Application of the Scintillation Camera to the Radiochromatogram

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A fast and accurate method of counting instant thin layer chromatographic segments with a scintillation camera and data processor is described. The error of bisecting segments through sections of free or bound pertechnetate is eliminated.

After establishment of routine radiopharmaceutical quality control using instant thin layer chromatography (ITLC), a search for a faster and more accurate graphic display was made. A solution to the problem was found by applying the "region of interest" functions of the Picker 3c Dynacamera. The combined use of the data processor and "curve photo" capability of the Dynacamera or a comparably equipped scintillation camera provides graphs that are far superior in accuracy to those utilizing bisected silica gel strips with a well counter and spectrometer.

Instant thin layer chromatography has been proven very effective in the evaluation of radiochemical impurities in radiopharmaceuticals *(1)*. The results vary relative to the technique used. Elimination of one of the common errors may be accomplished by counting the final results on a scintillation camera rather than by spectrometer and well counter.

Evaluation of free pertechnetate in a prepared radiopharmaceutical is performed with Gelman's Seprachrome (ITLC) Silica Gel chromatography kit. Approximately 2 μl of the prepared agent is placed on the lower end of the silica gel strip and allowed to dry completely. The strip is then placed vertically in a 4-ml solution of acetone which is allowed to rise to the top of the strip, the free pertechnetate migrating with it *(2)*. The strip is then removed from the solvent, allowed to dry, and then evaluated.

The counting procedure itself presents a high risk of error. Gelman's Seprachrome procedure manual *(1)* suggests bisecting the strip, with strip A containing the radiopharmaceutical and strip B the radiochemical impurity. The strips are counted individually, and the free pertechnetate calculated from the formula $B/(A+B) \times 100 = \%$ FTC. With the use of relatively short strips (10 cm), errors from technique, or oxidation of origin *(2)*, may cause smearing that might spread activity the length of the strip. Evaluating such a strip with the technique described above would result in erroneous interpretations.

This problem could be circumvented by cutting the strip into many parts, counting them separately, and plotting each individual strip. However, this is quite time consuming, and the accuracy of this procedure only improves as the size of each strip approaches zero.

Sunderland *(3)* has developed a method utilizing a ratemeter and strip recorder to plot the curve of labeled and free pertechnetate on a silica gel strip. The strip is passed by a G-M tube while the counts are processed on the ratemeter and plotted on the strip recorder. The end result is an accurate and permanent record of ITLC, with the risk of cutting eliminated.

Materials and Methods

Sunderland's *(3)* technique has been applied to the scintillation camera. The silica gel strip is placed under the crystal of the camera and monitored on the persistance scope. The rotation control is then adjusted in such a way that the strip is lying horizontally along the x-axis of the persistance scope on the Dynacamera. The Y "region of interest" is engaged, and adjusted to encompass the entire width of the silica gel strip. If the "monitor intensify" control is depressed, the strip should show on the scope in its entirety. The data processor is then engaged and the strip counted until a sufficient peak is demonstrated on the data processor scope. This will take 40–100 sec, with statistics improving with increasing time. The camera is previously calibrated for ^{99m}Tc, utilizing a 16% window and a low-energy parallel-hole collimator.

After the peak is built up to a desirable point, the processor is stopped, and the total counts computed. A Polaroid photograph is exposed using the "curve photo" [Fig. 1(A)]. The smaller peak is then isolated at its base by means of a "channel selector," counted, and photographed [Fig. 1(B)]. The percent free pertechnetate is calculated by dividing the counts of the isolated peak by

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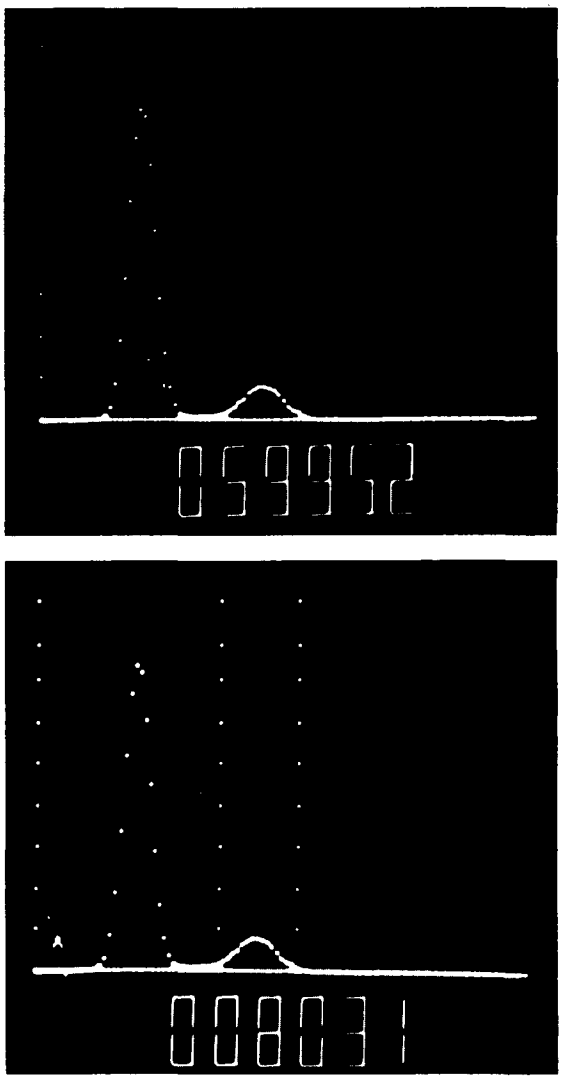

FIG. 1. Polaroid photograph of (A) total curve and (B) isolated free pertechnetate. Percent free pertechnetate = 27.7%.

the total counts of the strip and multiplying by 100. An accurate and permanent record of the ITLC is then ready for review.

This method may be employed on scintillation cameras other than the Picker, provided they are comparably equipped. Isolation of individual areas of the camera's crystal is a necessity. The use of a computer to plot activity versus distance is then required to graph the distribution of radiopharmaceutical over the length of the silica gel strip. Without the ability to isolate and count two individual sources, such as is done in kidney studies, this method of evaluating radiochromatography cannot be performed.

The same procedure was again conducted with "Analtech Preparative" 5 × 20-cm, 500-μg silica gel plates. Approximately 2 μl of the radiopharmaceutical evaluated previously was placed about 1 cm from the base of the plate and allowed to dry. The silica gel plate was then entered into a chromographic chamber containing acetone. The acetone was allowed to rise to the top of the plate, after which the plate was removed and dried. The plate was then counted with the scintillation camera as previously described.

Discussion

Unlike the silica gel strip, the silica gel plate demonstrated a third peak about Rf.=0.35 [Figs. 2 (A and B)]. This peak might possibly be misinterpreted as an additional impurity such as hydrolyzed technetium as described by Eckelman *(4)*. Further investigation revealed this peak to be secondary radiation generated by the origin. This peak only occurs when large amounts of activity (20-50 μCi) are spotted. This problem was quickly eliminated by spotting less activity. The fact that this peak does not appear on the ITLC silica gel strip is probably due to the relatively small distance between

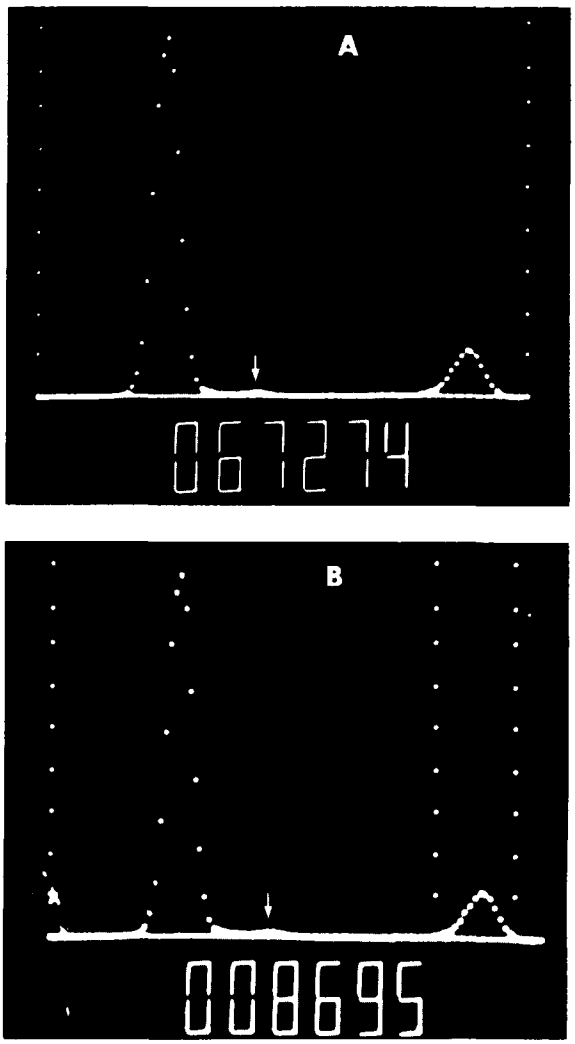

FIG. 2. (A and B) Polaroid photograph of radiochromatogram utilizing 20-cm silica gel plate. Note small peak at Rf.-0.35. Percent free pertechnetate = 16.1%.

TABLE 1. Average Percent Free Pertechnetate of Contaminated Samples

Agent	Well counter	Scintillation camera	Chromatogram scanner
MAA	10.2	3.9	5.8
DTPA	13.6	9.1	6.9
Diphosphonate	45.9	34.5	37.8
Sulphur colloid	6.2	4.4	2.7

the origin and the solvent front. This secondary radiation peak is probably incorporated in the high activity of the origin.

After proving superior to segment cutting, the scintillation camera was then compared with a radiochromatogram scanner. Comparisons were made with pyrophosphate, MAA, DTPA, sulfur colloid, and pertechnetate, using Whatman No. 1 paper developed in acetone (Table 1). The same strip was then counted on both the scintillation camera and a Packard Radiochromatogram scanner. In all cases, the scintillation camera gave results within 2% of the radiochromatogram scanner. The highest margin of errors occurs with free pertechnetate measuring under 1%. Activity this low becomes difficult to distinguish from background on the scintillation camera. The one problem the scintillation camera does eliminate is error incurred from triangulation and planimetry or from both.

Use of the scintillation camera in the counting process of ITLC in daily quality control has proven superior to counting with a well counter and spectrometer. Utilizing a well counter requires that the silica gel strip be cut into separate pieces. The scintillation camera, with 1/8-in. resolution, automatically divides the silica gel strip into approximately three uniformly separate segments per centimeter. A method of plotting, counting, and recording chromatographic strips is available to the Department for evaluating radiopharmaceuticals for gross impurities. Although the scintillation camera does not provide the resolution, collimation, and accuracy provided by the radiochromatogram scanner, it does offer an additional function to an already versatile instrument, and it affords most nuclear medicine departments the opportunity of performing accurate radiochromatography.

Acknowledgments

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