Evaluation of Technetium-99m-Ethylenedicysteine in Renal Disorders and Determination of Extraction Ratio

Levent Kabasakal, Saadet Atay, Vural A. Vural, Kutlan Özker, Kerim Sönmezoglu, Mustafa Demir, Ihami Uslu, Ali Tan Işitman and Çetin Önsel

Department of Nuclear Medicine, Cerrahpaşa Medical Faculty of Istanbul University, Istanbul, Turkey

This article evaluates the clinical usefulness of ^{99m}Tc-ethylenedicysteine (EC) in patients with various renal disorders. In addition, extraction ratios of ^{99m}Tc-EC in five volunteers were also determined. **Methods:** Twenty patients were intravenously injected with 200 MBq ^{99m}Tc-EC and 2.5 MBq [^{131}I]orthoiodohippurate (OIH) simultaneously and 11 blood samples were withdrawn within 60 min. Plasma clearance was determined on the basis of a two-compartment model. Imaging was performed in the posterior projection by acquiring three sets of images. Extraction ratios were determined from the blood samples obtained from the renal vein and abdominal aorta. **Results:** Renal clearance of ^{99m}Tc-EC was significantly lower than that of OIH ($p = 0.0003$) with good correlation ($r = 0.93$). Volume distributions of ^{99m}Tc-EC and OIH were 26584 ± 10807 ml/1.73 m^2 and 23148 ± 7602 ml/1.73 m^2, respectively ($p = 0.047$). The clearance half-lives of ^{99m}Tc-EC and OIH were 98 ± 54 min and 74 ± 54 min, respectively ($p = 0.049$). Protein binding of ^{99m}Tc-EC (33 ± 3.2%) was significantly less than that of OIH (62 ± 2.8%) ($p < 0.0001$). Red blood cell binding of ^{99m}Tc-EC was almost negligible (5.7 ± 4.3%). Similar extraction ratios were obtained from blood (0.68 ± 0.08) and plasma (0.70 ± 0.07) ($p = 0.062$). The 60-min excretion fractions were similar for ^{99m}Tc-EC and OIH, with values of 50% ± 20% and 51% ± 19%, respectively ($p = 0.9$). **Conclusion:** Technetium-99m-EC is a suitable radiopharmaceutical for routine renal dynamic studies. Although the biological behavior of ^{99m}Tc-EC seems to be different from that of OIH, their clearances demonstrate high correlation. Technetium-99m-EC provides excellent quality images and has high potential in the evaluation of quantitative renal functions.

Key Words: renal function evaluation; technetium-99m-ethylenedicysteine; orthoiodohippurate; renogram

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Recently, ^{99m}Tc-labeled L,L-ethylenedicysteine (^{99m}Tc-EC) has been introduced as a new agent for radionuclide renal function and imaging studies (*1*). Technetium-99m-EC is a metabolite of the brain-imaging agent ethylenecysteine dimer (ECD), and, unlike ^{99m}Tc-mercaptoacetyltriglycine (^{99m}Tc-MAG_3), it can be easily prepared at room temperature. Animal biodistribution studies have demonstrated that ^{99m}Tc-EC shows low kidney retention and low liver uptake with rapid urinary excretion through the primary route of active tubular secretion (*1*). In human volunteers, renal clearance of ^{99m}Tc-EC was significantly higher than that of ^{99m}Tc-MAG_3 and it was 75% of orthoiodohippurate (OIH) clearance (*2*). The renogram curves obtained with ^{99m}Tc-EC were superior to ^{99m}Tc-MAG_3 curves, primarily because of lower background and liver activity (*3*). Comparative studies in patients have demonstrated that ^{99m}Tc-EC clearance shows a strong correlation with OIH clearance, which may allow estimation of the effective renal plasma flow (ERPF) (*4–6*).

This study was carried out to evaluate the clinical usefulness of ^{99m}Tc-EC in patients with varying degrees of renal function impairment with concomitant use of OIH as an intrasubject standard. Furthermore, the extraction ratio of ^{99m}Tc-EC also was assessed in subjects with normal renal function to achieve a better understanding of the physiological behavior and renal handling of this agent.

MATERIALS AND METHODS

Patients

Between September 1993 and February 1994, we studied 20 patients, 10 women and 10 men (age range 19 to 50 yr; mean age 35.3 ± 8.7 yr), with various renal disorders who were referred to our department for renal investigations. The study protocol was approved by the Medical Faculty Ethical Committee and all patients gave written informed consent prior to the examination. Clinical diagnoses and creatinine clearance values of patients are listed in Table 1.

Radiopharmaceuticals

Technetium-99m-EC was prepared from kits according to the manufacturer's instructions using 900–1000 MBq (24–27 mCi) freshly eluted ^{99m}Tc. Labeling quality control was performed by thin-layer chromatography and labeling efficiency was found to be over 96% in every case. Iodine-131-OIH was obtained commercially from Amersham, UK.

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For correspondence or reprints contact: Levent Kabasakal, MD, Cerrahpasa Tip Fakültesi, Nükleer Tip Anabilim Dali, Aksaray, Istanbul, Turkey.

TABLE 1
Patient Data and Plasma Clearance of OIH and Technetium-99m-EC

Patient no.	Clinical diagnosis	Creatinine (ml/min/1.73²)	OIH (ml/min/1.73²)	^{99m}Tc-EC (ml/min/1.73²)
1	Hypertension	79	411	325
2	Hypertension	110	541	309
3	Chronic glomerulonephritis	42	257	192
4	Renal amyloidosis	44	211	188
5	Hypertension	81	817	496
6	Chronic renal insufficiency	23	48	39
7	Obstructive renal disease	39	189	190
8	Hypertension	103	670	604
9	Unilateral nephrectomy	75	512	535
10	Obstructive renal disease	52	286	162
11	Unilateral renal hypoplasia	78	400	340
12	Unilateral renal hypoplasia	39	312	249
13	Bilateral hydronephrosis	23	87	60
14	Hypertension	83	556	396
15	Henoch-Shonlein purpura	47	120	69
16	Chronic glomerulonephritis	54	372	224
17	Nephrotic syndrome	27	49	38
18	Nephrotic syndrome	34	89	72
19	Chronic pyelonephritis	89	594	396
20	Hypertension	59	289	196
Mean ± s.e.m.		59 ± 23	341 ± 181	251 ± 136

Clearance Studies

All patients were orally hydrated with 500 ml of water 10–15 min before the study. Immediately after the injection of 200 MBq ^{99m}Tc-EC, 2.5 MBq [^{131}I]OIH was injected intravenously through a three-way stopcock connected to an intravenous catheter and flushed with saline. Subsequently, 11 blood samples were withdrawn at 3, 6, 9, 12, 15, 20, 25, 30, 40, 50 and 60 min from the antecubital vein opposite to the injection site. The 2-ml blood samples were placed in weighted and heparinized tubes and centrifuged to separate the plasma. Plasma radioactivity was determined by counting 0.2-ml plasma samples in a gamma counter. Counts were corrected for the radioactivity decay, background activity and the crosstalk of ^{131}I into the ^{99m}Tc channel. Urine samples were collected by spontaneous voiding at 60 min and 0.2-ml samples of urine were also counted. Postvoidal residual urine correction was not performed. Plasma clearance and pharmacokinetic parameters were calculated using biexponential curve fit analysis with the formula (*7*):

$$Cl = \frac{D \times b_1 \times b_2}{(A \times b_2) + (B \times b_1)},$$

where D represents the injected dose, b_1 and b_2 represent the first and second slopes, respectively, and A and B represent the intercepts of the biexponential clearance curve. The administered patient dose was estimated from the standard activity and the weight difference of the syringes before and after injection.

Excretion fractions were determined from the 60-min urinary activity and expressed as the percentage of injected activity.

Protein binding of ^{99m}Tc-EC and OIH was determined from the 20-min plasma samples by ultrafiltration. Red blood cell (RBC) binding was determined from the 20-min blood and plasma samples and corrected for hematocrit. Hematocrit values were determined by a dilution technique.

Imaging Procedure

Imaging was performed with the patient in the supine position in the posterior projection using a large field of view gamma camera with a low-energy, all-purpose collimator. Three groups of images with 30 frames of 1 sec/frame, 12 frames of 5 sec/frame and 28 frames of 60 sec/frame were obtained. Regions of interest were drawn over the whole kidney on the first 3-min composite image using a computer. The renograms and split renal function values were generated with attenuation correction and taking into account radionuclide decay and background activity. Kidney depth was estimated from the body weight and height of the patients.

Extraction Ratio

Renal extraction ratios of ^{99m}Tc-EC were calculated in five volunteers with normal renal functions who underwent diagnostic arterial catheterization and angiography for suspected coronary artery disease. Immediately prior to the angiography procedure, 100 MBq ^{99m}Tc-EC were injected intravenously and two separate 5-ml anticoagulated blood samples were obtained at 5 and 30 min, from both right renal vein and abdominal aorta. A portion (0.5 ml) of these whole blood samples were set aside and the remainder were centrifuged to obtain plasma. The 0.5-ml samples of whole blood and plasma then were assayed for radioactivity. The extraction ratios were calculated from blood and plasma samples using the following equation (*8*):

$$ER = \frac{(C_A - C_V)}{C_A},$$

where C_A represents the arterial concentration and C_V represents the renal venous blood concentration.

During the procedure, the patient's blood pressure and heart rate were monitored. No significant blood pressure change was observed during the procedure. In Patients 3 and 5, moderate tachycardia was observed during the 30-min interval (Table 3).

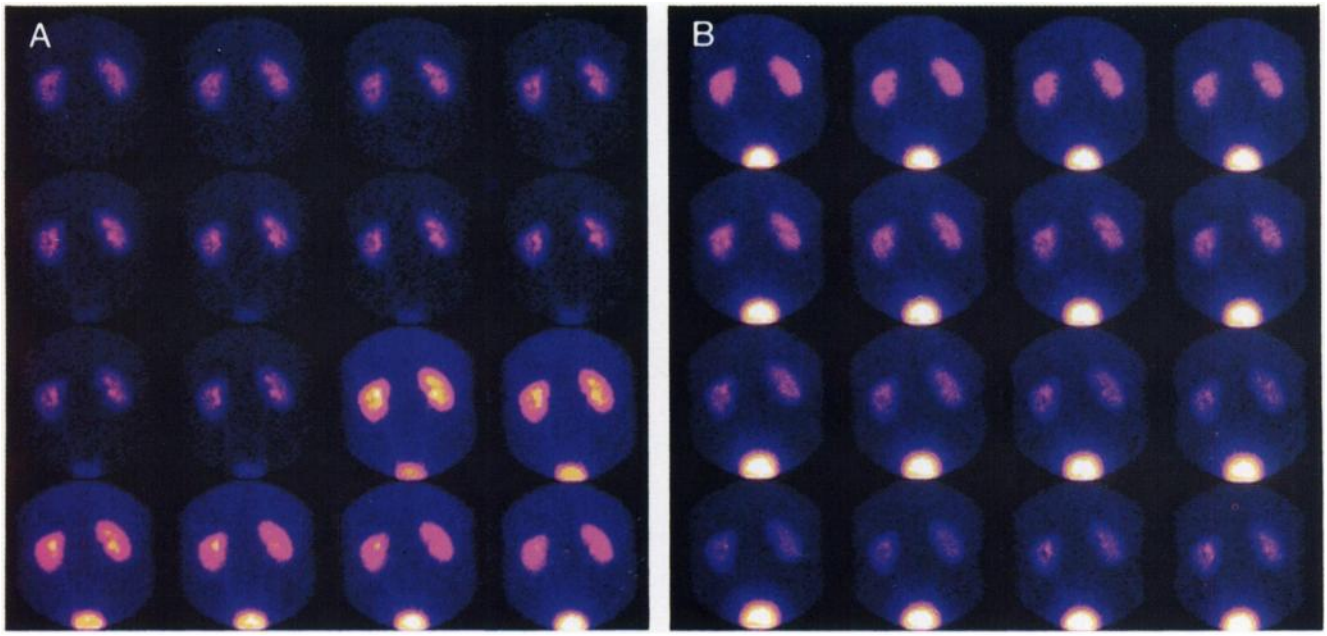

FIGURE 1. Sequential ^{99m}Tc-EC scintigraphic images. The first 10 images were obtained 5 sec/frame and the rest were obtained 60 sec/frame. Low background and liver activity and clear delineation of kidneys are shown.

Statistical analysis was performed using the Student's paired two-tailed t-test and a probability value of $p < 0.05$ was considered significant. Conventional regression analysis was also performed ($p \leq .05$, 95% confidence intervals).

RESULTS

Figure 1 shows the ^{99m}Tc-EC renal images of a patient. There is excellent delineation of the kidneys with high target:background ratios, even in patients with impaired renal function. Generally, faint liver uptake of ^{99m}Tc-EC was observed in the scintigraphic images.

The mean plasma clearance of ^{99m}Tc-EC was found to be significantly lower than OIH clearance ($p = 0.0003$), giving a ^{99m}Tc-EC/OIH clearance ratio of 0.75 ± 0.11. The ^{99m}Tc-EC clearance ranged between 38 ml/min/1.73 m^2 and 604 ml/min/1.73 m^2, whereas OIH clearance was between

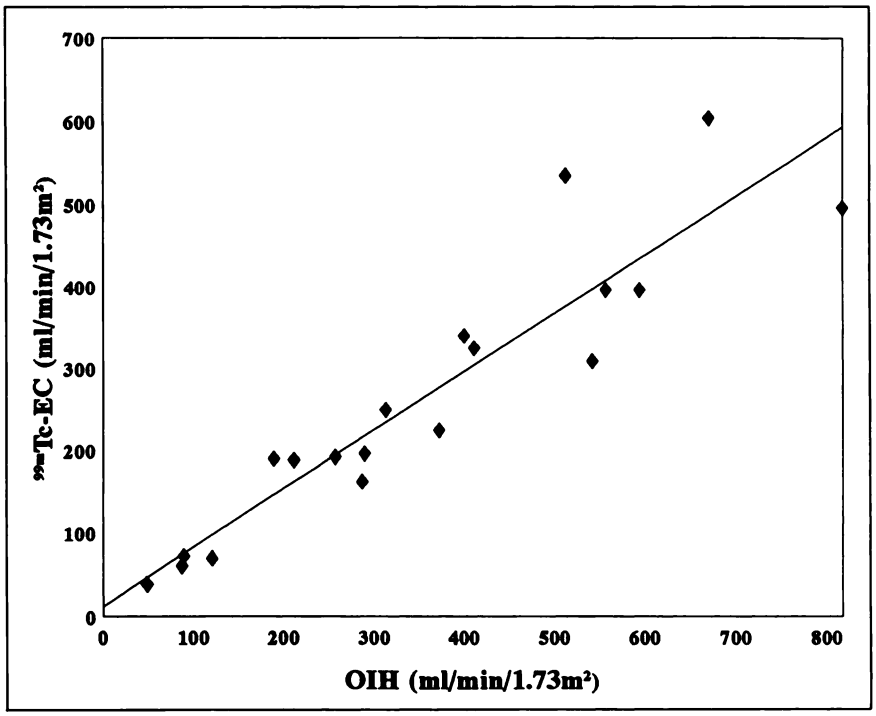

FIGURE 2. Correlation of clearances between ^{99m}Tc-EC (abscissa) and OIH (ordinate). Technetium-99m-EC clearance has high correlation with OIH ($r = .93$).

TABLE 2
Mean Volume Distributions, Compartment Coefficients, Excretion Fractions, Protein Binding, Red Blood Cell Binding and Clearance Half-lives of OIH and Technetium-99m-EC

	OIH	^{99m}Tc-EC
Volume distribution (ml)*	23148 ± 7602	26584 ± 10807
b_1*	0.017 ± 0.008	0.011 ± 0.005
b_2*	0.149 ± 0.07	0.105 ± 0.029
Excretion fraction (%)	51 ± 19	50 ± 20
Protein binding (%) (n = 17)*	62 ± 2.8	33 ± 3.2
RBC binding (%) (n = 17)*	32.6 ± 12.9	5.7 ± 4.3
Elimination half-life (min)*	74 ± 54	98 ± 54

*$p < 0.05$.
b_1 = slow component; b_2 = early component.

48 ml/min/1.73 m^2 and 817 ml/min/1.73 m^2 (Table 1). Figure 2 demonstrates the correlation of clearance between the two agents with a correlation coefficient of $r = .93$.

Total volume distributions of ^{99m}Tc-EC and OIH were 26,584 ± 10,807 ml/1.73 m^2 and 23,148 ± 7,602 ml/1.73 m^2, respectively (Table 2). The distribution volume of ^{99m}Tc-EC was significantly higher than that of OIH, giving a ratio of 1.15 ± 0.24 ($p = 0.047$).

The clearance half-life of ^{99m}Tc-EC was longer when compared to that of OIH ($p = 0.049$) (Table 2). The clearance half-lives of ^{99m}Tc-EC and OIH were calculated to be 98 ± 54 min and 74 ± 54 min, respectively. Figure 3 demonstrates the time-activity curves of ^{99m}Tc-EC and OIH obtained from plasma concentrations of all patients relative to the injected dose. Approximately 40 min after injection, the difference between plasma activity concentrations of ^{99m}Tc-EC and OIH was found to be progressively increased and was notably different at 60 min for ^{99m}Tc-EC. The compartmental constants of ^{99m}Tc-EC were lower than those of OIH ($p = 0.002$, $p = 0.016$ for b_1 and b_2, respectively) (Table 2).

The 60-min excretion fractions were almost identical with a value of 50 ± 20% and 51 ± 19 for ^{99m}Tc-EC and OIH, respectively ($p = 0.9$). The protein binding and RBC binding values, however, were significantly different. Protein binding fractions were 33% ± 3.2% for ^{99m}Tc-EC and 62% ± 2.8% for OIH ($p < 0.0001$) (Table 2). RBC binding ($n = 17$) was almost negligible for ^{99m}Tc-EC compared to OIH (5.7% ± 4.3 and 32% ± 12.9%, respectively).

The renal extraction ratio of ^{99m}Tc-EC and creatinine are presented in Table 3. The extraction ratio of ^{99m}Tc-EC between 5 and 30 min showed no significant change ($p = 0.16$). The extraction ratios obtained from plasma samples (0.70 ± 0.07) and blood (0.68 ± 0.08) were almost similar ($p = 0.062$).

DISCUSSION

Recent animal and human studies have reported promising results with ^{99m}Tc-EC in the evaluation of renal function with dynamic imaging. Currently, ^{99m}Tc-MAG_3 is the widely accepted agent for routine renal dynamic imaging. Comparative scintigraphic and functional dynamic studies have demonstrated that the renogram patterns obtained with ^{99m}Tc-EC and ^{99m}Tc-MAG_3 were almost identical. The time-to-peak activity, time-to-50% activity and split renal function parameters have demonstrated good correlation. Technetium-99m-EC was found to be better than ^{99m}Tc-MAG_3 mostly due to its simplicity of preparation and lower liver and background activities (*3*).

In our patient population, ^{99m}Tc-EC has demonstrated nearly one-half the protein binding value of OIH, which is in agreement with results from previous reports. The lower protein binding of ^{99m}Tc-EC in comparison to OIH probably is the main reason for the higher distribution volume of ^{99m}Tc-EC. The difference in volume distribution between two agents was not as significant as the difference between the protein binding values. A lower RBC binding value for ^{99m}Tc-EC may account for this discrepancy (Table 2).

Our time-activity curves showed that ^{99m}Tc-EC has almost similar plasma concentration to OIH up to 30 min and

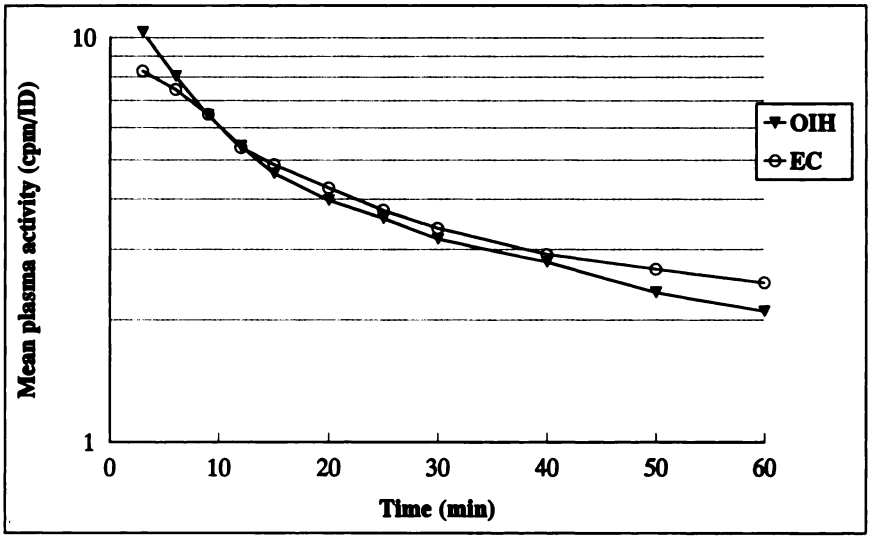

FIGURE 3. Time-activity curves of ^{99m}Tc-EC and OIH obtained by mean plasma concentrations of all patients relative to injected dose. Technetium-99m-EC has high plasma concentration compared to OIH.

TABLE 3
Extraction Ratios of Technetium-99m-EC and Creatinine Obtained 5 and 30 Min Postinjection

Patient no.	^{99m}Tc-EC					
	Creatinine		Blood		Plasma	
	5	30	5	30	5	30
1	0.17	0.17	0.70	0.62	0.71	0.60
2	0.18	0.23	0.76	0.79	0.78	0.81
3	0.22	0.19	0.55	0.58	0.60	0.59
4	0.16	0.23	0.63	0.65	0.73	0.67
5	0.17	0.19	0.80	0.74	0.80	0.75
Mean ± s.e.m.	0.18 ± 0.02	0.20 ± 0.02	0.69 ± 0.08	0.67 ± 0.07	0.72 ± 0.06	0.68 ± 0.08
Mean ± s.e.m.	0.19 ± 0.02		0.68 ± 0.08		0.70 ± 0.07	

exceeds OIH plasma concentration at 60 min (Fig. 2). Since, the elimination half-life is derived from the volume distribution and the clearance, the compartmental coefficients and half-lives of ^{99m}Tc-EC in both compartments were longer than those of OIH (Table 2).

The longer clearance half-life of ^{99m}Tc-EC may give slightly higher total body radiation exposure to the patient compared to ^{99m}Tc-MAG_3, since the reported half-life values for ^{99m}Tc-MAG_3 are not different from OIH (*9*). The higher plasma concentration of ^{99m}Tc-MAG_3 due to higher protein binding may lead to higher radiation exposure to the kidneys compared to ^{99m}Tc-EC (*9,10*).

The 0–60-min excretion fractions of ^{99m}Tc-EC were similar to those of OIH (Table 2). This finding is also in agreement with previously reported values (*2,4*). The discordance between differences in the clearance and excretion fractions can be explained by the slightly higher plasma concentration of ^{99m}Tc-EC (Fig. 3). For a better understanding of renal excretion of ^{99m}Tc-EC, its affinity to the tubular transport system needs further study. Although results from animal experiments have shown that ^{99m}Tc-EC is excreted primarily by tubular secretion, lower protein binding and high volume distribution of ^{99m}Tc-EC may lead to some excretion by glomerular filtration (*1,2*).

In this study, the extraction ratios of ^{99m}Tc-EC in blood and plasma samples were 0.68 ± 0.08 and 0.70 ± 0.07, respectively. The negligible RBC binding of ^{99m}Tc-EC, may be responsible for this similarity in blood and plasma extraction ratios ($p = .062$). The 5- and 30-min extraction ratios were also almost identical ($p = .166$) (Table 3). It seems that the extraction ratio of ^{99m}Tc-EC is lower than that of OIH. The reported values for OIH in humans were about 0.80 (*9,11*). On the other hand, in an animal study (*12*), the extraction ratio of ^{99m}Tc-EC was higher than that of OIH.

Although, we found plasma clearance of ^{99m}Tc-EC to be significantly lower than that of OIH, with a ^{99m}Tc-EC-to-OIH ratio of 0.75 ± 0.11, ^{99m}Tc-EC and OIH clearance have excellent correlation. This correlation may lead to establishing a reliable parameter for quantitating renal function and ERPF estimation. A formulation has been reported for estimating ERPF from ^{99m}Tc-EC clearance (*6*). From the results of our study, we derived a regression equation ($y = a + bx$) for estimating OIH clearance (y) from ^{99m}Tc-EC clearance (x), ($y = 29.9 + 1.22x$).

CONCLUSION

Technetium-99m-EC appears to be a useful alternative radiopharmaceutical for routine renal dynamic studies. Its labeling procedure is simple and ^{99m}Tc-EC provides high-quality images. We have shown that the biological behavior and pharmacokinetics of ^{99m}Tc-EC seem to be different than those of OIH. Technetium-99m-EC clearance, however, demonstrates high correlation with OIH clearance and therefore has high potential for renal imaging and the evaluation of quantitative renal function.

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FIRST IMPRESSIONS

FIGURE 1.

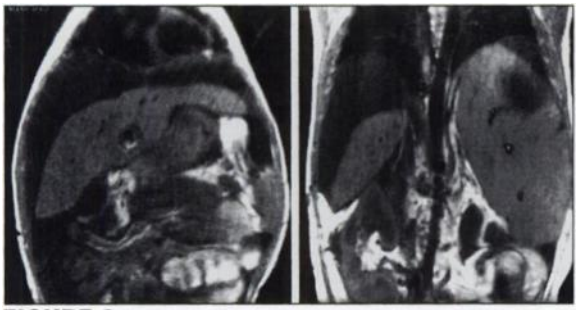

FIGURE 2.

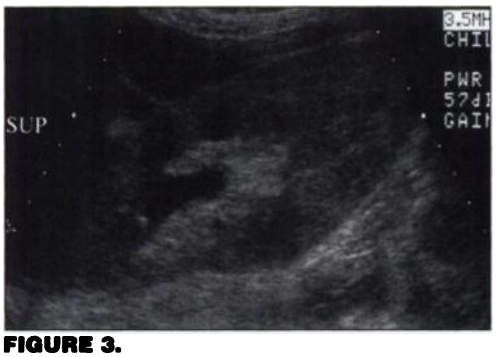

FIGURE 3.

PURPOSE

A 12-yr-old boy with a renal transplant due to juvenile nephronophthysis was referred for a renal scan because of deteriorating renal function. He also had liver failure and portal hypertension secondary to congenital hepatic fibrosis. The 2-min anterior image (Fig. 1) shows increased background activity and reduced uptake in the transplant. Prominent hepatic, splenic and cardiac blood-pool activity are noted as they reflect delayed blood clearance. There is increased distance between the liver and abdominal wall laterally and the heart superiorly, suggesting the presence of ascites. This was confirmed by abdominal MRI (Fig. 2). The relative photopenia of the kidney's upper pole is likely due to both overlying ascites and their more posterior position relative to the lower pole as demonstrated by ultrasound (Fig. 3). This case demonstrates scintigraphic clues indicating the presence of associated advance liver disease on a renal scan.

TRACER

Technetium-99m-MAG3

ROUTE OF ADMINISTRATION

Intravenous

INSTRUMENTATION

Siemens Orbiter single-detector gamma camera eqiupped with a low-energy, high-resolution collimator

CONTRIBUTORS

Zvi Bar-Sever, Leonard P. Connolly, Joan K. Rastegar and S. Ted Treves, Children's Hospital, Boston, MA