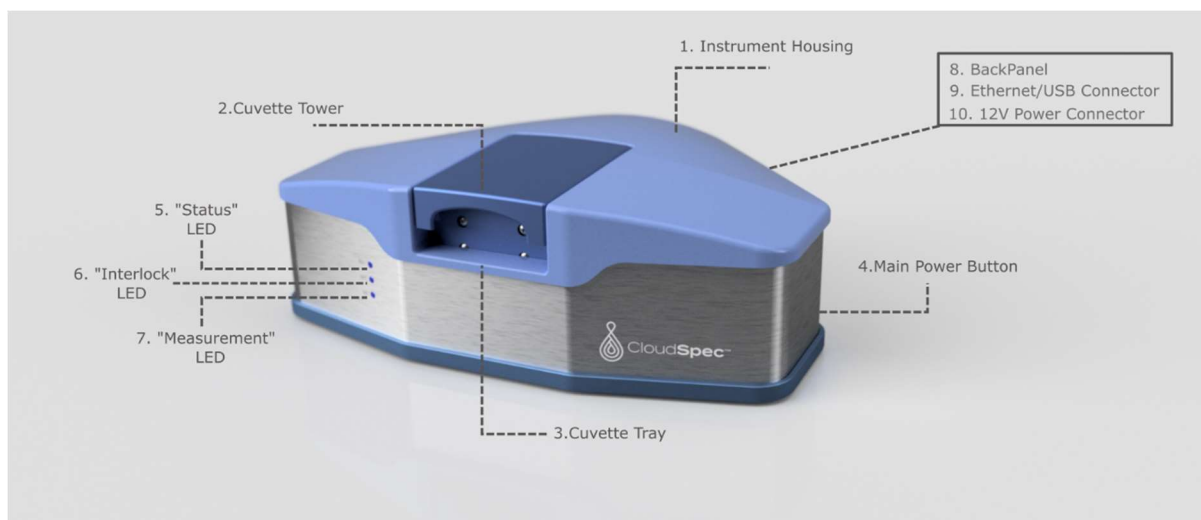


Figure 7: The effect of path length enhancement on an absorption measurement, here for a clear sample of Eosin.

## Product Features



## Calibrated Absorbance

Due to the complex way that light interacts with a sample inside an integrating sphere, the spectra obtained cannot be interpreted using the standard Beer-Lambert law approach, as seen below.

Many approaches have been developed to "calibrate" an integrating sphere device, but most require running lengthy sets of standard measurements.

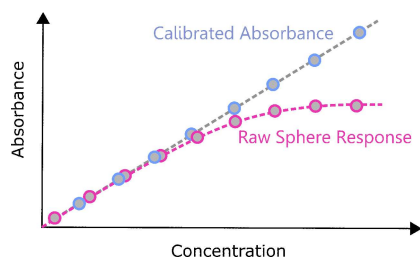


Figure 9: How the CloudSpec algorithm transforms the integrating-sphere response to standard UV-Vis response.

MaramaLabs have developed a powerful algorithm that corrects the spectra obtained so they are normalised to a standard path length. No lengthy empirical calibration need to be run to obtain quantitative UV-Vis spectra, as shown above.

The CloudSpec calibration algorithm ensures that all spectra obtained can be directly interpreted as what would be obtained in a standard 10mm path length cuvette. Therefore the Beer-Lambert law is revived and concentration curves can be developed.

## Features and Advantages

Feature	Explanation
No Filtering of Samples	Integrating sphere eliminates the effects of scattering/turbidity.
Interchangeable Cuvettes	Choose from 1mm, 2mm or 10mm depending on application.
Quantitative Results	Calibration algorithm reports absorbances for a 10mm cuvette
Detection down to 0.001A	Path length enhancement allows ultra-low limits of detection.
Simple to use software	Familiar to any user of UV-Vis
3 Spectra in one measurement	Extinction, absorption and scattering (calculated) output automatically

# Specifications

Parameter	Specifications
Instrument Type	Single-beam, array detector
Wavelength Range	250nm – 800nm
Resolution	~2nm (Spectral Band Width)
Absorbance Range (10mm Cuvette)	0.005 A – 2 A (Extinction spectrum) 0.001 A – 1 A (Absorbance Mode)
Cuvettes	Quartz 10mm (standard) Talk to us for special applications requiring 1mm or 2mm cuvettes
Detector	2048 Pixel CCD
Light Source	Pulsed Xenon
Sample Volume Required	1mL (10 mm Cuvette)
Measurement Time (Typical)	5 – 15 seconds (application dependent)
Dimensions (cuvette tray closed)	45cm (W) x 30cm (D) x 15cm(H)



Figure 9: : The Cloudspec is connected via USB to a standard Windows PC

# Applications

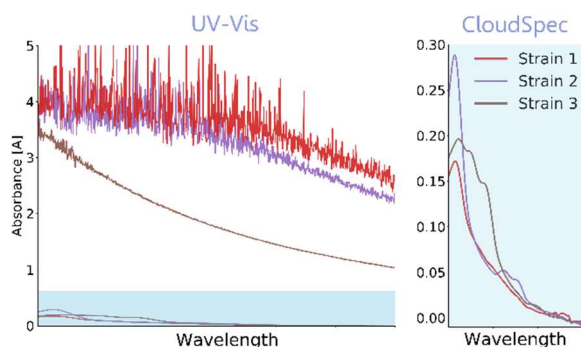


## Biological Samples

Applications for UV-Vis in biological samples typically include measuring protein (280nm), DNA/RNA, and cell concentration, as well as measuring reaction kinetics, metabolite production and detection of contaminants.



However many biological samples, such as cell cultures, can have significant turbidity due to light scattering from cells, and spinning down of samples is often required to measure.

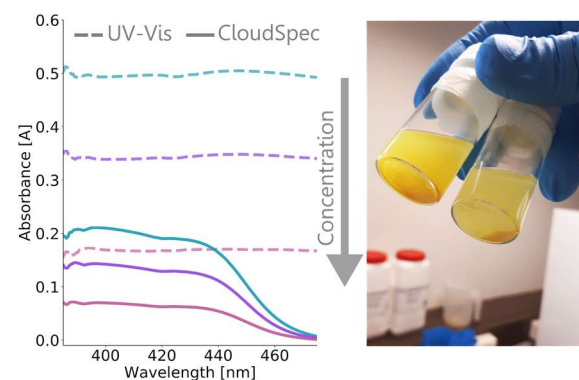


As seen above, the UV-Vis spectrum of unfiltered cultures of three different bacterial strains contain very little spectral information because of scattering.

The CloudSpec spectra (right) reveal a wealth of peaks that differ from strain to strain. This highlights the potential for the CloudSpec to [allow an entirely new way to measure biological samples](#).

## Insoluble Materials

Many materials are non-soluble in convenient solvents and tend to form particles or aggregates in solution. The particles can be strongly scattering and may also settle. [Insoluble materials cannot therefore be studied using standard UV-Vis analysis](#).



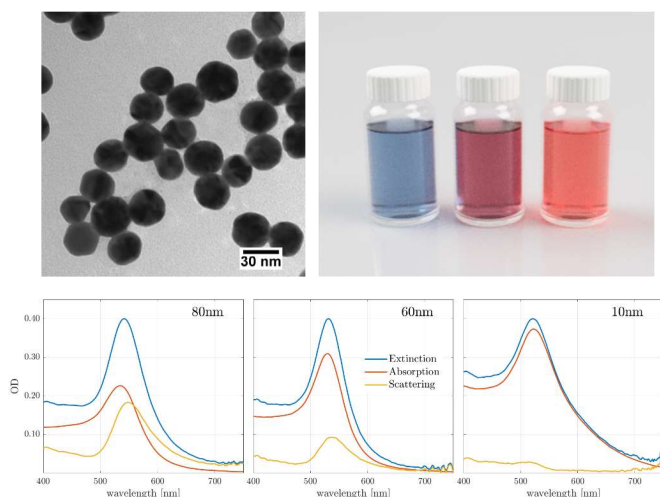
Metal organic frameworks (MOFs) are such an example (shown right). When measured in UV-Vis, spectra are broad and contain little information due to scattering, whereas the CloudSpec spectra have detailed features, here corresponding to a band-edge of the MOF material.

Many interesting applications will be possible for optical characterisation of such difficult samples using the CloudSpec.

## Nanoparticle Characterisation

UV-Vis is a common tool for measuring the optical spectra of nanoparticles, as it can give important information on particle size, concentration and batch consistency. UV-Vis is limited however because only the extinction of the particles is measured.

With the CloudSpec, **extinction**, **absorption** and **scattering** spectra of nanoparticles can be measured directly, yielding significantly more information about the sample. Quantifying scattering and absorbance can be important for both size characterisation and engineering of particles for specific applications.

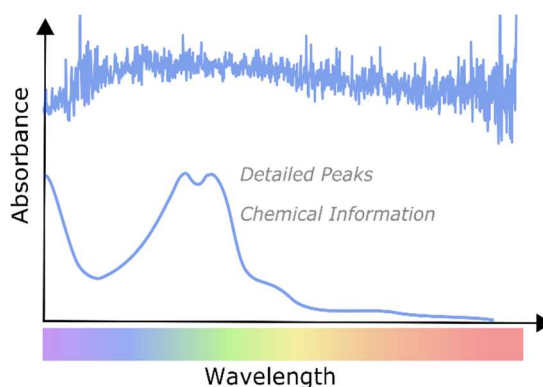


As seen on the left, gold nanoparticles of increasing size, measured in the CloudSpec exhibit varying degrees of scattering and absorbance, which cannot be measured in UV-Vis.

## Food and Beverage

Many samples in the food and beverage industry are turbid, sometimes only slightly turbid as in fruit juice or finished wine, sometimes extremely turbid, for example milk. All these samples can be measured with the CloudSpec to monitor concentrations of compounds or simply colour, as any point in the production process, independently of turbidity.

Even in a sample as turbid as undiluted milk, we observe clear absorption peaks associated with the fat-soluble vitamins (see below, the noisy spectrum is the extinction).



MaramaLabs is also developing special tools and services for wine analysis, for measurement of colour and phenolics content at all steps in the production process. Check out our dedicated wine-related products website: <https://cloudspec.co.nz>



## Get measuring today.

The CloudSpec represents an entirely new way to think about UV-Vis.

Not only does measuring *absorbance in the presence of turbidity* bring obvious time-saving and practical advantages for many sample types, but the added feature of measuring absorption scattering and extinction simultaneously may yield an *entirely new way to characterise complex liquids*.

The applications shown here are just a few examples of where this new technique can be employed. There are many more exciting ideas that the CloudSpec enables, and many that have yet to be explored.

Get in touch with MaramaLabs to begin improving the way you measure your samples.

### Contact Us

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