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POLYACRYLAMIDE FLUOROGRAPHY

(Bonner and Laskey, Eur. J. Biochem. 46, 83; Laskey and Miller, Eur. J. Biochem. 56, 335). This method allows a 10x increase in ^{35}S and ^{14}C detection and much greater increase for ^{3}H. 400 dpm of ^{14}C or ^{35}S will be detected in a 24 hr exposure.

A. PPO-DMSO method

Reagents: PPO (Sigma), DMSO, reagent grade, Fisher.
20% w/w PPO in DMSO: To 1.82 liters DMSO (density = 1.1) in brown 1 gel container add 500 g PPO. Or 20g PPO + 73 ml DMSO/gel. Also 20% PPO available from NEN.

Procedure:

1. Directly after electrophoresis or after staining, soak the gel in about 20x its volume of DMSO for 30 min, followed by a second 30 min. immersion in fresh DMSO. The baths can be reused in the same sequence.
2. Immerse the gel in 4 volumes of 20% w/w PPO in DMSO for 3 h.
3. Immerse the gel in 20 volumes of water for 1 hr.
4. Dry gel under vacuum.
5. Place dried gel in contact with RP Royal X-Omat film now called XAR-5, pre-flashed to a density of 0.15 absorbance until above fog background, and expose at -70 °C.

B. EN^{3}HANCE method (revised from NEN instructions). Note: This has proved far less sensitive than above method, perhaps because the fluor is volatile and evaporates during drying of gel under vacuum pump. Greatly reduced sensitivity is found even after minimal drying time of one hour.

1. Place gel in a minimum of 3 volumes of EN^{3}HANCE (150 ml for a large 1.6mm slab). Agitate gently 1 hr.
2. Wash with 2 changes of cold tap water, total of 1 hr.
3. Dry gel and process as in step 5 above.