Diabetes and biodistribution of pertechnetate ($Na^{99m}TcO_4$) in rats

Diabetes e biodistribuição de pertecnetato de sódio ($Na^{99m}TcO_4$) em ratos

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ABSTRACT

Purpose: This study aimed to clarify if diabetes induced in rats changes the biodistribution of the radiopharmaceutical sodium pertechnetate ($Na^{99m}TcO_4$) and, consequently, the accuracy of the scintigraphic exams. **Methods:** We used 14 male Wistar rats, randomly allocated in 2 group: the diabetic group (D) rats (n=7) were submitted to the induction of diabetes with streptozotocin, and 7 non diabetic rats were used as controls (C). After 7 days, in the 14 animals it was administered 0,1 mL of $Na^{99m}TcO_4$ (0.66 MBq) through orbital plexus and, after 30 minutes, the radiopharmaceutical sodium pertechnetate was evaluated, being compared the uptske of this in the several studied organs of diabetic animals and controls. **Results:** The biodistribution of $Na^{99m}TcO_4$ was significantly higher in the liver and smaller in the bladder, thyroid and stomach of diabetic mice when compared to the control group rats. **Conclusion:** Diabetes induced in rats alters the biodistribution of $Na^{99m}TcO_4$, and this finding could have clinical implications on scintigraphic exams.

Key words: Bioavailability. Tc 99m Pertechnetate. Diabetes. *S*treptozotocin. Rats.

RESUMO

Objetivo: Contribuir para esclarecer se o diabetes induzido em ratos altera a biodistribuição do radiofármaco pertecnetato de sódio ($Na^{99m}TcO_4$). **Métodos:** Estudo do tipo experimental com utilização de 14 ratos machos *Wistar*, divididos aleatoriamente em 2 grupos, um grupo submetido à indução do diabetes com estreptozotocina, e o outro somente observado. Após 7 dias, nos 14 animais foi administrado 0,1 mL de $Na^{99m}TcO_4$ (o.66 MBq) via plexo venoso orbital e, após 30 minutos, foi avaliada a biodistribuição do radiofármaco, comparando-se a captação deste nos diversos órgãos estudados de animais diabéticos e controles. **Resultados:** A biodistribuição do $Na^{99m}TcO_4$ foi significativamente maior no fígado e menor na bexiga, tireóide e estômago de ratos diabéticos quando comparados aos ratos do grupo controle. **Conclusão:** O diabetes induzido em ratos altera a biodistribuição do radiofármaco $Na^{99m}TcO_4$, podendo esses achados ter implicações clínicas na interpretação de exames cintilográficos.

Descritores: Biodisponibilidade. Pertecnetato. Diabetes. Estreptozotocina. Ratos.

Introduction

The radionuclides are employed in many fields of knowledge. In health sciences they contribute to the improvement of diagnosis and treatment, enabling advances in clinical and experimental research. Most diagnostic tests like scintigraphy, reveals the uptake of radioisotopes in several organs and tissues[1-4].The most used radioisotopes are gamma radiation emitting, as a source of radiation or tracers[1].

Since the 60s, the ^{99m}Technetium (^{99m}Tc) is used in the biomedical area because it is easily obtained from molybdenum/technetium (99Mo/99mTc) generators, it has short half-life (6h), low emission energy, it is easy to label red blood cells, cellular structures or molecular, it has low cost and low environmental impact[5-8]. These features make ^{99m}Tc the most widely used radioisotope, administered in the form of sodium pertechnetate ($Na^{99m}TcO4$) or attached to other molecules. Their biodistribution is evaluated by scintigraphy or other systems to detect radioativity[6].

The scintigraphic image reflects morphology and organ function. Thus, different radioactive compounds can be used to study the physiology of organs and tissues. Therefore, a diseased tissue may contain higher or lower uptake of

radioactivity, depending on its disfunction[8]. Scintigraphic examinations are used in the diagnosis of tumors, metastasis, gastric emptying, esophageal motility, thyroid nodules, bleeding, and in the monitoring of systemic diseases and their consequences, among them, diabetes[9-15].

The metabolic imbalance associated with diabetes causes secondary disorders in multiple organ systems, which can cause alterations in biodistribution of radioisotopes. Changes in scintigraphic examination results can generate false positive images, repetition of exams and increased patient exposure to radiation. Some studies advocate the use of streptozotocin to induce experimental diabetes, because it is a cytotoxic chemical agent, specific for pancreatic beta cells, causing severe primary insulin insuficiency and high glucose levels, followed by the establishment of diabetes in the subsequent 24 hours. This model, although with a high mortality rate is relatively easy, destroying the endocrine cells with preservation of exocrine function of pancreas[16-18].
Considering such principles, the objective of this study was to evaluate the biodistribution of sodium pertechnetate ($Na^{99m}TcO4$) in different organs of rats with diabetes.

Methods

We used 14 three months old Wistar rats, weighing 265g ± 31g, provided by the vivarium of the Health Sciences Center, Federal University of Rio Grande do Norte, Brazil. All animals were weighed, placed in individual cages with water and food (Purina ® Labina) *ad libitum* and acclimated in the laboratory for 7 days. They were kept under controlled temperature (21 degrees), humidity (60-70%), lighting (12/12 h) light / dark cycle and handled in accordance with the Ethical Code for Animal Experimentation (Council for International Organization of Medical Sciences) and the rules of the Brazilian College of Animal Experimentation. They were distributed randomly into two groups: diabetes (D, n = 7) and control group (C, n = 7). The animals in group D were subjected to induction of diabetes by injecting streptozotocin at the dose 80mg/kg, intraperitoneally (IP). Seven days after the injection of streptozotocin, glucose was measured by collecting blood from the dorsal vein of the tail, using the equipment Accu-Chek Advantage, Roche Diagnostics, Mannheim, Germany (2003). We considered diabetic the animals with fasting glucose above 200 mg/dL. The C animals were not subjected to any procedure, only observed.

On day 7, all animals were anesthetized with thiopental (20mg/kg-IP) and Ketamine (20mg/Kg-IM), administered 0, l ml $Na^{99m}TcO4$ intravenously into the orbital plexus, and radioactivity dose was 0.68 MBq. After 30 minutes, the animals were killed with a lethal dose of anesthetic thiopental (100mg/kg), intracardiac, and

samples were harvested from pancreas, brain, thyroid, lung, heart, stomach, liver, kidney, bladder and right femur. The samples were washed in 0.9% NaCl, weighed on a digital precision balance (Bel-Mark 160-II-Italy ®) and taken for detection of radioactivity through the 1470 Auto Gamma counter, WizardTM-Perkin-Elmer (Finland) with automatic correction of decay. The percentage of radioactivity per gram (% ATI/g) of each organ was calculated by dividing the activity per gram of tissue by the total radioactivity administered to each animal. The experiment was completely randomized with statistical analysis by Student t test. The level of significance for the test was 5% (95% CI), ie, $p<0.05$ was considered statistically significant.

Results

Table 1 shows the results of %ATI/g in group D and C. In group D rats we observed a significant increase of the biodistribution of Na99mTcO4 in the liver and reduction in the uptake of thyroid, bladder and stomach when compared with group C rats. ($p <0.05$). The other organs did not show difference in %ATI/g, comparing with controls.

Table 1. Biodistribution of $Na^{99m}TcO_4$ in organs and respective groups

Organs	% ATI/g		p-valor[1]
	Diabetes	Control	
Brain	0.02 ± 0.008	0.01 ± 0.003	0.093109
Thyroid	0.46 ± 0.154	3.89 ± 1.447	0.00075*
Lung	0.41 ± 0.157	0.39 ± 0.141	0.803036
Heart	0.23 ± 0.067	0.29 ± 0.061	0.223256
Stomach	0.43 ± 0.373	3.33 ± 1.593	0.004118*
Liver	2.97 ± 1.745	0.34 ± 0.050	0.009782*
Kidney	0.27 ± 0.161	0.42 ± 0.074	0.106062
Bladder	0.19 ± 0.055	0.34 ± 0.094	0.015865*
Femur	0.15 ± 0.058	0.14 ± 0.032	0.709645
Pancreas	0.15 ± 0.047	0.13 ± 0.046	0.518932

Mean±SD. *Significant diference $p<0.05$.
1- p-valor of Student t test.

Discussion

Research shows the interference of anesthetic drugs, chemotherapy and herbals on the biodistribution of sodium pertechnetate ($Na^{99m}TcO4$). Recently, studies from our laboratory demonstrated changes in the uptake of radiopharmaceuticals in the postoperative of major surgeries involving experimental models of short bowel syndrome and bariatric surgery[19-23].

Diabetes mellitus and associated metabolic disturbances interfere with homeostasis, which may have caused changes in the biodistribution of pertechnetate on this study. We detected an increase in radioisotope uptake in the liver and a reduction in the stomach, bladder and thyroid gland of diabetic rats compared to controls. Bertin et al[24] argued that the control of gastric emptying involves neurological, hormonal and metabolic factors and that the role of diabetes on gastric motility is little known. However El-Shaldy et al[25] attributed the delay in gastric emptying to the inhibition of the hormone motilin by hiperglicemia. Theoretically, it was expected to obtain a higher uptake of gastric pertechnetate in rats of the experimental group, given the prior knowledge of several other trials on diabetic gastroparesis associated with autonomic neuropathy. However, we observed the opposite. This may affect the results of scintigraphy for the study of gastric emptying in diabetic patients commonly performed during the search of dispeptic-symptoms[14,26-31].

Another autonomic dysfunction in diabetes is the neurogenic bladder. Many researchers adopt as diagnostic of this pathology an increased tracer retention in the bladder during the performance of radionuclide cystography. The bladder dysfunction would result from neurological, muscular and urothelial injuries, uriginated on hiperglicemia[32,33].

We suppose that the low bladder uptake of sodium pertechnetate observed in this study was due to a marked polyuria in the experimental group rats, which may have contributed to greater intravesical radioactive energy dissipation. In relation to thyroid disorders associated with diabetes, it is a phenomenon widely reported in the current literature. It is estimated that diabetes is associated with hypothyroidism, Hashimoto's thyroiditis and nontoxic goiter in about 20-56% of cases involving mainly patients with type II diabetes[34-36]. It is therefore of great importance for thyroid evaluations. In this sense, thyroid scintigraphy is used in complementing clinical data and ultrasound. The use ^{99m}Tc displays advantages over iodine^{131}, because it labels the gland, allows its morpho-functional evaluation, with less patient exposure to radiation without interfering with the glandular function[36].

There was a lower thyroid uptake of sodium pertechnetate ($Na^{99m}TcO4$) in our diabetic animals due to a probable subclinical hypothyroidism, as evidenced by

Akbar et al, studying thyroid dysfunction in patients with type II diabetes[37]. Passos et al[38] stated that a reduction in the synthesis of thyroglobulin precursor results in a lower uptake of sodium pertechnetate by the gland, since there is not enough protein to be marked. Thus, we alert to possible changes in thyroid scintigraphy in diabetic patients.

A final analysis concerns the greater hepatic uptake of pertechnetate in the rats of the experimental group. Clinical trials point to the diabetes in the pathogenesis of liver diseases, with emphasis on non-alcoholic steatohepatitis, cirrhosis and hepatocellular carcinoma, including transplanted patient[39-45]. The liver participates directly in the glucose metabolism, raising or lowering the glucose uptake as vary their organic levels. It is postulated that a normalization of blood glucose, would result in regression of these pathological processes in inicial stage[46]. In this study, a likely steatohepatitis could have caused an increased uptake of sodium pertechnetate in the liver of diabetic rats, because once present in the circulation, the radiopharmaceuticals label not only erythrocytes but also leukocytes, showing a higher bioavailability in inflammed sites[47,48]. This phenomenon could alter the results of liver scintigraphy when searching for primary tumors, liver cirrhosis and functional assessments in detecting metastases.

Conclusion

Diabetes altered the biodistribution of sodium pertechnetate ($Na^{99m}TcO4$) in the liver, stomach, thyroid and urinary bladder of rats, emphasizing the need for a careful analysis of the results, if scintigraphic examinations are to be performed in patients with this disease.

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