1.4 Determination of optical rotation and specific rotation

Many substances possess the inherent property to rotate the plane of incident polarized light; this property is called optical activity. The measurement of optical activity is used for pharmacopoeial purposes mainly to establish the identity of the substance. It may also be employed to test the purity of the substance (absence of optically non-active foreign substances) and as an assay procedure.

Optical rotation

The optical rotation is the angle through which the plane of polarization is rotated when polarized light passes through a layer of a liquid. Substances are described as dextrorotatory or levorotatory according to whether the plane of polarization is rotated clockwise or counterclockwise, respectively, as determined by viewing towards the light source. Dextrorotation is designated (+) and levorotation is designated (-).

In the *International Pharmacopoeia* the optical rotation (α) is expressed in angular degrees. In the SI, the angle of optical rotation is expressed in radians (rad).

The optical rotation is measured on a layer of suitable thickness at the wavelength specified in the monograph. If the sodium D line is specified, the sodium light of wavelength 589.3 nm (a mean value for a doublet at 589.0 nm and 589.6 nm) should be used. The wavelength of the mercury green line at 546.1 nm is also frequently used. If the wavelength specified lies in the ultraviolet range, the use of a photoelectric polarimeter is necessary.

The measurement of optical rotation should be carried out at the temperature indicated in the monograph, usually at 20-25 °C. Some substances have large temperature coefficients and for them special care should be taken to adjust the temperature indicated.

Specific optical rotation (specific rotation)

The specific optical rotation of a liquid substance is the angle of rotation measured as specified in the monograph, calculated with reference to a layer 100 mm thick, and divided by the relative density (specific gravity) measured at the temperature at which the rotation is measured.

The specific optical rotation of a solid is the angle of rotation measured as specified in the monograph, and calculated with reference to a layer 100 mm thick of a solution containing 1 g of the substance per mL.

$$\text{Specific rotation} = \frac{10\,000\,a}{lc} = \frac{10\,000\,a}{ldp}$$

where *a* is the observed rotation, *l* is the length of the observed layer in mm, *c* is the number of g of substance contained in 100 mL of solution, *d* is the relative density and *p* is the number of g of substance contained in 100 g of solution.

In the *International Pharmacopoeia* the specific optical rotation is expressed as $[\alpha]_{\lambda}^{t}$ where *t* is the temperature and λ the wavelength. For solid substances the solvent, if different from water, and the concentration are further described. The general directions concerning wavelength and temperature as given above for optical rotation refer equally to the measurement of specific optical rotation. In the SI specific optical rotation (optical rotatory power) is given in m^2·rad/kg and molar optical rotatory power (α_n) in m^2·rad/mol.

Apparatus

Optical rotation is measured with a polarimeter. The zero point of the polarimeter is determined with the tube empty but closed for liquid substances and filled with the specified solvent for solutions of solid substances.

Generally, a polarimeter accurate to 0.05° of angular rotation, and capable of being read with the same precision, suffices for pharmacopoeial purposes; in some cases, a polarimeter accurate to 0.01° of angular rotation, and read with comparable precision, may be required.

Polarimeters for visual measurement: commercial instruments are normally constructed for use with sodium light or a mercury-vapour lamp. The manufacturer's instructions relating to a suitable light source should be followed.

Photoelectric polarimeters: where it is directed in the individual monograph to determine the optical rotation photoelectrically, use a photoelectric polarimeter capable of an accuracy of at least 0.01°.

Measurement of optical rotation

The accuracy and precision of optical rotatory measurements will be increased if they are carried out with due regard for the following general considerations.

Optical elements of the instrument must be brilliantly clean and in exact alignment. The match point should lie close to the normal zero mark. The light source should be rigidly set and well aligned with respect to the optical bench. It should be supplemented by a filtering system capable of transmitting light of a sufficiently monochromatic nature. Precision polarimeters are generally designed to accommodate interchangeable discs to isolate the D line from sodium light or the 546.1 nm line from the mercury spectrum. With polarimeters not thus designed, cells containing suitably coloured liquids may be employed as filters.

Observations should be accurate and reproducible to the extent that differences between replicates, or between observed and true values of rotation (the latter value having been established by calibration of the polarimeter scale with suitable standards), shall not exceed one-fourth of the range given in the individual monograph for the rotation of the substance being tested.

Polarimeter tubes should be filled in such a way as to avoid creating or leaving air bubbles, which interfere with the passage of the beam of light. Interference from bubbles is minimized by the use of tubes in which the bore is expanded at one end. However, with tubes of uniform bore, such as semi-micro or micro tubes, care is required for proper filling.

In closing tubes having removable end-plates fitted with gaskets and caps, the latter should be tightened only enough to ensure a leak-proof seal between the end-plate and the body of the tube. Excessive pressure on the end-plate may set up strains that result in interference with the measurement. In determining the optical rotation of a substance of low rotatory power, it is desirable to loosen the caps and tighten them again between successive readings in the measurement of both the rotation and the zero point. In this way, differences arising from end-plate strain will generally be revealed and appropriate adjustments to eliminate the cause can be made.

The requirements for optical rotation and specific rotation apply to the dried, anhydrous, or solvent-free material in all those monographs in which standards for loss on drying, water, or solvent content are given. In calculating the result, the loss on drying, water, or solvent content determined by the method specified in the monograph should be taken into account.

Recommended procedure

If the substance is a solid, weigh a suitable portion and transfer it to a volumetric flask by means of water, or other solvent if specified in the monograph, reserving a portion of the solvent for the blank determination. Add enough solvent to bring the meniscus close to, but still below, the mark, and adjust the temperature of the flask contents by suspending the flask in a constant-temperature bath. Add solvent to the mark, and mix. Transfer the solution to the polarimeter tube, preferably within 30 minutes from the time the substance was dissolved, taking care to standardize the elapsed time in the case of substances known to undergo racemization or mutarotation. During the elapsed time interval, maintain the solution at the required temperature.

If the substance is a liquid, adjust its temperature, if necessary, and transfer it directly to the polarimeter tube.

When a polarimeter is used for visual measurement, make at least 6 readings of the observed rotation at the required temperature. Take half the readings in a clockwise and the other half in a counterclockwise direction. Substitute the reserved solvent for the solution, and make an equal number of readings on it. In the case of liquid substances, carry out blank determinations on the empty, dry tube. The zero correction is the average of the blank readings, and is subtracted from the average observed rotation if the two figures are of the same sign, or added if they are opposite in sign, to give the corrected observed rotation.

When a photoelectric polarimeter is used, a smaller number of readings is required, depending on the type of instrument.