

## 2.2. SINGLE-CRYSTAL X-RAY TECHNIQUES

Table 2.2.5.1. The distance displacement (in mm) measured on the film versus angular setting error of the crystal for a screenless precession ( $\bar{\mu} = 5^\circ$ ) setting photograph

Angular correction, $\varepsilon$ , in degrees and minutes	$\Delta$ r.l.u.	Distance displacement (mm) for three crystal-to-film distances		
		60 mm	75 mm	100 mm
0	0	0	0	0
15'	0.0175	1.1	1.3	1.8
30'	0.035	2.1	2.6	3.5
45'	0.0526	3.2	4.0	5.3
60'	0.070	4.2	5.3	7.0
1° 15'	0.087	5.2	6.5	8.7
1° 30'	0.105	6.3	7.9	10.5
1° 45'	0.123	7.4	9.2	12.3
2°	0.140	8.4	10.5	14.0

Alternatively,  $\Delta = \delta/D \simeq \sin 4\varepsilon$  can be used if  $\varepsilon$  is small [from equation (2.2.5.1)].

## Notes

- (1) A value of  $\bar{\mu}$  of  $5^\circ$  is assumed although there is a negligible variation in  $\varepsilon$  with  $\bar{\mu}$  between  $3^\circ$  (typical for proteins) and  $7^\circ$  (typical for small molecules).
- (2) Crystal-to-film distances on a precession camera are usually settable at the fixed distance  $D = 60, 75, \text{ and } 100$  mm.
- (3) This table should be used in conjunction with Fig. 2.2.5.1.
- (4) Values of  $\varepsilon$  are given in intervals of  $5'$  as this is convenient for various goniometer heads which usually have verniers in  $5', 6'$  or  $10'$  units. The vernier on the spindle of the precession camera is often in  $2'$  units.

relative to the zero-layer photograph. This effect can be eliminated by initial translation of the cassette by  $D \tan \mu$ .

## 2.2.5. Precession geometry

The main book dealing with the precession method is that of Buerger (1964).

## 2.2.5.1. General

The precession method is used to record an undistorted representation of a single plane of relp's and their associated intensities. In order to achieve this, the crystal is carefully set so that the plane of relp's is perpendicular to the X-ray beam. The normal to this plane, the zone axis, is then precessed about the X-ray-beam axis. A layer-line screen allows only relp's of the plane of interest to pass through to the film. The motion of the crystal, screen, and film are coupled together to maintain the coplanarity of the film, screen, and zone.

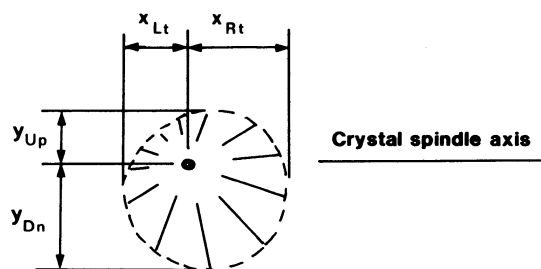


Fig. 2.2.5.1. The screenless precession setting photograph (schematic) and associated mis-setting angles for a typical orientation error when the crystal has been set previously by a monochromatic still or Laue.

## 2.2.5.2. Crystal setting

Setting of the crystal for one zone is carried out in two stages. First, a Laue photograph is used for small molecules or a monochromatic still for macromolecules to identify the required zone axis and place it parallel to the X-ray beam. This is done by adjustment to the camera-spindle angle and the goniometer-head arc in the horizontal plane. This procedure is usually accurate to a degree or so. Note that the vertical arc will only rotate the pattern around the X-ray beam. Second, a screenless precession photograph is taken using an angle of  $\sim 7\text{--}10^\circ$  for small molecules or  $2\text{--}3^\circ$  for macromolecules. It is better to use unfiltered radiation, as then the edge of the zero-layer circle is easily visible. Let the difference of the distances from the centre of the pattern to the opposite edges of the trace in the direction of displacement be called  $\delta = D\Delta$  so that for the horizontal goniometer-head arc and the dial:  $\delta_{\text{arc}} = x_{\text{Rt}} - x_{\text{Lt}}$  and  $\delta_{\text{dial}} = y_{\text{Up}} - y_{\text{Dn}}$  (Fig. 2.2.5.1). The corrections  $\varepsilon$  to the arc and camera spindle are given by

$$\Delta = \frac{\delta}{D} = \frac{\sin 4\varepsilon \cos \bar{\mu}}{\cos^2 2\varepsilon - \sin^2 \bar{\mu}} \text{ in r.l.u.}, \quad (2.2.5.1)$$

where  $D$  is the crystal-to-film distance and  $\bar{\mu}$  is the precession angle.

It is possible to measure  $\delta$  to about 0.3 mm ( $\delta = 1$  mm corresponds to  $14'$  error for  $D = 60$  mm and  $\bar{\mu} \simeq 7^\circ$  [Table 2.2.5.1, based on *IT* II (1959, p. 200)]).

## 2.2.5.3. Recording of zero-layer photograph

Before the zero-layer photograph is taken, an Nb filter (for  $\text{Mo } K\alpha$ ) or an Ni filter (for  $\text{Cu } K\alpha$ ) is introduced into the X-ray beam path and a screen is placed between the crystal and the film at a distance from the crystal of

$$s = r_s \cot \bar{\mu}, \quad (2.2.5.2)$$

where  $r_s$  is the screen radius. Typical values of  $\bar{\mu}$  would be  $20^\circ$  for a small molecule with  $\text{Mo } K\alpha$  and  $12\text{--}15^\circ$  for a protein with  $\text{Cu } K\alpha$ . The annulus width in the screen is chosen usually as 2–3 mm for a small molecule and 1–2 mm for a macromolecule. A clutch slip allows the camera motor to be disengaged and the precession motion can be executed under hand control to check for fouling of the goniometer head, crystal, screen or film cassette;  $s$  and  $r_s$  need to be selected so as to avoid this happening. The zero-layer precession photograph produced has a radius of  $2D \sin \bar{\mu}$  corresponding to a resolution limit  $d_{\text{min}} = \lambda/2 \sin \bar{\mu}$ . The distance between spots  $A$  is related to the reciprocal-cell parameter  $a^*$  by the formula

$$a^* = \frac{A}{D}. \quad (2.2.5.3)$$

## 2.2.5.4. Recording of upper-layer photographs

The recording of upper-layer photographs involves isolating the net of relp's at a distance from the zero layer of  $\zeta_n = n\lambda/b$ , where  $b$  is the case of the  $b$  axis antiparallel to the X-ray beam. In order to determine  $\zeta_n$ , it is generally necessary to record a cone-axis photograph. If the cell parameters are known, then the camera settings for the upper-level photograph can be calculated directly without the need for a cone-axis photograph.

In the upper-layer precession photograph, the film is advanced towards the crystal by a distance

$$D\zeta_n \quad (2.2.5.4)$$

and the screen is placed at a distance