A simple procedure for destaining of polyacrylamide gels by current at right angles to the direction of the electrophoretic separation

The removal of excess dye from gels used in electrophoresis is a necessary, time consuming task which as a rule requires a higher proportion of voltage and current of the power source than the electrophoretic separation itself. Uneven destaining encountered when the electrophoresis of the dye proceeds lengthwise in the same direction as the separation of proteins presents a drawback which prolongs the destaining procedure. Destaining by current at right angles[1–4] to the gel axis, however, needs the construction of a complicated apparatus. The apparatus usually requires gels of exactly known dimensions and/or custom-made parts of plastic. The arrangement designed here diminishes the work necessary in the mechanic's shop to a minimum and can be employed universally.

Apparatus and power supply

Direct current for destaining is supplied by a rectifier with Graetz connection of four, type 36 NP 76 or KY 705, Tesla silicone junction diodes (500 mA, cut-off voltage 700 V) fed by a regulating transformer. For complete destaining within 20 to 25 min a current rate of 30 mA per gel column 60 × 6 mm has been used. The dimensions of the apparatus permit eight columns to be destained simultaneously, the maximum current required being 240 mA. When dealing with gels of larger surface increasing the current intensity should be avoided since it would lead to undesirable overheating and even to dehydration of the gel. For this reason we kept the current at 240 mA for the given surface of viscose pads and prolong the destaining period.

The individual parts of the apparatus, consisting of six parts, are as follows (from the top):

(1) stainless-steel plate, the cathode, 1 × 95 × 125 mm, round edged;
(2) viscose sponge pad 72 × 110 × 12 mm (dimensions in wet state);
(3) cellophane foil, 95 × 125 mm;
(4) rubber spacer for gels, 2.5 × 95 × 125 mm;
(5) viscose sponge pad, 72 × 110 × 12 mm (dimensions in wet state);
(6) stainless-steel plate, the anode, 1 × 95 × 125 mm, round edged.

Destaining procedure

(a) The viscose sponge 5 is placed on the anode plate 6 and wetted with 7 % acetic acid until saturated.

(b) On top of the pad is put the rubber spacer (4) which has longitudinal slits in which the gels, prewashed with 7 % acetic acid, are accommodated. The gels are inserted with the aid of a pair of tweezers.

(c) The gels are covered with cellophane to prevent the dye from diffusing into the upper viscose pad before the current is switched on.

(d) A viscose pad saturated with 7 % acetic is placed on top of the spacer and on top of this the cathode plate. The layers are held together by rubber strips at three points. The gels must be in contact with the viscose pads on both sides, *i.e.* they must not extend over the edges of the pad.

(e) The assembled apparatus is accommodated in a glass dish, cathode upside and the current (30 mA per column, 240 mA max.) is switched on. At intervals we

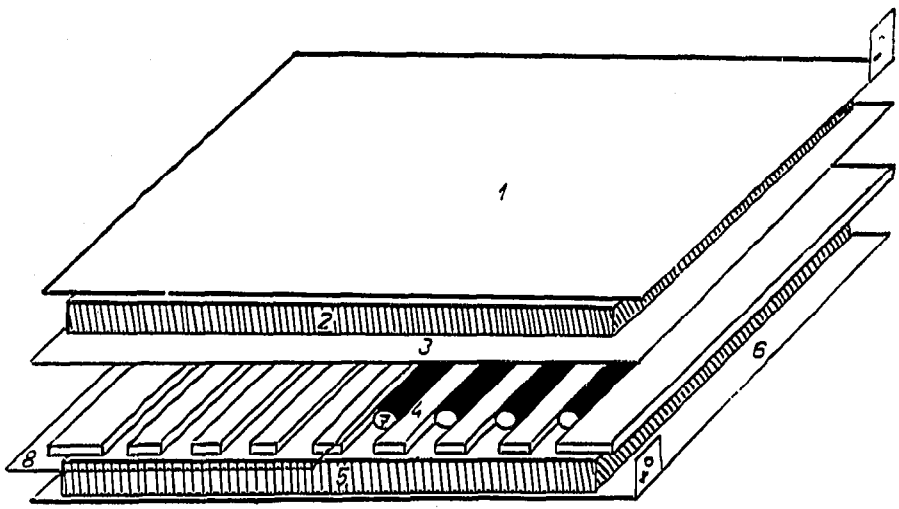

Fig. 1. Destaining apparatus. 1 = Cathode; 2 = viscose pad; 3 = cellophane; 4 = rubber spacer pad; 5 = viscose pad; 6 = anode; 7 = gels; 8 = insulating polyethylene foil on unused slits.

keep a check on the current to ensure that it does not exceed the required value.

(f) After 20 to 25 min the current is switched off (the voltage stays below 50 V) and the apparatus is dissembled. The gels are stored in 7 % acetic acid with traces of the dye. The viscose sponge pads are wrung out and thus most of the dye removed from the lower pad. The rest of the dye can be removed by soaking of the sponge. The dye, however, need not be removed completely since each pad is always used with the same electrode.

Institute of Organic Chemistry and Biochemistry, Z. PRUSÍK
Czechoslovak Academy of Science, Prague (Czechoslovakia)

1 C. F. MATSON, *Clin. Chem.*, 10 (1964) 644.
2 J. A. M. SCHRAUWEN, *J. Chromatog.*, 15 (1964) 256.
3 L. K. NAGY, B. ROGERSON AND N. TOMKUSS, *Nature*, 212 (1966) 923.
4 R. FARMER, P. TURANO, AND W. J. TURNER, *J. Chromatog.*, 24 (1966) 204.

Received July 10th, 1967