Sequential Biological Dosimetry after a Single Treatment with Iodine-131 for Differentiated Thyroid Carcinoma

R. M'Kacher, J.-D. Légal, M. Schlumberger, B. Aubert, N. Beron-Gaillard, A. Gaussen and C. Parmentier
Radioprotection Laboratory-Nuclear Medicine and Department of Physics, Institut Gustave Roussy, Villejuif, France

To determine the cytogenetic and genotoxic risk associated with therapeutic exposure to ^{131}I (3.7 GBq) in 50 patients with differentiated thyroid carcinoma, we estimated the dosimetric index that reflects the dose to the circulating lymphocytes on Day 4 and at several time intervals after exposure over a period of 2 yr. **Methods:** Chromosomal aberrations were scored in peripheral lymphocytes obtained before and then 4 days, 3 mo, 6 mo, 1 yr and 2 yr after the first administration of 3.7 GBq ^{131}I according to two methods: conventional cytogenetics and chromosome 4 painting. **Results:** The dosimetric index was 0.52 Gy on Day 4, 0.49 Gy at 3 mo, 0.45 Gy at 6 mo, 0.44 Gy at 1 yr and 0.42 Gy at 2 yr by conventional cytogenetics and 0.47 Gy on Day 4, 0.45 Gy at 3 mo, 0.44 Gy at 6 mo, 0.43 Gy at 1 yr and 0.42 Gy at 2 yr by chromosome 4 painting. We found a decrease in the frequency of chromosomal aberrations between Day 4 and 3 mo after exposure. This may be due to the decrease of lymphocyte counts shortly after ^{131}I administration, which will recover later on. In contrast, the number of anomalies remained constant starting 3 mo after ^{131}I administration. **Conclusion:** These techniques permit retrospective biological dosimetry for up to 2 yr after therapeutic exposure to ^{131}I.

Key Words: biological dosimetry; iodine-131; thyroid cancer

J Nucl Med 1997; 38:377–380

The scoring of chromosomal aberrations in human lymphocytes is a well-established method for personal dosimetry in radiological protection (*1*). The dicentric chromosome is considered the aberration of choice because its production is nearly specific for ionizing radiation, and its natural occurrence is low. However, its unsuitability for measuring a dose received some years prior to the blood sampling is a major drawback. This drawback may now be overcome by scoring stable translocations by fluorescence in situ hybridization (FISH) with whole chromosome probe libraries (*2,3*). The persistence of these radiation-induced translocations may be used for retrospective biological dosimetry (*4,5*).

Chromosomal changes have been described following treatment with ionizing radiation (*6–9*), but up to now, only few quantitative studies have been performed in thyroid cancer patients (*7–9*).

In our recent study (*10*), biological dosimetry based on scoring of chromosomal aberrations in peripheral lymphocytes has been applied to 30 thyroid cancer patients treated with ^{131}I. On Day 4 after the first administration of 3.7 GBq ^{131}I, the mean dosimetric index that reflects the mean dose to the circulating lymphocytes was 0.54 Gy (95% CI; 0.45–0.62 Gy) by conventional cytogenetics and 0.48 Gy (95% CI; 0.42–0.61 Gy) by chromosome 4 painting.

We have extended this study to a larger series of thyroid cancer patients. Furthermore, we have followed these patients for up to 2 yr after the first treatment with ^{131}I to ascertain whether: (a) results of biological dosimetry performed 4 days or several months after therapy with ^{131}I are similar and (b) biological dosimetry based on counting stable and unstable anomalies leads to similar dose estimates over this period of time.

Received Apr. 11, 1996; revision accepted Jul. 5, 1996.
For correspondence or reprints contact: C. Parmentier, PhD, Institut Gustave Roussy, 94805 Villejuif Cedex, France.

METHODS

Patients

The study was performed in 50 patients (8 men, 42 women; age 25–78 yr; mean age 46 yr) treated for differentiated thyroid cancer. All patients had undergone total thyroidectomy. None of these patients had been treated with external radiotherapy or ^{131}I therapy before study. A standard treatment dose of 3.7 GBq ^{131}I was administered 4–5 wk after total thyroidectomy for ablation of thyroid remnants while the patient was hypothyroid. A total-body scan was performed on Day 4 after the treatment. Patients then underwent clinical follow-up and biological evaluation at 3 mo, 1 and 2 yr and total-body scanning performed with 37 to 185 MBq ^{131}I at 6 mo post-treatment.

A blood sample was obtained from each patient before treatment and on Day 4. A blood sample was then drawn at 3 mo in 21 patients, at 6 mo in 23 patients, at 1 yr in 5 patients and at 2 yr in 18 patients after the first administration of 3.7 GBq ^{131}I.

A total-body scan was performed 4 days after administration of 3.7 GBq ^{131}I using a homemade digitized whole-body rectilinear scanner. This scanner measures radioactive uptake in any focus as well as whole-body retention of ^{131}I.

This study was performed in accordance with local ethical rules, and all patients gave informed consent.

Conventional cytogenetics methods were applied to score unstable aberrations (dicentrics, rings and breaks) and chromosome 4 painting (FISH) to score stable aberrations (translocations, insertions and deletions). Two hundred metaphases per blood sample were scored for each method.

Lymphocyte Culture and Chromosome Preparation

Five milliliters of medium were added to 0.5 ml of blood sample and incubated. The medium consisted of 5 ml RPMI 1640 supplemented with 10% fetal calf serum, 0.1 ml phytohemagglutinin M (Gibco, BRL, Grand Island, NY), 1% glutamin, 1 m*M* sodium pyruvate, 1% BrdU and antibiotics (penicillin and streptomycin). Colcemid (0.1 μg/ml) was added 2 hr before harvesting, and slides with chromosomes in metaphase were prepared following the standard methanol/acetic (3/1, v/v) procedure. The slides were stored at −20°C until use (*3*).

Conventional Cytogenetics

The slides were stained by Fluorescent Plus Giemsa coloration. Only complete metaphases (46 centromeres) were scored for dicentrics, rings and breaks under a light microscope (*7*).

Fluorescence In Situ Hybridization

Whole chromosome 4 painting was performed with a specifically labeled FITC probe (Spectrum green, GIBCO) (*5,6*). The

TABLE 1
Percentage of Aberrations before and after Administration of 3.7 GBq of Iodine-131

	Conventional cytogenetics						Chromosome 4 painting						^{131}I	Decr
	0 day	4 days	3 mo	6 mo	1 yr	2 yr	0 day	4 days	3 mo	6 mo	1 yr	2 yr	Ret	lym
C2	1.33	7.2	4.5	4.5	4.09	3.68	2	5	4	4	4	4	3.3	4
C6	0	2.5	2	2	2	1.75	0	3	3	3	3	2.6	1.7	8
C7	0	3	—	3	—	1.75	0	3	—	2.9	—	2.5	6.8	16.9
C13	1.5	5.5	3	3	—	2.5	2	6	4.5	4	—	4	5.9	6
C15	0	2.22	2	2	2	1.75	1	5.5	4	3.5	3.5	3.5	4.3	—
C22	0	3	—	3	3	2.5	0	3	—	2.5	2.5	2.5	1.8	24.1
C35	0	3	3	—	—	3	0	2.5	2.5	—	—	2.5	2.6	7.5
C31	1	4.5	—	—	—	2.22	0	3	—	—	—	2.5	1	20.1
C12	0	3	—	—	—	2	0	3	—	—	—	3	1.5	2.1
C82	0	4	—	—	—	4	0	3	—	—	—	2	0.5	—
K3	0	3	—	—	—	3	0	3	—	—	—	3	1.4	—
K1	0	2	—	—	—	1.5	0	2	—	—	—	2	3.3	—
C42	0	4	3.5	3	—	3	2	5	5	4.5	—	4	—	—
C63	1	4	—	3.5	3	3	0	3	—	3	3	3	2.4	—
C9	1	3.5	3.5	3	—	2.5	1	3	3	2.5	—	2.5	16.4	11.6
C5	0	5	—	2.5	—	—	0	3.5	—	3	—	—	2.7	5.5
C10	0	3.5	3	—	—	—	0	2	2	—	—	—	4.8	6.7
C11	0	3.5	—	3	—	—	0	4	—	3.5	—	—	23.3	10.1
C14	0	3.5	—	2.5	—	—	0	3	—	2.75	—	—	9.1	—
C19	0	5	4	—	—	3	0	4	2.5	—	—	2	7.4	3.5
C25	0.5	3	—	3	—	2	0	3	—	3	—	3	10.4	9
C26	0	3.5	3.33	2.5	—	2	0	3	3	3	—	2.5	6.1	—
C34	0	4	—	2.5	—	—	0	3.5	—	3	—	—	12.9	—
C45	0	3	2.5	2.5	—	—	0	3	2.5	2.5	—	—	8.3	1.4
C58	0	4	—	3	—	—	0	3	—	2	—	—	15.5	—
C64	0	3	—	2	—	—	0	4	—	3.5	—	—	6.2	—
C65	0	4	—	4	—	—	0	3	—	2.5	—	—	14.3	—
C66	0	2.5	—	2	—	—	0	2.5	—	2	—	—	—	—
C55	2	8	—	—	—	—	1	3.04	—	—	—	—	—	8.7
C68	0	4	—	4	—	—	0	4	—	4	—	—	—	0.5
C69	0	3	2	2	—	—	0	2.5	2.7	3	—	—	—	9.2
C57	1	5	—	—	—	—	0	2.7	—	—	—	—	1.1	8
C44	0	2	1.75	—	—	—	0	2	2	—	—	—	1	2.6
C56	0	4	3.5	—	3	—	0	3	2.8	—	2.5	—	3.6	13.7
C70	1.42	4.1	3	2.5	—	—	1.5	4	4.7	4	—	—	11.6	18.1
C67	0	3.5	—	—	—	—	2.3	5	—	—	—	—	7.6	5.6
C33	1.42	4.1	—	—	—	—	1.5	4	—	—	—	—	11.6	18.1
C73	0	5	5.5	—	—	—	1.3	4.3	4	—	—	—	2.3	12.8
C74	0	2.5	—	—	—	—	0	2	—	—	—	—	2.5	6
C75	0	3	2.5	—	—	—	1	3	3	—	—	—	3.7	14.3
C76	0	3	3.5	—	—	—	0	3	3	—	—	—	4.4	17.2
C77	0	2.5	3	—	—	—	1	4	3	—	—	—	0.8	4.1
C78	1	3.5	—	—	—	—	2.5	4	—	—	—	—	4.4	8.1
C79	0	2.5	—	—	—	—	0	2.5	—	—	—	—	7.4	—
C80	1	3	—	—	—	—	0	2.5	—	—	—	—	0.7	7.9
C81	0	2.5	—	—	—	—	0.5	2.6	—	—	—	—	0.5	—
C83	0	4	3.5	—	—	—	2	4.3	4.5	—	—	—	5.3	5.3
K2	0	2.5	—	—	—	—	0	2.5	—	—	—	—	4.1	—
C87	0	3	—	—	—	—	0	3	—	—	—	—	2	7
C88	0	4	4	—	—	—	0	2.5	2.5	—	—	—	7	—
Mean	0.28	3.61	3.17	2.83	2.85	2.50	0.45	3.29	3.25	3.11	3.08	2.84		

Decr lym = decrease of lymphocytes after iodine administration (%); ^{131}I ret = ^{131}I whole-body retention measured 4 days after ^{131}I administration.

slides were analyzed under a fluorescence microscope by visual scoring of translocations, insertions, deletions and breaks.

Statistical Analysis

The dosimetric index was obtained by plotting the number of chromosomal aberrations in peripheral lymphocytes on the dose-effect curve established by ^{131}I in vitro exposure (dose rate ranged from 0.3 10^{-3} to 14.7 10^{-3} cGy/min) of normal lymphocytes (*10*). We compared this curve to that obtained by external irradiation with ^{60}Co (dose rate 0.1 Gy/min). No significant differences were detected in the frequency of aberrations induced by in vitro irradiation, with either ^{60}Co or ^{131}I. In fact, previous studies have shown that the dose response curve is not significantly modified by the type of energy nor by the dose rate of ionizing radiation

TABLE 2
Mean Dosimetric Index at Several Time Intervals after Administration of 3.7 GBq Iodine-131

	Conventional cytogenetics		Chromosome 4 painting	
Time	Dose (Gy)	95% CI (Gy)	Dose (Gy)	95% CI (Gy)
4 days	0.52	0.47–0.55	0.47	0.42–0.51
3 mo	0.49	0.43–0.58	0.45	0.33–0.50
6 mo	0.45	0.41–0.50	0.44	0.34–0.56
1 yr	0.44	0.32–0.60	0.43	0.32–0.61
2 yr	0.42	0.34–0.49	0.42	0.30–0.49

(*12,13*). The reproducibility of this assay was assessed by measuring the dosimetric index in lymphocytes of five healthy donors irradiated with ^{60}Co (dose rate 0.1 Gy/min) at known doses: the coefficient of variation was 14.8% for 0.2 Gy, 12.2% for 0.5 Gy and 4.3% for 1 Gy. Therefore, the reproducibility of the assay system appears to be acceptable even for low doses.

RESULTS

Table 1 shows the frequency of unstable anomalies detected by conventional cytogenetics and stable anomalies detected by chromosome 4 painting at different sampling times before, 4 days, 3 mo and up to 2 yr after treatment with ^{131}I.

The mean dosimetric index of the 50 patients on Day 4 after administration of 3.7 GBq was equal to 0.52 Gy (95% confidence interval (CI): 0.48–0.60 Gy) by conventional cytogenetics and 0.47 Gy (95% CI: 0.42–0.51 Gy) by chromosome 4 painting (Table 2). This is in accordance with our previous data (*10*).

Iodine-131 retention on Day 4, as measured by a total-body scan, ranged from 1% to 23% of the administered activity, and ^{131}I uptake in thyroid remnants and in metastases ranged from 0% to 9% of the administered activity. A close relationship was found between the dosimetric index estimated by both conventional cytogenetics and chromosome 4 painting and total-body retention of ^{131}I on Day 4 after ^{131}I administration. In contrast, no relationship was found on Day 4 between the dosimetric index and ^{131}I uptake in the thyroid remnants and distant metastases.

The follow-up of these patients showed the persistence of all kinds of anomalies with both methods for up to 2 yr (Fig. 1). Table 2 shows the mean dosimetric index at several times after the administration of 3.7 GBq ^{131}I by conventional cytogenetics and chromosome 4 painting. A mean decrease in the frequency of chromosomal aberrations was found between Day 4 and 3 mo after exposure. This may be related to a 1% to 24.1% decrease in lymphocyte counts on Day 4 starting 3 mo after exposure; the number of anomalies remained constant with both methods for up to 2 yr. During this time, the number of reciprocal translocations increased compared to terminal translocations.

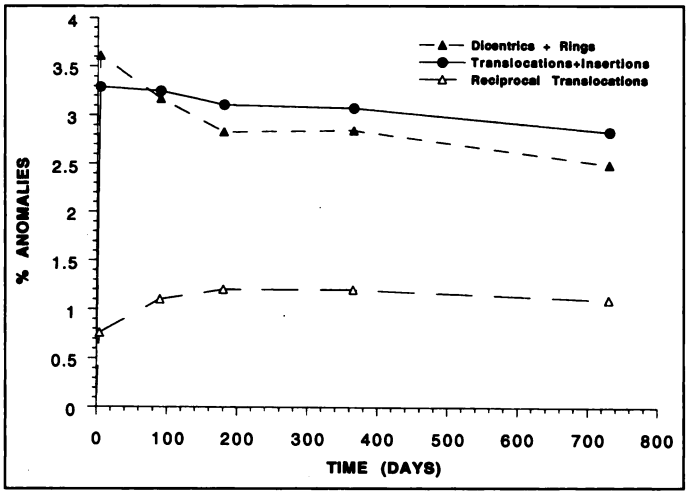

FIGURE 1. Variation of unstable and stable anomalies with time.

DISCUSSION

Physical dosimetry estimates of the absorbed dose by circulating lymphocytes are similar for the blood and bone marrow. As a result, the dosimetric index of the circulating lymphocytes reflects the dose to the bone marrow (*14*).

The present study confirms our previous report on a larger series of patients (*10*):

1. The dosimetric index at Day 4 is 2–4 times higher than that derived from mathematical models in euthyroid subjects (*14*).
2. It is closely related to whole-body retention of ^{131}I at Day 4.
3. Results of both methods are in close agreement.

Furthermore, this study shows that chromosomal anomalies induced by radiation persist over long periods of time (2 yr) and that retrospective biological dosimetry can be established in patients exposed to ionizing radiations.

According to the literature, a marked decrease in dicentric chromosomes should occur with time (*15*) because these anomalies are lethal. However, translocations should persist with time (*5*). Our results demonstrate the persistence of dicentric chromosomes for up to 2 yr, which is in agreement with the average life of lymphocytes which is 3 yr. Thus, both methods are suitable for retrospective dosimetry during this time frame. In fact, both methods provided similar results at each time point. Furthermore, dose estimates did not vary with time and their apparent decrease at 3 mo may be attributed to the decrease in lymphocytes after the administration of ^{131}I and to lymphocytes from less irradiated regions that subsequently enter the circulation (*16*). This decrease is proportional to the mean absorbed body dose (*17*). To make up for the decrease in lymphocyte counts after administration of ^{131}I, either the stem cells may accelerate their mitotic activity, or lymphocytes from other organs such as the spleen, which are less exposed to irradiation, may migrate into the bloodstream. This lowers the frequency of chromosomal anomalies found in lymphocytes (*18*).

Blood samples taken at 3 and 6 mo and 1 and 2 yr after administration of ^{131}I demonstrated more reciprocal translocations than those taken at 4 days. Stem cells produce new lymphocytes to make up for the loss after ^{131}I treatment (Table 1). These new lymphocytes will carry the reciprocal translocations induced by irradiation in the stem cells, and the terminal translocations, which are less viable, will not be present in the blood samples beyond 4 days. These methods appear more suitable for retrospective dosimetry than scoring of micronuclei, which are eliminated after 1 yr (*19,20*).

CONCLUSION

The present study needs to be extended to larger time intervals after exposure to confirm the reliability of chromosome painting for the detection of past exposures and to define the limits of its resolving power. The spectrum of aberrations detected with chromosome painting after long intervals from radiation exposure favors the use of this analysis (*21*).

ACKNOWLEDGMENTS

This work was supported by a grant from the Institut Gustave Roussy (CRC # 95-25), Electricité de France (EDF # 1H1794) and Institut de Protection et de Sûreté Nucléaire (IPSN # 40105B).

We are grateful to the nurses, technologists and secretaries of the nuclear medicine department for their support and participation in this study. We thank A. Bernheim and A. Duverger for their help in reading the slides and N. Foray for the statistical analysis of the results. We are indebted to I. Kuchenthal for preparing the manuscript.

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Autopsy of a Cadaver Containing Strontium-89-Chloride

Frank V. Schraml, Laurence F. Parr, Sawson Ghurani and Eugene D. Silverman

Nuclear Medicine Division and Anatomic Pathology, National Naval Medical Center, Bethesda, Maryland; and Radiology and Nuclear Medicine Department, Uniformed Services University of the Health Sciences, Bethesda, Maryland

An autopsy was performed on a patient who died after receiving ^{89}Sr-chloride for treatment of bone pain from metastatic prostate carcinoma. Coordination between nuclear medicine physicians, radiation safety division personnel and pathologists resulted in minimal radiation exposure and the acquisition of dosimetry data.

Key Words: strontium-89-chloride; metastatic carcinoma; autopsy; biodistribution studies

J Nucl Med 1997; 38:380–382

Strontium-89-chloride has proven efficacy in the treatment of bone pain associated with metastatic prostate (*1,2*) and metastatic breast carcinoma (*1–3*). Patients who receive ^{89}Sr therapy have advanced metastatic disease, and many are markedly debilitated. Although ^{89}Sr-chloride is generally reserved for patients who have a life expectancy of at least a few months, occasional deaths may occur soon after treatment.

Received Apr. 8, 1996; revision accepted Jul. 31, 1996.

For correspondence contact: Frank V. Schraml, MD, Division of Nuclear Medicine, Radiology Department, National Naval Medical Center, Bethesda, MD 20889-5600.

CASE REPORT

An 83-yr-old man with Stage C prostate carcinoma and pain associated with widespread bony metastases [confirmed by bone scan (Fig. 1)] was referred to the nuclear medicine division for ^{89}Sr-chloride therapy. The patient had no other known significant medical problems. He was treated with 162.1 MBq (4.38 mCi) ^{89}Sr-chloride [$T_{1/2}$], 50.5 days, beta Emax 1.463 MeV(100%)], intravenously, given over 2 min. Because of difficulty adequately managing the patient's pain at home, prior arrangements had been made for the patient's admission to the hospital after administration of strontium. The patient was therefore admitted to the hospital, in stable condition, after his discharge from the nuclear medicine clinic. He died approximately 4 days later.

The body was not moved until the radiation safety staff arrived and performed appropriate safety surveys with a thin window "pancake" Geiger-Mueller detector to determine levels of contamination. The body was then tagged, wrapped in bed linens, transported to the morgue using universal precautions and autopsied.

Two pathologists and a technologist conducted the autopsy 1 day after the patient's death (i.e., 5 days after ^{89}Sr treatment). Before proceeding, each member of the pathology staff donned two pairs of standard latex surgical gloves, a standard polyester surgical