THE DEVELOPMENT OF *META*-IODOBENZYLGUANIDINE ANALOGUES FOR THE THERAPY OF NEUROENDOCRINE AND OTHER TUMOURS*

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Abstract. Radioiodinated *meta*-iodobenzylguanidine (MIBG) has been extensively used in the diagnosis and therapy of neuroendocrine tumours such as neuroblastoma. We have developed a no-carrier-added synthesis (n.c.a.) for MIBG as well as other analogues which may improve clinical utility. In SK-N-SH human neuroblastoma cells *in vitro*, the uptake of n.c.a. [^{131}I]MIBG remained constant over a 2–3-log activity concentration range. In contrast, the uptake of [^{131}I]MIBG prepared by an exchange radioiodination (ex-[^{131}I]MIBG) steadily decreased over the same range demonstrating the saturability of uptake under these conditions. Similar differences in uptake were seen in normal mouse heart and adrenals, the normal target tissues for MIBG. While no advantage of n.c.a [^{131}I]MIBG over ex-[^{131}I]MIBG was seen in athymic mice hosting SK-N-SH neuroblastoma xenografts, higher tumour uptake and tumour-to-normal tissue ratios were observed when SK-N-BE(2C) xenografts were used. Since neuroblastoma is often associated with micrometastases, an MIBG analogue labelled with the -particle emitting ^{211}At could be advantageous. A method has been developed for the efficient synthesis of *meta*-[^{211}At]astatobenzylguanidine (MABG). A number of *in vitro* assays and tissue distribution studies showed that MABG is an excellent analogue of MIBG. From clonogenic assays using SK-N-SH neuroblastoma cells, it was calculated that the D_0 value for MABG (215 Bq/ml) was more than 1000-fold lower than that of n.c.a. [^{131}I]MIBG. A ^{18}F-labelled analogue of MIBG, 4-[^{18}F]fluoro-3-iodobenzylguanidine ([^{18}F]FIBG), has been prepared and is shown to have a higher uptake in SK-N-SH cells than MIBG. Because it may be an invaluable tool in combination with [^{18}F]FIBG, a method has been developed for the synthesis of its radioiodinated analogue, [^{131}I]FIBG. It was shown that SK-N-SH cells retained FIBG to a significantly higher degree than MIBG over a 3-day period, suggesting that [^{131}I]FIBG may deliver a higher integrated dose to the tumour than [^{131}I]MIBG.

1. INTRODUCTION

Although relatively rare, neuroblastoma is the most common among the solid malignant pediatric tumours; about 15% of all cancer deaths in children are due to neuroblastoma. Despite the use of intensive multimodal therapy regimens, which have increased remission rate and duration, the long term survival of stage IV disease has remained less than 15 per cent. Targeted radiotherapy is an alternative approach because it could allow the delivery of curative doses to tumour while minimizing normal tissue toxicities. Originally developed as an adrenomedullary imaging agent [1], radioiodinated *meta*-iodobenzylguanidine, MIBG (Figure 1) has found use in the diagnosis and treatment of a number of neuroendocrine tumours such as neuroblastoma, pheochromocytoma and carcinoid [2–5]. Although MIBG is an effective diagnostic agent for neuroblastoma and pheochromocytoma [6], its therapeutic efficacy is less than desired [7]. To address this problem, our laboratory is involved in the development of newer analogues of MIBG with the goal of improving its clinical usefulness. This paper describes some of our efforts in this area.

2. NO-CARRIER-ADDED MIBG

Radioiodinated MIBG used in the clinic is prepared by an exchange radioiodination (ex-MIBG) and thus contains a substantial amount of unlabelled carrier. Like norepinephrine, MIBG is taken up by the norepinephrine transporter by an active uptake-1 mechanism [8]. Although there are conflicting reports on the dependence of uptake on specific activity, one study has shown that at higher loading doses (lower specific activity), the uptake of [^{123}I]MIBG was reduced in rat hearts [9]. To investigate

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the effect of specific activity in detail, the availability of radioiodinated MIBG at a no-carrier-added (n.c.a.) level is advantageous. Towards this goal, a silicon precursor of MIBG, 3-trimethylsilylbenzylguanidine (TMSBG) was prepared starting from 3-bromotoluene in 5 steps. It was possible to prepare MIBG of very high specific activity and in >90% radiochemical yield by reacting TMSBG with radioiodine and *N*-chlorosuccinimide in trifluoroacetic acid for 5 min at room temperature.

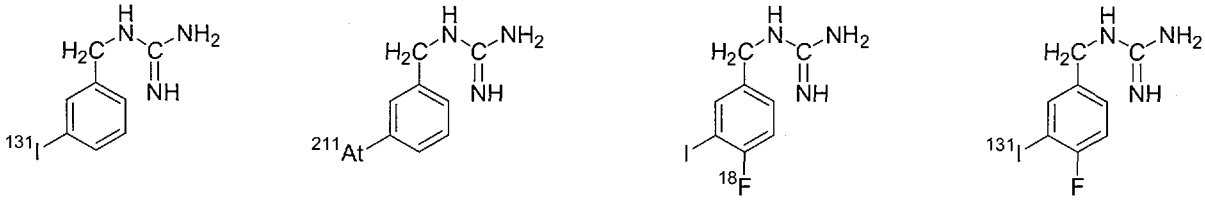

FIG. 1. Structures of [^{131}I]MIBG, [^{211}At]MABG, [^{18}F]FIBG and [^{131}I]FIBG.

2.1. Uptake of n.c.a.- and ex-[^{131}I]MIBG by SK-N-SH human neuroblastoma cells as a function of activity concentration

SK-N-SH human neuroblastoma cells (4–5 × 10^5 cells per well in 500 µl incubation medium) were incubated in quadruplicate with varying concentrations (1.2–233 nM) of ex-[^{131}I]MIBG (370 mBq/mg) in 24-well plates. In parallel, an equivalent activity range of n.c.a. [^{131}I]MIBG (10 000-2 000 000 cpm) was also incubated. After a 2-h incubation, the cell-associated activity was determined. Non-specific uptake was determined by repeating the experiment with another neuroblastoma cell line, SK-N-MC which does not take up MIBG. As shown in Table 1, the specific uptake of n.c.a. [^{131}I]MIBG remained fairly constant whereas that of ex-[^{131}I]MIBG steadily decreased over the 2–3-log activity concentration range. These data clearly demonstrate that the uptake of MIBG by SK-N-SH cells under these conditions is saturable.

TABLE I. UPTAKE OF N.C.A. [^{131}I]MIBG AND EX-[^{131}I]MIBG BY SK-N-SH CELLS AS A FUNCTION OF ACTIVITY CONCENTRATION

	Specific uptake (per cent of input) of [131I]MIBG	
Log Input (count per minute)	No-carrier-added	Exchange Preparation
4.0	48.9 ± 0.8	41.1 ± 1.1
5.0	48.0 ± 1.1	39.1 ± 0.4
5.4	47.3 ± 0.6	30.7 ± 1.5
5.7	45.1 ± 1.2	21.5 ± 1.6
6.0	43.5 ± 0.6	11.4 ± 0.6
6.3	44.3 ± 2.8	6.4 ± 0.6

2.2. Tissue distribution of n.c.a. [^{131}I]MIBG in normal mice

Sympathetically innervated tissues such as heart and adrenals sequester MIBG and can serve as valuable indicators of uptake-1 mediated targeting. To assess the uptake of n.c.a. [^{131}I]MIBG in normal tissues, especially the above, the tissue distribution of the n.c.a. [^{131}I]MIBG in normal mice was compared with that of ex-[^{131}I]MIBG over a period of 24 h. The myocardial uptake of n.c.a. preparation was significantly ($p < 0.05$) higher than that of ex-[^{131}I]MIBG at all time points studied. For example, the heart uptake of the n.c.a. preparation 1 h after injection was 26.4 ± 5.2% ID/g; in comparison, for ex-[^{131}I]MIBG it was 9.2 ± 1.3%, a 3-fold difference. Initially, the difference between

the adrenal uptake of the two preparations was not significant; however, by 24 h, the value for n.c.a. preparation was 4-fold higher than that for ex-[^{131}I]MIBG. These results suggest that n.c.a.[^{131}I]MIBG may be advantageous for clinical applications.

2.3. Effect of specific activity on uptake in vivo *in human neuroblastoma xenograft model*

2.3.1. SK-N-SH model

A comparative biodistribution of n.c.a.[^{131}I]MIBG and ex-[^{131}I]MIBG (2 :g of unlabelled MIBG per mouse) was carried out in separate groups of BALB/c *nu/nu* athymic mice hosting SK-N-SH xenografts. Over a period of 48 h, no significant difference in tumour uptake was seen between the two preparations. For example, at 4 h after administration, when maximum tumour accumulation was observed, the uptake was 3.2 $\pm$ 0.3%ID/g and 2.7 $\pm$ 1.0 %ID/g for the exchange and n.c.a. preparations, respectively. Selective targeting of highly innervated tissues such as heart and adrenals was seen; however, levels of uptake expressed as %ID/g were less than those seen in normal mice. In addition, specific activity did not have an effect on the myocardial or adrenal uptake. However, when the study was performed using non-tumour bearing athymic mice, the myocardial uptake was higher for the n.c.a. preparation. Yet another study was performed in which n.c.a. preparation was administered alone, or with varying amounts of unlabelled MIBG in athymic mice with xenografts. This was done to insure that differences in radiopharmaceutical quality did not obscure any specific activity effects. Again, no differences in tumour uptake were seen. However, at 4 h after administration, the heart uptake was reduced by a factor of 1.5 by the presence of carrier (3 g per mouse).

2.3.2. SK-N-BE(2C) model

Contrary to the above results with SK-N-SH model, higher uptake of n.c.a. [^{131}I]MIBG compared with ex-[^{131}I]MIBG was seen in tumour, heart and adrenals when the biodistribution was carried out in MF1 *nu/nu* athymic mice bearing SK-N-BE(2C) xenografts. In addition, tumour-to-normal tissue ratios were higher for the n.c.a. preparation. For example, the tumour-to-liver ratio for n.c.a. [^{131}I]MIBG was 4.4 $\pm$ 1.8 at 24 h, almost twice that of ex-[^{131}I]MIBG (2.3 $\pm$ 1.0; $p < 0.01$).

There are several factors that might have contributed to the differences observed between the two studies. For example, the strain of mice used for the two studies was different. As mentioned above, reduced heart and adrenal accumulation was seen between normal to athymic mice, suggesting that the uptake may be species-dependent. The most important difference between the two studies was the xenograft model itself. *In vitro* studies have shown that the uptake is saturable in both cell lines at a concentration of about 100 nM. Based on this, one would predict that the amount of unlabelled MIBG used in the SK-N-SH study would have been sufficient to saturate tumour uptake. A plausible explanation may be the differences in the NET. An inverse correlation between the expression of NET and tyrosine hydroxylase, the key regulatory enzyme of catecholamine synthesis, has been observed for SK-N-SH cells [10]. It may be possible that the expression of NET gene is diminished for SK-N-SH cells when implanted *in vivo*. Preliminary results have shown that receptor gene expression by SK-N-SH, but not SK-N-BE(2C) cells, is diminished when the cells are grown as xenografts in MF1 *nu/nu* mice. If this is true for BALB/c *nu/nu* mice also, then a considerable amount of [^{131}I]MIBG uptake by the specific uptake-1 mechanism would be compromised in the SK-N-SH model *in vivo*. Then, the majority of the uptake would be by passive diffusion, where added carrier could actually enhance the uptake.

3. META-[^{211}At]ASTATOBENZYLGUANIDINE

Neuroblastoma, the tumour most commonly treated with [^{131}I]MIBG, often is characterized by micrometastatic disease. Unfortunately, the physical properties of β-particles, such as those emitted by ^{131}I, are suboptimal for smaller tumours. The range in tissue of β-particles is of the order of

millimeters. As a result of this, the fraction of absorbed dose deposited in small tumours decreases as the tumour volume decreases, making [^{131}I]MIBG less than ideal for the therapy of micrometastatic tumours. Alpha particles, on the other hand, have ranges of 50–100 :m and hence their energy is fully absorbed within a few cell diameters. In addition, α-particles are radiations of high linear energy transfer (LET), and thus have higher relative biological effectiveness. Astatine-211 is a 7.2 h α□emitter of particular interest for endoradiotherapy. Being a halogen, it is generally easy to introduce ^{211}At onto organic molecules by adapting radioiodination chemistry. Furthermore, astatinated compounds often retain the biological characteristics of their iodinated counterparts [11]. Thus, an astatinated analogue of MIBG, *meta*-[^{211}At]astatobenzylguanidine (MABG; Figure 1) could be potentially useful for the treatment of micrometastatic neuroblastoma.

3.1. Synthesis of MABG

MABG was prepared by the astatination of TMSBG, the silicon precursor used for the preparation of n.c.a. [^{131}I]MIBG. Astatine-211 was produced by the cyclotron irradiation of natural bismuth metal targets by the ^{209}Bi(α, 2n)^{211}At nuclear reaction. Dry distillation was used to isolate ^{211}At from the target generally in chloroform. The ^{211}At activity from this solution was extracted into a small volume of 0.1 N NaOH and treated with *N*-chlorosuccinimide and the silicon precursor. Unlike radioiodination, a temperature of 50–70°C was necessary for astatination. Under these conditions, more than 85% of radiochemical yields were obtained.

3.2. In vitro *uptake of MABG by SK-N-SH cells: Mechanistic studies*

To investigate whether substitution of ^{211}At for iodine compromised the molecular properties of MIBG, experiments were performed in SK-N-SH human neuroblastoma cells *in vitro* to see whether, like MIBG, MABG is taken up by an active uptake-1 mechanism. For this, the cells (5×10^5 per well per 0.5 ml medium) were preincubated with various uptake-1 inhibitors for 30 min in 24-well plates. The medium was removed, fresh medium containing MABG (2.8 kBq per well) was added and incubated at 37°C for an additional 2 h, and cell-associated activity was determined. In addition, an experiment was done to determine the effect of lower temperature on uptake to see whether tracer accumulation was due to an energy-dependent process. For this, the cells as above were incubated with MABG at 4°C.

Because MIBG is an analogue of the neuronal transmitter norepinephrine, the addition of norepinephrine can inhibit the uptake of MIBG by SK-N-SH cells. Indeed, the uptake of MABG was reduced to 84%, 13% and 4% of control values by 1, 10 and 1000 :M norepinephrine. The tricyclic antidepressant desipramine (DMI) is an inhibitor of the uptake-1 mechanism. When pretreated with 0.1, 0.5 and 1 :M DMI, the uptake of MABG was reduced to 21%, 12% and 11% of control values, respectively. To determine the energy-dependency of MABG uptake by SK-N-SH cells, three conditions were used. When MABG was preincubated with 1.5 mM dithionite, which depletes the oxygen from the medium, the uptake was reduced to 18% of control values. Ouabain (1mM), which inhibits ATPase and hence the uptake-1 mechanism, also reduced MABG uptake to 8% of controls. Finally, incubation at 4°C also resulted in the reduction (to 8%) of MABG uptake. These data clearly demonstrate that MABG, like MIBG, is transported via an active uptake-1 mechanism in SK-N-SH neuroblastoma cells.

3.3. Biodistribution in normal mice

The biological similarity of MIBG and MABG was further investigated by performing tissue distributions in normal mice. As shown in Figure 2, the accumulation of both tracers in normal mouse

tissues was quite similar over a 24 h period. Differences in the uptake in adrenals, one of the target tissues for MIBG were not statistically significant at any time points. The heart uptake of MABG was 80–83% of that seen for n.c.a. [^{131}I]MIBG at 1 and 4h; however, the difference was statistically

significant only at 4 h (13.0 ± 1.6% vs 16.1 ± 2.6%; $p < 0.05$). By 24 h, the value for MABG was about 1.3-fold that for [^{131}I]MIBG, but the difference was not statistically significant. The uptake of MABG in adrenals and heart was reduced to 50% and 33%, respectively, of control values when the mice were pretreated with the uptake-1 inhibitor DMI. This suggests that the accumulation of ^{211}At these tissues was mediated by a specific uptake mechanism. A potential concern with using astatinated radiopharmaceuticals is their *in vivo* instability. Thyroid uptake, an indicator of *in vivo* dehalogenation, was similar for both tracers suggesting a low degree of dehalogenation for both compounds. However, it is important to note that the uptake of astatide in mouse thyroid is only 10–50% that of iodide over the time course of this study [12]. In addition, spleen and lungs are two organs with relative selectivity for astatide about 10 times that seen for iodide. The above results show similar values of uptake for both tracers in these tissues, suggesting that MABG may be reasonably stable *in vivo*.

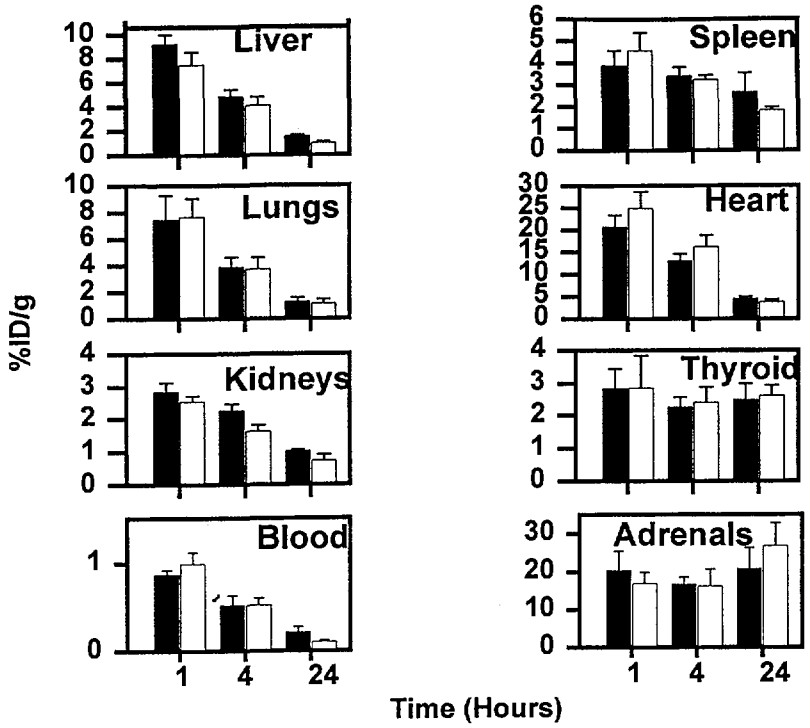

FIG. 2. Tissue distribution of n.c.a. [^{131}I]MIBG and MABG in normal mice.

3.4. Tissue distribution in athymic mouse neuroblastoma xenograft model

The potential usefulness of MABG for targeted radiotherapy was further evaluated by doing a paired-label tissue distribution of MABG and n.c.a. [^{131}I]MIBG in athymic mice bearing SK-N-SH neuroblastoma xenografts. As shown in Figure 3, at 8 h after injection, the tumour uptake of MABG and n.c.a. [^{131}I]MIBG was 3.8 ± 0.8%ID/g and 3.1 ± 0.7%ID/g, respectively, and the difference was statistically significant ($p < 0.05$). At all time points the tumour uptake of MABG was higher than that of n.c.a. [^{131}I]MIBG. Pretreatment of mice with DMI reduced the tumour uptake of MABG by 43%, suggesting that its accumulation was related to a specific uptake-1 mechanism. The uptake of MABG in other tissues was generally higher than that of n.c.a. [^{131}I]MIBG, and the difference in uptake between the two tracers increased with time. Although uptake in thyroid was similar for both tracers, uptake in lung, spleen and stomach was higher for ^{211}At, suggesting that MABG has a greater susceptibility to dehalogenation in this model than seen in normal mice. This, and the probable higher lipophilicity of MABG, may be the reasons why tissue uptake of MABG was generally higher in all tissues.

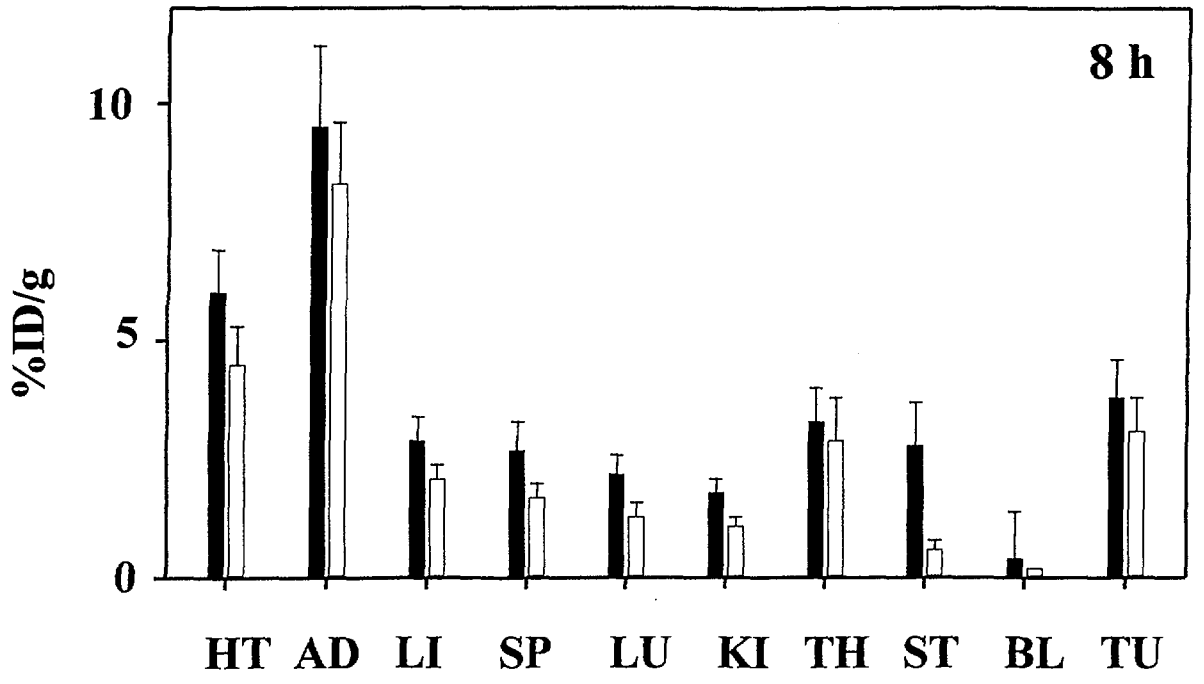

FIG. 3. Paired-label tissue distribution of MABG and n.c.a. [^{131}I]MIBG in athymic mice hosting SK-N-SH xenograft 8 h after injection.

3.5. Cytotoxicity

3.5.1. Inhibition of [^{3}H]thymidine uptake

SK-N-SH cells were initially treated with varying concentrations of ex- [^{131}I]MIBG, n.c.a. [^{131}I]MIBG and MABG for 30 min. After washing, the cells were incubated with thymidine-deficient medium for 24 h. Subsequently, the ability of cells to incorporate thymidine was determined by incubating them with [^{3}H]thymidine for 30 min. The amount of thymidine incorporated was reduced to less than 50% of the control level with as little as 118 Bq of MABG. No significant reduction in thymidine uptake was seen even with 3 kBq of n.c.a. [^{131}I]MIBG. The thymidine uptake was completely impaired with about 370 kBq of n.c.a. [^{131}I]MIBG, whereas at this level more than 50% of thymidine incorporation was seen with ex-[^{131}I]MIBG.

3.5.2. Clonogenic survival

The proliferative capacity of untreated SK-N-SH cells and those treated with MABG, [^{211}At]astatide and n.c.a. [^{131}I]MIBG was determined using a limiting dilution clonogenic assay [13]. The D_0 values, the amount of initial radioactivity concentration necessary to reduce the survival to 37%, were calculated from these data. A D_0 value of 215 Bq/ml was calculated for [^{211}At]MABG. In comparison, the value for n.c.a. [^{131}I]MIBG was 384 kBq/ml implying a more than 1,000-fold higher cytotoxicity for the α-particle emitting analogue. That the exquisite cytotoxicity of [^{211}At]MABG is indeed due to its specific uptake and retention in SK-N-SH cells was demonstrated by the fact that the D_0 for [^{211}At]astatide, 17.8 kBq/ml, was more than 80-fold higher than that for [^{211}At]MABG.

4. 4-FLUORO-3-[^{131}I]IODOBENZYLGUANIDINE

Positron emission tomography (PET) is a superior imaging technique. An MIBG analogue labelled with a positron emitter would be attractive for diagnostic oncology. Towards this goal, we have prepared 4-[^{18}F]fluoro-3-iodobenzylguanidine ([^{18}F]FIBG; Figure 1) [14]. Preliminary results

have indicated that [^{18}F]FIBG is a suitable analogue of MIBG and may find application in the PET imaging of neuroendocrine tumours and the myocardium.

With regard to the oncologic strategy of using PET as a prelude to radionuclide therapy, utilization of radionuclides of the same element for both diagnosis and treatment would be advantageous, particularly if PET is to be used for dosimetry planning. For example, ^{124}I and ^{131}I, while not ideal, may be a useful pair of radionuclides for labeling MIBG or an MIBG analogue such as FIBG with increased retention in neuroblastoma cells. Another strategy is to have a molecule which can be labelled with either the therapeutic or positron-emitting nuclide. FIBG presents such an opportunity since it contains both fluorine and iodine. Because our results with [^{18}F]FIBG suggest that 4-fluoro-3-[^{131}I]iodobenzylguanidine([^{131}I]FIBG; Figure 1) may offer higher binding to neuroblastoma cells than MIBG itself, we developed a no-carrier-added synthesis of [^{131}I]FIBG from a silicon precursor and evaluated its potential usefulness.

4.1. Synthesis of [^{131}I]FIBG

Since n.c.a. [^{131}I]MIBG could be prepared in excellent radiochemical yield from a silicon precursor, we decided to follow the same strategy for the preparation of radioiodinated FIBG. Towards this end, a silicon precursor, 4-fluoro-3-(trimethylsilyl) benzylguanidine (FTMSBG) was prepared in 5 steps. When the conditions used for the conversion of TMSBG to MIBG were applied, only 60–65% of [^{131}I]FIBG was obtained from FTMSBG. A radiolabelled byproduct, in an amount roughly equal to half of [^{131}I]FIBG, was also formed. Its HPLC behaviour and *in vitro* binding to SK-N-SH cells indicate that this compound is MIBG. Radiochemical yields of 75–80% for [^{131}I]FIBG were obtained, however, when FTMSBG was radioiodinated using hydrogen peroxide as the oxidant in aqueous acidic conditions at 50°C.

4.2. In vitro evaluation

When performed in a paired-label format, the specific binding of [^{131}I]FIBG to SK-N-SH cells remained fairly constant (45–60%) over a 2–3-log activity range, and was 11–14% higher ($p < 0.05$) than that of [^{125}I]MIBG. The uptake of [^{131}I]FIBG was blocked to varying degrees by several interventional agents, and by performing the incubation at 4°C. The uptake-1 inhibitor DMI (1.5 :M) reduced the binding of [^{131}I]FIBG to 13% of the control value. Ouabain (1 mM) and incubation at 4°C reduced the uptake to 31% and 8% of the control value, respectively, suggesting that the uptake of [^{131}I]FIBG in this cell line was energy-dependent. The specificity of [^{131}I]FIBG uptake was further demonstrated by the reduction of its binding to 8%, 6% and 5% of the control value by 50 :M norepinephrine, 10:M MIBG and 10 :M FIBG, respectively. These results suggest that uptake of [^{131}I]FIBG by this cell line is specific and is mediated through an active uptake-1 mechanism.

In addition to higher uptake, retention of a radiotherapeutic agent by the tumour for time period compatible with the physical half-life of the radionuclide is important for its efficacy. The ability of SK-N-SH cells to retain [^{131}I]FIBG and [^{125}I]MIBG was determined in a paired-label format. After incubating the cells with both tracers for a period of 2 h, the cells were washed to remove unincorporated activity. Subsequently, cells were incubated with fresh medium without and with desipramine. The cell-associated activity at various intervals was determined. As shown in Table 2, 76% of the originally bound [^{131}I]FIBG activity was retained in SK-N-SH cells after 3 days compared with 30% for [^{125}I]MIBG. Using these binding data, time-activity curves were constructed assuming that both tracers were labelled with ^{131}I. The area under the FIBG time-activity curve extrapolated to infinity was about twice that for MIBG, suggesting a significant advantage in radiation absorbed dose to this cell line might be achievable with [^{131}I]FIBG. Further, it was demonstrated that DMI enhanced the washout of initially bound [^{125}I]MIBG and [^{131}I]FIBG, indicating that tracer retention is mediated by the re-uptake of released activity.

TABLE II. PAIRED-LABEL RETENTION OF [^{131}I]FIBG AND [^{125}I]MIBG BY SK-N-SH CELLS AS A FUNCTION OF TIME.

Time (Hours)	Cell-associated activity (Per cent of Input)			
	[^{131}I]FIBG	[^{131}I]FIBG + DMI	[^{125}I]MIBG	[^{125}I]MIBG + DMI
0	74.1 ± 3.1	74.1 ± 3.1	66.6 ± 3.7	66.6 ± 3.7
2	78.4 ± 0.6	53.0 ± 1.2	67.0 ± 0.4	40.6 ± 1.0
4	77.3 ± 2.3	38.5 ± 3.2	64.2 ± 2.6	25.8 ± 2.2
8	69.8 ± 4.3	27.3 ± 0.9	54.9 ± 3.3	14.5 ± 0.5
24	72.9 ± 2.6	23.5 ± 2.0	45.8 ± 4.6	10.8 ± 0.7
48	64.5 ± 5.0	25.6 ± 0.9	34.9 ± 2.0	12.7 ± 0.4
72	50.6 ± 5.0	29.6 ± 3.1	17.3 ± 2.5	14.9 ± 1.4
96	25.7 ± 5.7	26.7 ± 3.3	7.7 ± 1.3	12.6 ± 1.4

5.1. Biodistribution in normal mice

A paired-label tissue distribution of [^{131}I]FIBG and [^{125}I]MIBG was performed in normal mice over a period of 7 days. High uptake of [^{131}I]FIBG was seen in both heart and adrenals. The [^{131}I]FIBG/[^{125}I]MIBG myocardial uptake ratio increased from about 1 at 1 h to 1.3, 3.8 and 12.2 at 4 h, 1 d and 3 d, respectively. Adrenal uptake was similar for both tracers up to 2 days; however, a 1.4–2-fold higher retention of [^{131}I]FIBG was seen from 3 to 7 d. One hour after injection, the heart and adrenal uptake of [^{131}I]FIBG in DMI-treated mice was reduced to 48% and 60% of the control values, respectively confirming the specificity of uptake in these tissues.

Retention of [^{131}I]FIBG was higher in most other tissues also. Thyroid was a notable exception. For example, at 24 h, the thyroid uptake of ^{131}I (1.9 ± 0.4%ID/g) was half that of ^{125}I (3.7 ± 1.7%ID/g; $p < 0.05$) and this difference increased with time. This suggests that FIBG is less susceptible towards deiodination.

6. CONCLUSIONS

A method has been developed to prepare radioiodinated MIBG at a no-carrier-added level. *In vitro* studies and some *in vivo* studies indicate that n.c.a. [^{131}I]MIBG may be advantageous for clinical applications. It was possible to prepare MABG in high radiochemical yields and it retained the molecular properties of MIBG to a considerable degree. MABG was shown to be extremely cytotoxic and should find applications in the treatment of metastatic neuroblastoma. It has been shown that MIBG is taken up by several medulloblastoma cell lines and that MABG is cytotoxic to some of these as a result of the specific uptake of MABG (data not given). Since neoplastic meningitis, a disease that should be amenable to α-particle therapy, is often associated with medulloblastoma, MABG could be a suitable endoradiotherapeutic agent for this type of cancer. Fluorine substitution in MIBG resulted in a molecule which was retained to a higher degree by the tumour cells. Thus therapeutic efficiency of [^{131}I]FIBG can be anticipated to be higher than that of [^{131}I]MIBG. Hopefully, clinical investigations with some of these more potent MIBG analogues will be initiated in the near future.

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