Effects of Ouabain on Technetium-99m-Q12 and Thallium-201 Extraction and Retention by Isolated Rat Heart

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The mechanisms of myocardial extraction and retention of the new cationic lipophilic radionuclide imaging agent ^{99m}Tc-Q12 are currently unknown. We hypothesized that ^{99m}Tc-Q12 has satisfactory single-pass extraction independent of active transport processes and longer cellular retention than ^{201}Tl for rapid and sustained cardiac imaging to differentiate perfusion defects. **Methods:** Isolated rat hearts were perfused at constant flow with Krebs-Henseleit buffer enriched with bovine red blood cells (30%–40%). The indicator dilution method was used to measure the single-pass maximum extraction (E_{max}) and net extraction ($E_{net}(t)$) of ^{201}Tl and ^{99m}Tc-Q12 over 15 min during control perfusion (n = 11) and during normal (1 μM, n = 6) and high cardiotoxic (50 μM, n = 11) dose infusions of the digitalis glycoside, ouabain. **Results:** The E_{max} of ^{201}Tl was greater than ^{99m}Tc-Q12 E_{max} (0.73 ± 0.01 and 0.29 ± 0.01, respectively). At 3 min of perfusion, ^{201}Tl E_{net} was greater than ^{99m}Tc-Q12 E_{net} (0.40 ± 0.01 and 0.11 ± 0.00, respectively). Between 3 and 15 min, ^{201}Tl E_{net} was decreasing by a rate of 2% per minute while ^{99m}Tc-Q12 E_{net} was decreasing by less than 0.1% per minute. Ouabain decreased ^{201}Tl E_{max} but did not change ^{99m}Tc-Q12 E_{max}. High-dose ouabain decreased ^{201}Tl E_{net} at 3 min and ^{99m}Tc-Q12 E_{net} at 10 and 15 min. **Conclusion:** Ouabain reduced ^{201}Tl E_{max} but not ^{99m}Tc-Q12 E_{max}. Therefore, the cellular extraction process for ^{99m}Tc-Q12 is different from that of ^{201}Tl. Since the $E_{net}(t)$ of ^{99m}Tc-Q12 was reduced in the presence of high doses of ouabain while E_{max} was unchanged, ^{99m}Tc-Q12 extraction and retention appear to be controlled by different processes. Extraction and release kinetics of ^{99m}Tc-Q12 were not changed with a low dose analogous to the human therapeutic levels of ouabain.

Key Words: cardiac glycosides; radioisotope dilution technique; thallium-201; technetium-99m-Q12; myocardial perfusion

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Several ^{99m}Tc compounds based on a cationic "mixed-ligand" structure have been developed and extensively studied by Deutsch et al. (*1*). One such compound, ^{99m}Tc-Q12 [trans(1,2-bis (dihydro-2,2,5,5,-tetramethyl-3(^{2}H)-furanonato-4-methyleneimino)ethane) bis(tris(3-methoxy-1-propyl)phosphine)^{99m}Tc(III)], has recently been developed into a kit formulation and tested favorably in clinical trials (*2,3*). Canine myocardium in vivo extracts ^{99m}Tc-Q12 in proportion to myocardial blood flow over a physiologically relevant range of flows and the tracer is retained without measurable redistribution for at least 4 hr (*4*). The mechanisms of myocardial extraction and retention of ^{99m}Tc-Q12 are currently unknown. Accordingly, we hypothesized that ^{99m}Tc-Q12 has significant, single-pass myocardial extraction and better retention than the conventional perfusion tracer, ^{201}Tl. We further speculated that myocardial ^{99m}Tc-Q12 extraction was not dependent on active transport processes and therefore would be unaffected by digitalis glycoside inhibition of Na^+-K^+ ATPase. These postulates were tested in the isolated perfused rat heart using the indicator dilution method (*5*). We compared extraction and retention of ^{99m}Tc-Q12 and ^{201}Tl during control coronary perfusion and during infusions of therapeutic (1 μM) and cardiotoxic (50 μM) doses of the digitalis glycoside, ouabain.

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METHODS

Animal Preparation

The animal experimental protocol was approved by the Institutional Animal Care and Use Committee of the University of Cincinnati. Male rats (250–400 g) were anesthetized with 50 mg/kg of pentobarbital-sodium intraperitoneally and given 1000 IU/kg heparin intraperitoneally. The heart was removed quickly and arrested in iced (4°C) normal Krebs-Henseleit (KH) buffer solution containing in m*M*: NaCl, 118; KCl, 4.7; $CaCl_2$, 2.5; $MgSO_4$, 1.2; KH_2PO_4, 1.2; $NaHCO_3$, 25; glucose, 5.5; and Na_2EDTA, 0.5. The heart was then transferred to a Langendorff perfusion apparatus, for coronary perfusion with modified KH buffer containing 30%–40% washed bovine red blood cells and equilibrated to 95% O_2/5% CO_2 with a silicone membrane oxygenator. Sodium bicarbonate was added to the perfusate to obtain a normal arterial perfusate pH range (7.3 to 7.5). Perfusion was controlled with a peristaltic pump. Perfusate flow was measured gravimetrically correcting for the specific gravity of the coronary effluent perfusate. The heart was placed in a water-jacketed chamber (38°C) with air in the space surrounding the heart and electrically paced above the normal sinus rate. The coronary flow rate was set to 2–3 ml/min to obtain an aortic pressure above 30 mmHg. The pO_2, pCO_2 and pH of the coronary perfusate were measured before and after each experiment to insure that they remained in the physiological range (434 ± 6 torr, 33 ± 1 torr, 7.37 ± 0.01, respectively). The heart was stabilized for approximately 10 min at the final coronary flow rate before ouabain or tracer injection. Left ventricular pressure (LVP) was monitored by placing a fluid-filled balloon attached to a Statham transducer into the lumen of the left ventricle. The balloon was inflated to achieve an initial end-diastolic pressure of 3–10 mmHg. Aortic (coronary perfusion) pressure (AoP) was measured with a Statham transducer through a side port near the aorta. LVP and AoP were continuously monitored on a strip-chart recorder. The perfusate temperature at the cannula inlet port was periodically measured with a needle probe and was 36–38°C. Baseline hemodynamic measurements were recorded immediately prior to tracer injection for the control group or immediately prior to ouabain injection for the test groups.

Radiopharmaceutical Cocktail

Thallium-201 was obtained commercially and ^{99m}Tc-Q12 was produced according to the method of De Rosch et al. (*6*) from commercially available kits. The impermeable vascular reference tracer ^{111}In-albumin was produced from ^{111}In-acetate labeled to

DTPA-conjugated albumin and the product label was verified by HPLC analysis in the radiochemistry laboratory according to the methods described by Hnatowich et al. (*7*). The three isotopes were combined in a cocktail consisting of 230 μCi ^{99m}Tc-Q12, 90 μCi ^{201}Tl and 50 μCi ^{111}In-albumin per ml diluted with filtered KH solution. The 100-μl, three-isotope cocktail was injected as a bolus in the coronary perfusate stream directly above the aortic inlet over 3–5 sec.

Sample Collection

Venous effluent for tracer indicator dilution measurements was collected in preweighed vials through a funnel placed under the heart. Collection began immediately after cocktail injection into the coronary artery perfusion cannula. A total of 23 samples were collected over 15 min with 5-sec collection duration up to 1 min and 10-sec collection duration after 1 min. Effluent samples were weighed and isotope activity counted in a gamma well counter. The activities of ^{201}Tl, ^{99m}Tc and ^{111}In were determined by correcting for energy crossover, background and decay and expressed as counts per minute per milliliter for each isotope. All perfusate passing through the heart not collected in sample vials was collected in a beaker, divided into sample vials and counted for isotope activity along with the heart for calculation of total injected dose.

Ouabain Infusion

Ouabain was measured on a precision balance scale, added to distilled water and heated to compose a solution of 10 m*M*/liter. The stock solution was diluted with distilled water to a final injectate concentration of either 38 μM or 1.9 m*M* to deliver either 1 μM or 50 μM, respectively, at the existing perfusate flow rate. Either concentration of ouabain was infused at 80 μl/min directly into the aortic cannula at the point of isotope injection by a syringe infusion pump. The rat heart is rather insensitive to cardiac glycosides as compared to humans, but the two doses of ouabain chosen were reported to produce threshold inotropy and maximal inotropy for rat myocardium (*8,9*).

Indicator Dilution Technique

Myocardial extraction for each tracer was calculated from the venous effluent activity and expressed as the fraction of specific tracer activity appearing per second h(t) (*5,10*):

$$h(t) = F_s \times \frac{C(t)}{q_o}, \qquad \text{Eq. 1}$$

where t = the time after injection, C(t) = (activity/ml) of the sample, q_o = (activity injected) and F_s = the plasma flow rate (ml/sec).

Instantaneous extraction [E(t)] of the diffusible (d) tracers was computed relative to the vascular reference (r) tracer as:

$$E(t) = 1 - \frac{h_d(t)}{h_r(t)}, \qquad \text{Eq. 2}$$

where $h_r(t)$ is the fraction of the injected dose of the vascular reference tracer (^{111}In-albumin) appearing in the effluent per unit time and $h_d(t)$ is the fraction of the injected dose of the diffusible tracer (^{201}Tl or ^{99m}Tc-Q12) appearing in the effluent per unit time. The maximum instantaneous extraction (E_{max}) is the maximum E(t). Net extraction ($E_{net}(t)$) was calculated as:

$$E_{net}(t) = 1 - \frac{\int_0^t h_d(t)\, d\lambda}{\int_0^t h_r(t)\, d\lambda}, \qquad \text{Eq. 3}$$

where λ is a dummy variable for integration. Since the ^{111}In-labeled albumin presumably remains intravascular, the difference between the amount of ^{111}In and ^{99m}Tc or ^{201}Tl appearing in the venous effluent represents the amount of diffusible tracer that has escaped the extravascular space. A negative difference, $h_d(t)$ greater than $h_r(t)$, indicates washout of either ^{99m}Tc-Q12 or ^{201}Tl from the myocardium. The control $E_{net}(t)$ curves were subjected to nonlinear biexponential curve fitting. The time constants are related to the half-time by the relationship $t_{1/2}$ = ln(2)/(time constant).

Experimental Protocols

After a 10-min perfusion stabilization period, ouabain was infused into the aortic cannula so that the concentration in the perfusate at the heart was either 1 or 50 μM. Infusion continued for 10 min before isotope injection and for the duration of sample collection. Isotope injection took place immediately after the equilibration period without vehicle infusion for control hearts. Effluent collection continued for 15 min after isotope injection.

Data Analysis

Hemodynamic parameters in the ouabain groups were compared to the control group by Student's t-test. Aortic and left ventricular pressures at the end of the perfusion period were compared to values immediately prior to tracer injection (baseline) by paired Student's t-test. Instantaneous maximal extraction (E_{max}) and net extraction (($E_{net}(t)$) from the ouabain groups were compared to the control group by Student's t-test. E_{max}, $E_{net}(t)$ and time constants between tracers were compared using a paired Student's t-test. $E_{net}(t)$ over time was examined with repeated measures ANOVA followed by a Bonferroni t-test. A p value < 0.05 was accepted as significant. Data are presented as mean ± s.e.

RESULTS

Hemodynamics

Hemodynamic data are shown in Table 1. Electrical overdrive pacing was not universally achieved in the control or high-dose ouabain groups. High doses of ouabain (100 μM) induced severe arrhythmias (data not shown) consistent with published data for rat ventricular strips (*8*).

Tracer Extraction

The different cardiac extraction and washout characteristics of ^{201}Tl and ^{99m}Tc-Q12 are demonstrated in the representative transport function plot from a control heart (Fig. 1). Thallium-201 E_{max} was greater than ^{99m}Tc-Q12 E_{max} (0.73 ± 0.01 and 0.29 ± 0.01, respectively), as shown in Figure 2. This can also be seen in the h(t) plot (Fig. 1) by comparing the maximum difference between the ^{201}Tl and ^{111}In data points to the maximum difference between the ^{99m}Tc-Q12 and ^{111}In data points. Ouabain decreased ^{201}Tl E_{max} compared to control (low dose, $p < 0.006$; high dose, $p < 0.03$, Fig. 2) but had no effect on ^{99m}Tc-Q12 E_{max}.

Tracer Retention

By 15–20 sec, both ^{201}Tl and ^{99m}Tc-Q12 appear to backdiffuse (washout) from a tissue space or extravascular compart-

TABLE 1
Isolated Rat Heart Hemodynamics

Variable	Control Mean (n = 11)	s.e.	Low ouabain Mean (n = 6)	s.e.	High ouabain Mean (n = 11)	s.e.
CF	2.44	0.23	2.48	0.08	2.32	0.16
HR	256	15	300*	0	248	19
AoP (baseline)	67	14	52	14	76	14
AoP (15 min)	75	13	72	27	77	13
LVP (baseline)	68	10	73	10	57	9
LVP (15 min)	68	10	88*†	9	119*†	13

*$p < 0.05$ compared to controls by Student's t-test.
†$p < 0.05$ compared to baseline by paired Student's t-test.
CF = coronary flow (ml/min/g heart); HR = heart rate (bpm); AoP = aortic coronary perfusion pressure (mmHg); LVP = left ventricular pressure (mmHg).

ment within the myocardium as demonstrated by the negative slope of the $E_{net}(t)$ curve (Fig. 3). At 3 min of perfusion, ^{99m}Tc-Q12 E_{net} was lower than ^{201}Tl (0.11 ± 0.00 and 0.40 ± 0.01, respectively, $p < 0.0001$). Only minimal washout of ^{99m}Tc-Q12 from rat myocardium, however, was observed between 3 and 15 min compared to ^{201}Tl (0.1% ± 0.0% and 2.0% ± 0.1% per minute, respectively, $p < 0.0001$).

Low-dose ouabain tended to decrease ^{201}Tl $E_{net}(t)$ (Fig. 4) but tended to increase ^{99m}Tc $E_{net}(t)$ (Fig. 5), although neither trend gained statistical significance. High-dose ouabain significantly decreased 3-min ^{201}Tl E_{net} ($p < 0.001$) and 10- and 15-min ^{99m}Tc-Q12 E_{net} ($p < 0.05$). High-dose ouabain tended to decrease 5 min ^{201}Tl E_{net} ($p = 0.085$) and 3- and 5-min ^{99m}Tc-Q12 E_{net} ($p = 0.086$ and 0.052, respectively).

Both ^{201}Tl and ^{99m}Tc-Q12 demonstrated biphasic myocardial clearance with a fast early and slow late phase. For the control experiments, the early phase ^{201}Tl time constant was less than the early phase ^{99m}Tc-Q12 time constant (1.710 ± 0.389 and 2.646 ± 0.251 min^{-1}, respectively, $p < 0.05$). The late phase time constant for ^{201}Tl was greater than the ^{99m}Tc-Q12 late phase time constant (0.070 ± 0.006 and 0.012 ± 0.001 min^{-1}, respectively, $p < 0.0001$) for the control experiments. Biexponential numerical simulations predict that by 25 min ^{201}Tl E_{net} will equal ^{99m}Tc-Q12 E_{net} in the control group and that at all later times ^{99m}Tc-Q12 E_{net} would exceed ^{201}Tl E_{net}.

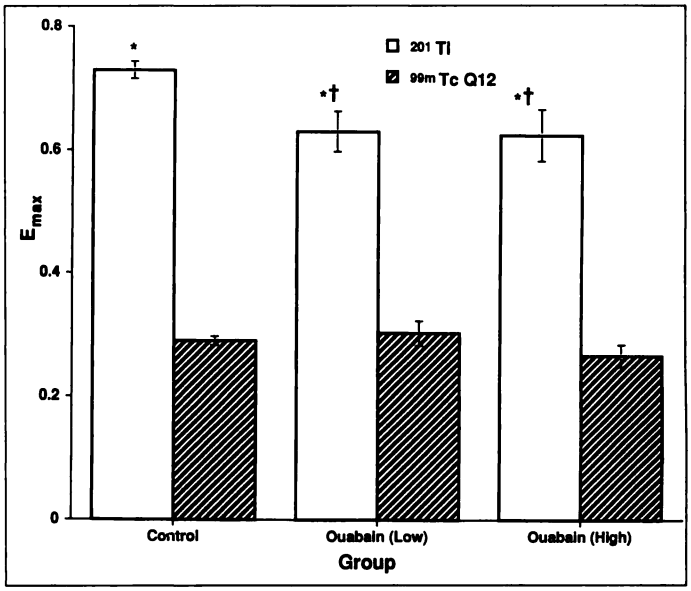

FIGURE 2. Maximum extraction (E_{max}) for ^{99m}Tc-Q12 (hatched bars) and ^{201}Tl (clear bars) for controls (n = 11), low-dose (normal) ouabain infusion (1 μM, n = 6) and high-dose (cardio-toxic) ouabain infusion (50 μM, n = 11) in isolated rat heart. *$p < 0.05$ compared to Q12. †$p < 0.05$ compared to control group.

DISCUSSION

Extraction of ^{201}Tl was greater than ^{99m}Tc-Q12 while ^{99m}Tc-Q12 retention was greater than ^{201}Tl. The average ^{201}Tl E_{max} for controls (0.73) was higher than that reported by others in isolated rabbit hearts (*10*). The present E_{max} determination for ^{99m}Tc-Q12 in isolated rat hearts (0.28) is consistent with reports of ^{99m}Tc-Q12 E_{max} in isolated rabbit hearts (*11*), ^{99m}Tc-sestamibi in isolated rabbit hearts (*10*) and ^{99m}Tc-tetrofosmin in isolated rat hearts (*12*).

Neither low-dose (1 μM) nor high-dose (50 μM) ouabain altered ^{99m}Tc-Q12 E_{max} in the present study. Comparable ouabain doses had no effect on ^{99m}Tc-sestamibi or ^{99m}Tc-tetrofosmin extraction by isolated perfused hearts or isolated myocytes (*10,13,14*). The reported effect of ouabain on ^{201}Tl E_{max} has been more variable (*10,13,15,16*). Ouabain reduced ^{201}Tl E_{max} in our isolated rat hearts (Fig. 2) consistent with expected Na^+-K^+ ATPase mediated uptake. This finding is consistent with the study of McCall et al.

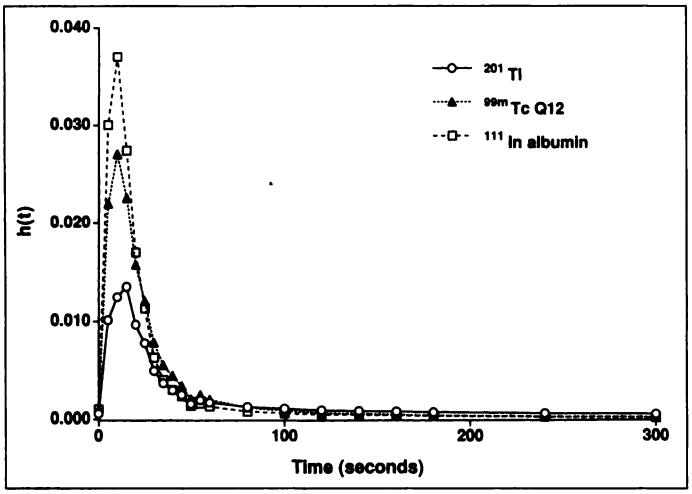

FIGURE 1. Plot of h(t) (mean ± s.e.) truncated at 300 sec obtained from control isolated rat heart. The difference between the ^{111}In-albumin curve (squares) and the ^{99m}Tc-Q12 (triangles) or ^{201}Tl curve (circles) indicates extraction/washout.

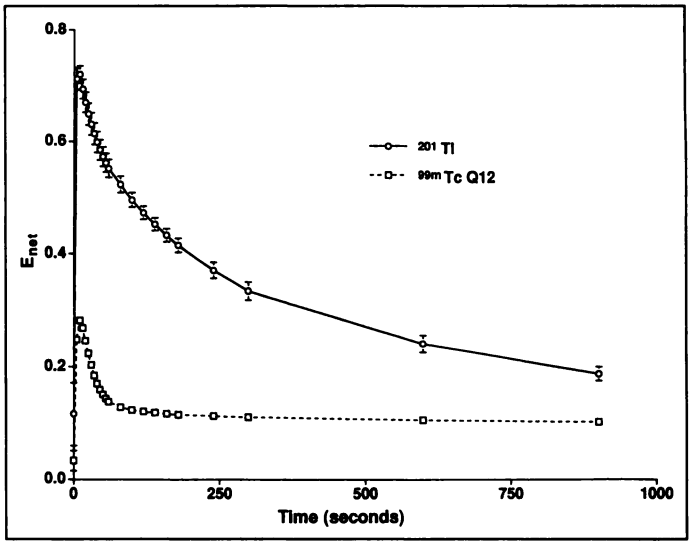

FIGURE 3. Net extraction ($E_{net}(t)$, mean ± SE) of ^{201}Tl and ^{99m}Tc-Q12 for all eight control isolated rat hearts.

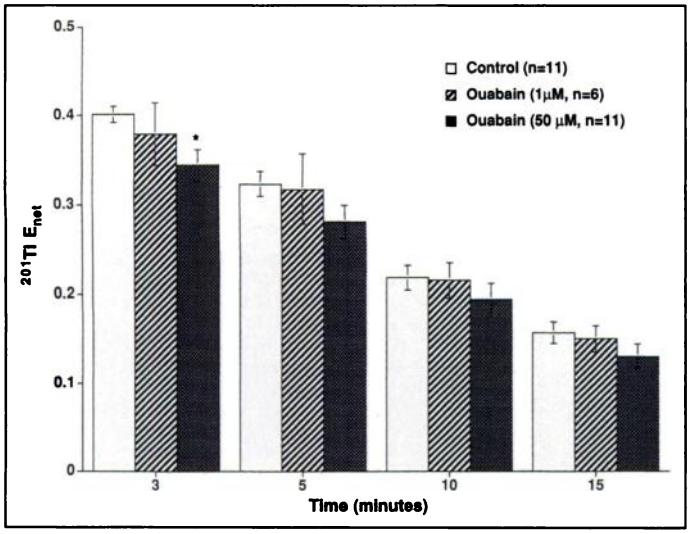

FIGURE 4. Thallium-201 E_{net} as a function of time after injected dose in the isolated rat hearts. *$p < 0.05$ compared to control group.

(*16*), in which ^{201}Tl uptake decreased with all doses of ouabain above 0.1 μM. These data cause us to suggest that ^{201}Tl extraction by rat myocardium may at least be partially dependent on Na^+-K^+ ATPase activity while extraction of ^{99m}Tc-Q12 is not.

Both ^{99m}Tc-Q12 and ^{201}Tl washout of rat myocardium biexponentially. Beginning at approximately 1 min, ^{99m}Tc-Q12 washout decreased while ^{201}Tl washout continued at a higher rate as evidenced by the higher late $E_{net}(t)$ time constant for ^{201}Tl compared to ^{99m}Tc-Q12 and the $E_{net}(t)$ curves in Figure 3. Washout of ^{201}Tl from rabbit myocardium (*17*) and rat myocardium (*18*) was shown to be biexponential with a fast early and slow late phase. A fast early washout phase of ^{99m}Tc-Q12 has also been observed by Johnson et al. (*19*). We did not observe ^{99m}Tc-Q12 washout in canine myocardium in vivo (*4*). Since, however, the earliest cardiac tissue samples were taken at 30 min after tracer injection, we would have missed an early washout period.

The 3-min ^{99m}Tc-Q12 E_{net} by isolated rat hearts compares well to that reported for ^{99m}Tc-tetrofosmin (*12*) but is lower than the ^{99m}Tc-sestamibi E_{net} in isolated rabbit hearts (*10*). The observed $E_{net}(t)$ difference between ^{99m}Tc-sestamibi and ^{99m}Tc-Q12 may be species-dependent but may also be explained by the early fast washout phase of ^{99m}Tc-Q12 (Fig. 3), which does not appear to be a characteristic of ^{99m}Tc-sestamibi. It is possible that the washout of ^{99m}Tc-Q12 compared to ^{99m}Tc-sestamibi may be due to a smaller volume of distribution for ^{99m}Tc-Q12 or to a reversible sequestration mechanism for ^{99m}Tc-Q12.

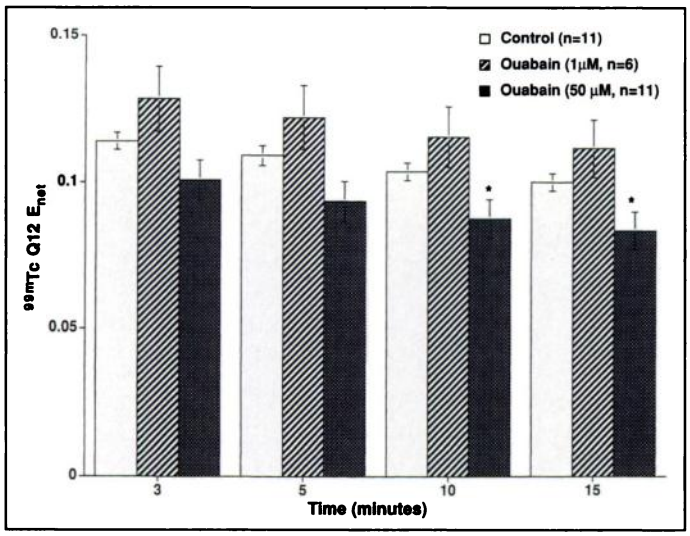

FIGURE 5. Technetium-99m-Q12 E_{net} as a function of time after injected dose in the isolated rat hearts. *$p < 0.05$ compared to control and low-dose ouabain groups.

Three-minute ^{99m}Tc-Q12 E_{net} tended to increase, while the concordant ^{201}Tl E_{net} tended to decrease in isolated rat hearts exposed to 1 μM ouabain. This is consistent with that reported for ^{201}Tl and ^{99m}Tc-sestamibi E_{net} in isolated rabbit hearts (*10*). High doses of ouabain (50 μM) significantly decreased ^{201}Tl 3-min E_{net} in our study probably as a result of the decreased E_{max} since the 10- and 15-min E_{net} values were not significantly reduced. Ouabain (50 μM) decreased ^{99m}Tc-Q12 $E_{net}(t)$, although only the 10- and 15-min values reached statistical significance. Since there was no effect of ouabain on ^{99m}Tc-Q12 E_{max}, the late phase washout may have been accelerated by the ouabain.

Ouabain concentrations greater than 1 μM causes biochemical and ultrastructural changes as well as evidence of metabolic inhibition in isolated perfused guinea pig heart (*21*). During metabolic inhibition in cultured fetal chick myocardial cells, net retention of both ^{201}Tl and ^{99m}Tc-sestamibi decreased, ^{201}Tl influx decreased and ^{99m}Tc-sestamibi influx increased initially before declining with more severe cell injury. Since 50 μM ouabain infusion to isolated rat hearts may induce severe arrhythmias, we are inclined to attribute the accelerated washout of ^{99m}Tc-Q12 we observed to a metabolic impairment unrelated to Na^+-K^+ ATPase activity inhibition. Ouabain may also cause an increase in vascular tone altering capillary flow (*21*), which in turn may alter E_{max} or E_{net}.

Clinical Implications

Net retention at 15 min favors ^{201}Tl over ^{99m}Tc-Q12, but at 15 min, when ^{201}Tl was still being released from the myocardium, ^{99m}Tc-Q12 release was minimal. These data imply that cardiac imaging with the aid of ^{201}Tl should begin as soon as possible to take advantage of the higher ^{201}Tl extraction. Imaging of ^{99m}Tc-Q12 can be delayed if necessary since washout of ^{99m}Tc-Q12 after 1 min was minimal. Clinical ^{201}Tl tomographic cardiac imaging often begins 10–15 min after isotope injection. This practice eliminates the extraction advantage of ^{201}Tl over ^{99m}Tc-Q12. Additionally, 25 min are typically required to complete a serial cardiac tomogram, which further reduces any ^{201}Tl extraction advantage. Assuming a biexponential rate of ^{201}Tl washout, myocardial concentrations of ^{99m}Tc-Q12 and ^{201}Tl relative to injected dose are predicted to be equal by 25 min after injection. Since ^{99m}Tc has a 6-hr physical half-life compared to the 73-hr physical half-life of ^{201}Tl, ^{99m}Tc-Q12 can be administered in doses several times that of ^{201}Tl without increasing the radiation dose to the patient. Thus, in a clinical setting, the total amount of ^{99m}Tc activity in the heart is greater than ^{201}Tl at the time of imaging. These results support a potential clinical advantage of ^{99m}Tc-Q12 over ^{201}Tl.

Limitations of Experimental Models

Because of technical and physiological circumstances, the average heart rate of the low-dose ouabain group was higher than a control group and high-dose ouabain group. Some of the hearts in the high-dose ouabain group could not be captured by electrical stimulation presumably due to the ouabain effect, while some of the control group hearts failed to capture due to technical difficulties. It is, however,

unlikely that the differences in contractility or heart rate between the groups significantly affected the extraction or retention of either ^{99m}Tc-Q12 or ^{201}Tl. Contractility and heart rate were shown previously to be unimportant in the myocardial kinetics of ^{201}Tl (*17,22*). There was no correlation observed between heart rate and either $E_{net}(t)$ or E_{max} of ^{201}Tl or ^{99m}Tc-Q12 among hearts in the control group in the present study.

The isolated rat heart presents challenges when performing indicator dilution measurements. The long sample collection time required in this study will tend to smear h(t) temporally. Smaller sample sizes would increase temporal resolution at the cost of a decrease in sample activity levels and increased uncertainty in sample weight measurements. Since E_{max} is dependent on instantaneous h(t) and since $h_r(t)$ is more narrow than $h_d(t)$, instantaneous extraction may be underestimated. The $E_{net}(t)$ calculation is an integration which reduces the effect of noisy data. Poor temporal resolution, however, contributes uncertainty to the integration at early time points when h(t) has a large slope. The integration may be underestimated before the h(t) peak and overestimated after the h(t) peak. An examination of the equation for $E_{net}(t)$ shows that overestimating the integration of $h_r(t)$ leads to an overestimation of $E_{net}(t)$.

We observed a transient reversible increase in AoP (5–10 mmHg) and decrease in LVP (25%) lasting 10–20 sec upon injection of the radioisotope cocktail. These changes were not associated with any single compound (data not included) and were observed in the hearts in all groups. It is possible that an increase in vascular resistance led to an increase in AoP and a transient decrease in coronary perfusion flow rate which would tend to increase instantaneous tracer extraction. Since the phenomenon was observed in all groups, we expect that these changes did not bias the results from any single group with respect to the others.

CONCLUSION

No studies of cellular mechanisms of ^{99m}Tc-Q12 have been performed, but the extraction and release of ^{201}Tl and ^{99m}Tc-Q12 may be metabolism-dependent but controlled by different cellular mechanisms. Ouabain reduced ^{201}Tl E_{max} but not ^{99m}Tc-Q12 E_{max}. Therefore, the cellular extraction process for ^{99m}Tc-Q12 is different from that of ^{201}Tl. Since the $E_{net}(t)$ of ^{99m}Tc-Q12 was reduced in the presence of high doses of ouabain while E_{max} was unchanged, the extraction and retention of ^{99m}Tc-Q12 appear to be controlled by different processes. Late phase washout of ^{99m}Tc-Q12 appears to be accelerated by high doses of ouabain. Extraction and release kinetics of ^{99m}Tc-Q12 were not changed with therapeutic levels of ouabain.

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