A study of professional radiation hazards in CT scan and nuclear medicine workers using GTG-banding and solid stain

Vahid Changizi¹, Mohammad Hossein Alizadeh², Akbar Mousavi³

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Abstract
Background: CT scan and nuclear medicine exams deliver a great part of medical exposures. This study examined professional radiation hazards in CT scan and nuclear medicine workers.

Methods: In a cross sectional study 30 occupationally exposed workers and 7 controls (all from personnel of a laboratory) were selected. Physical dosimetry was performed for exposed workers. Blood samples were obtained from the experimental and control groups. Three culture mediums for each one were prepared in due to routine chromosome analysis using G-banding and solid stain.

Results: There were significant increased incidence of chromatid gap (ctg) and chromatid break (ctb) with mean±SD frequencies of 3±0.84 and 3.1±1.40 per 100 cells respectively in the nuclear medicine workers versus controls with mean±SD frequencies of 1.9±0.69 and 1.3±0.84 for ctg and ctb, respectively. Chromosome gaps (chrg) were higher significantly in the nuclear medicine population (2.47±0.91) than in controls (1.4±0.9) (p<0.05). In CT scan group the ctg and ctb were increased with a mean±SD frequency of 2.7±0.79 and 2.6±0.91 per 100 cells respectively compared with control group. The mean±SD frequencies of the chrb were 2.0±0.75 and 0.86±0.690 per 100 cells for exposed workers and control group, respectively.

Conclusion: This study showed chromosome aberrations in peripheral lymphocytes using solid stain method are reasonable biomarker reflecting personnel radiation damage.

Keywords: CT scan, Nuclear physics, Chromosome aberration, G-banding, Solid stain.


Introduction
Humans are being exposed by natural ionizing radiation sources such as soil, air, building materials, cosmic irradiation and even body elements. Besides, anybody may have artificial irradiation experiences.¹ United Nations Scientific Committee on the Effects of Atomic Radiation (UNSCEAR) has reported that mine workers, medical personnel in radiology, radiotherapy and nuclear medicine fields are highly at risk of receiving low doses of ionizing irradiations in long terms (2,3).

Nowadays artificial rays have wide uses for medical diagnostic and therapeutic processes. Therefore they are the most important factors in society exposures (4,5). Ionizing radiations are power clastogen causing phosphodiester break in DNA (6). The other important side effects of absorbing ionizing irradiation is double strand break (DSB) forming chromosome aberrations (CAs). CAs are important biomarkers for biological dosimetry. People with increase of CAs in peripheral blood lymphocytes (PBLs) are at high risks for cancer (7,8).

Computed tomography scan (CT scan) and nuclear medicine exams deliver a great part of medical exposures. Nuclear medi-

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cine uses radioisotopes for diagnostic and therapeutic cases through oral or injection. Radioisotopes decay and their irradiation expose to patient, his family and nuclear medicine personnel. It is low doses in diagnostic exams. Personnel are being exposed during transport of drug, injection to patient, and generator milking for many years (9). Professional radiation workers are under long term exposures. In radiology ward multislice computed tomography technique has a wide spectrum of uses. However it uses high radiation doses. International Commission on Radiologic Protection (ICRP) has reported organ doses in CT scan. It is close to that of atomic bombardment remainders (10).

According to above explanations we decided to study chromosome aberrations and effective dose in CT scan among nuclear medicine personnel. Besides, we compared chromosome response to clastogen medium between these exposed personnel and ordinary people.

Methods

Subjects

In a cross-sectional study, occupationally exposed workers were 30 technicians of CT scan and nuclear medicine (all personnel and 7 individuals from the hospital administrative staffs with no exposure history as the control group.

This study was confirmed by the University Committee of Ethics. Every person was informed of the study aims and procedures and written consent form was obtained from all participants. All of them were in healthy conditions at the time of exam.

Occupationally exposed workers were classified in two groups of CT scan and nuclear medicine. CT scan group included 15 people (10 (67%) men and 5 (33%) women) with mean±SD age of 37±7.34 years (range: 24-49 years). Also nuclear medicine group included 15 people with mean±SD age of 35.4±9.24 years (range: 22-54 years). In this study only diagnostic nuclear medicine workers were considered. Table 1 shows characteristics of participants.

The mean±SD work experiences for CT scan and nuclear medicine groups were 12.4±7.08 (range: 1-15 years) and 6.6±5.13 years (range: 1-15 years), respectively. All of them were working 44 hours/week.

Physical dosimetry

Physical dosimetry was done for every one by film badge. Total dose was acquired for every two months and for 6 periods before blood sampling. With this dosimetry total effective dose, Hp (10) and skin dose, Hp (0.07) are assessable.

Blood sampling

After the last period of dosimetry, blood sampling was performed in two steps under sterile conditions. In each step 5 ml peripheral blood was obtained by 10ml syringe and maintained in heparin tube (mediplus lithium heparin). Sampling was performed for experimental and control groups at the

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Exposed subject</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of individual</td>
<td>15</td>
<td>15</td>
</tr>
<tr>
<td>Age (year)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(Mean ± SD)</td>
<td>35.4±9.24</td>
<td>37±7.34</td>
</tr>
<tr>
<td>(Range)</td>
<td>(22-54)</td>
<td>(24-49)</td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male %</td>
<td>60%</td>
<td>36.7</td>
</tr>
<tr>
<td>Female%</td>
<td>40%</td>
<td>33.8</td>
</tr>
<tr>
<td>Smoking status</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-smokers</td>
<td>3</td>
<td>15</td>
</tr>
<tr>
<td>Smokers</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Duration of exposure (year)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(Mean ± SD)</td>
<td>6.6±5.13</td>
<td>12.4±7.08</td>
</tr>
<tr>
<td>(Range)</td>
<td>(1-15)</td>
<td>(1-26)</td>
</tr>
<tr>
<td>Family history of cancer</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>
same time. All samples were coded randomly and under reasonable thermal condition of 4°C-20°C up 10 hours were transferred to the cell culture laboratory by coolant pack for instant cell culture.

**Lymphocytes culture**

Three culture mediums for each one were prepared as follows:

1. For routine chromosome analysis using G-banding and solid stain
2. For chromosome breakage study using mitomycin-c (mmc)
3. Storage culture medium

Whole blood cultures were created by adding 0.5ml heparinized whole blood into 4ml of RPMI 1640 medium (Gibco company production), supplemented with 1 phytohemaglutinin (PHA), 20 fetal bovine serum (FBS), antibiotics (penicillin100/ml and streptomycin 100µg/ml). All cultures were incubated in a CO₂ incubator.

To arrest dividing lymphocytes in metaphase, colchicine at a concentration of 0.5mg/ml, 2 hours prior harvest was added to the culture. After 48 hours incubation the culture were centrifuged at 1000rpm for 10 min. The supernant was removed. Remaining cells were resuspended in a hypotonic solution of KCl (0.075), incubated for 20 minute at 37℃ incubator. In next step centrifugation was performed for 5min at 1000 rpm and the cell were fixed with three exchanges of solution of a fresh mixture of methanol: acetic acid (3:1). Similar to without mitomycin-c approach centrifugation and resuspension were carried out three times and then the cells transferred onto wet, cold slides. The slides were coded and staining was performed by 5% Giemsa (Sigma).

The frequency of CA₅ in the lymphocytes of the exposed and control groups and that of two exposed groups were compared using Fisher’s exact test. The influence of age, work experience with radiation and annually effective dose on the CA₅ were tested by Pearson correlation. The influence of sex was tested by the independent test. Statistical evaluations were done by Fisher’s exact test using SPSS v.21 software.

**Results**

Demographic characteristics of control and exposed personnel are listed in Table 1. These groups did not significantly differ in age, sex and smoking habits (p> 0.05). Family history of cancer was not reported in three studied groups.

Mean±SD of occupationally annual effective doses for nuclear medicine and CT scan groups were 1.4±2.3 mSv (range: 0.05-8.99 mSv) and 0.007±0.01 mSv (range: 0-0.06 mSv) respectively. This value for nuclear medicine workers was significantly higher than that for CT scan workers (p< 0.05). Mean frequencies of chromosome aberrations for three populations (with and without mitomycin-c) are shown in Tables 2 and 3. Nuclear medicine population had the highest CA₅ among the three groups.

There were significant increased incidence of chromatid gap (chg) and chromo-
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Table 2. Frequency of aberrant cells without mmc in lymphocytes of exposed workers and controls

<table>
<thead>
<tr>
<th>Group</th>
<th>Sample size</th>
<th>No. of cell scored</th>
<th>Total aberrant cell</th>
<th>Chromatid gap N (Mean ± S.D)</th>
<th>Chromatid breaks (Mean ± S.D)</th>
<th>Chromosome Gaps N (Mean ± S.D)</th>
<th>Chromosome breaks N (Mean ± S.D)</th>
<th>Fragments N (Mean ± S.D)</th>
<th>Rearrangements N (Mean ± S.D)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>7</td>
<td>700</td>
<td>37</td>
<td>(1.86±0.69)</td>
<td>(1.29±0.84)</td>
<td>(1.4±0.90)</td>
<td>(0.86±0.69)</td>
<td>(0.29±0.48)</td>
<td>0</td>
</tr>
<tr>
<td>Nuclear medicine</td>
<td>15</td>
<td>1500</td>
<td>169</td>
<td>(3±0.84)</td>
<td>(3.13±1.40)</td>
<td>(2.47±0.91)</td>
<td>(2.20±0.86)</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Ct scan</td>
<td>15</td>
<td>1500</td>
<td>146</td>
<td>(2.73±0.79)</td>
<td>(2.6±0.79)</td>
<td>(2.07±1.03)</td>
<td>(2±0.75)</td>
<td>(0.2±0.56)</td>
<td>1</td>
</tr>
</tbody>
</table>

Table 3. Frequency of aberrant cells with mmc in lymphocytes of exposed workers and controls

<table>
<thead>
<tr>
<th>Group</th>
<th>Sample size</th>
<th>No. of cell scored</th>
<th>Total aberrant cell</th>
<th>Chromatid gap N (Mean ± S.D)</th>
<th>Chromatid breaks N (Mean ± S.D)</th>
<th>Chromosome Gaps N (Mean ± S.D)</th>
<th>Chromosome breaks N (Mean ± S.D)</th>
<th>Fragments N (Mean ± S.D)</th>
<th>Rearrangements N (Mean ± S.D)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>7</td>
<td>700</td>
<td>273</td>
<td>(11.5±0.97)</td>
<td>(11±1.4)</td>
<td>(7.7±1.7)</td>
<td>(7.7±0.75)</td>
<td>(0.71±0.75)</td>
<td>(0.29±0.48)</td>
</tr>
<tr>
<td>Nuclear medicine</td>
<td>15</td>
<td>1500</td>
<td>683</td>
<td>(13.8±2.8)</td>
<td>(12±3.04)</td>
<td>(8.4±1.9)</td>
<td>(9.6±1.6)</td>
<td>(1±1.1)</td>
<td>(0.4±0.5)</td>
</tr>
<tr>
<td>Ct scan</td>
<td>15</td>
<td>1500</td>
<td>630</td>
<td>(13.47±1.76)</td>
<td>(11±1.23)</td>
<td>(7.9±2.5)</td>
<td>(9.1±1.8)</td>
<td>(0.87±1.06)</td>
<td>(0.07±0.45)</td>
</tr>
</tbody>
</table>

Chromatid break (chb) with mean±SD frequencies of 3±0.84 per 100 cells and 3.1±1.40 per 100 cells respectively in the nuclear medicine workers versus controls with a mean±SD frequency of 1.86±0.69 and 1.29±0.84 for chg and chb respectively. P-values for these comparisons were 0.05 and 0.06 respectively. Chromosome gap (chrg) and chromosome breaks (chrb) were higher in the nuclear medicine group (2.47±0.91 and 2.2±0.86 respectively) than in controls (1.4±0.9 and 2.2±0.86 respectively). These increases were significant for chrg (p<0.05) and on the borderline for chrb (p=0.08). In this group fragment (F) and rearrangement (r) were greater than those in controls with mean±SD frequencies of 0.4±0.7 and 0.13±0.35 versus 0.29±0.48 and 0 respectively. However these increases were not significant (p=0.60 and p=0.99 respectively).

In CT scan population the increased incidence of the chg and the chb were found with a mean±SD frequency of 2.73±0.79 and 2.6±0.91 per 100 cells respectively, while controls had 1.86±0.69 and 1.29±0.48 per 100 cells for chg and chb. The increase was significant for chg (p<0.05) and not significant for chb (p=0.10). Also there was no significant increase for the chrg (p=0.10); but the increased incidence of the chrb was significant (p<0.05) with a mean±SD frequency of 2±0.75 per 100 cells versus 0.86±0.69 per 100 cells for controls.

There were no significant increases for fragments (F) and rearrangements (r) (p=0.10, p=0.40, respectively) for CT scan workers versus controls.

All types of the chromosome and the chromatid aberrations were higher in the nuclear medicine workers than in the CT scan workers. However they were not significant (Tables 2 and 3).

In breakage study the effect of mitomycin-c as a clastogen medium was studied on chromosome and chromatid aberrations. The results revealed no significant changes in spite of a little increase in frequency of chromosome and chromatid aberrations in the exposed personnel versus the controls (Table 3) (p>0.05). Figure 1 shows chg and chb with solid stain technique.

Discussion

In the present study, G-banding and solid stain assays were used to evaluate chromosome aberrations in peripheral lymphocytes of 30 occupationally exposed workers in nuclear medicine and CT scan wards compared with 7 individuals as the control group. Also annually effective dose was...
evaluated in these personnel using film badge. The measured doses were considerably below the permitted levels (20mSv/year). Our study revealed significantly higher chromosome aberrations in nuclear medicine exposed individuals compared with CT scan workers and controls. Nuclear medicine workers milk the generator, inject radionuclides to the patients and take medical images. These activities take a long time and obviously make long term exposures.

Chromatid type damages were found higher than chromosome type aberration in the exposed workers and the control group. It is in line with the results reported by Garaj-Vrhorac et al. and Hagelstrom et al. It could be due to the low ionizing radiation doses chronically received. However lifestyle and environmental factors are also effective parameters in this subject (9,1).

This study showed compared to other damages, fragments and rearrangements (ring, dicentric) had lower incidence in the exposed workers. It is in agreement with the results reported by Francis Maffi et al. Nuclear medicine and CT scan personnel are exposed by low level of chronic X and γ rays leading mostly single strand break. Chromosome translocations need double strand breaks. It could be due to higher level of ionizing radiations. Besides ring and dicentric chromosome damages are unstable and shown as a consequence of an in vitro or an acute in vivo ionizing irradiation. Translocations are stable chromosome aberrations to be accumulated in cells increasing risk of cancers. G-banding did not show any translocation. However it is suggested to do fluorescent in situ hybridization (FISH) for making sure.

We could not find correlation between age and chromosome aberrations. It is in agreement with some earlier studies (19,20). However some other studies reported contradictory results (21,22). Instability of chromosome damages could hide the age effect.

There was no significant correlation between chromosome damage and gender. It is similar to the results reported by Sari-Minodier et al and Francesca Maffei et al (5,17), and contrary to the results reported by Zakeri et al and Amerunnisa et al (3).
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Conflict of interests
The authors have no conflicts to disclose.

References
1. Health Risks from Exposure to Low Levels of Ionizing Radiation: BEIR VII – Phase 2 (Free Executive Summary)

Acknowledgments
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History of work experience had no correlation with chromosome damages. It is in agreement with the cytogenetic evaluation on occupationally exposed individuals reported by Monika et al (4). It could be due to instability of chromosome damage and considering radiation safety guidelines and using protection.

There was no positive correlation between the effective dose and the redundancy of chromosome aberration in low dose radiation. Some previous studies have revealed these results. Also some studies found, it was difficult to understand a dose-effect relationship for low doses of radiation (23,24). This study did not find any correlation between smoking habit and chromosome aberrations.

In this study the culture medium was treated by clastogenic factor of myotocin-c. This factor is used for Fanconi’s anemia diagnosis. We interested to find if it could be a radiosensitizer or not. We found myotocin-c had no radiosensitizer effect.

Conclusion
This study showed that chromosome aberrations in peripheral lymphocytes using solid stain method are reasonable biomarker reflecting personnel radiation damage. Despite the annual effective doses among CT scan and nuclear medicine workers were significantly below the maximum annual occupational dose limit (20mSv/year); their chromosome aberrations redundancies in peripheral lymphocytes were higher than the controls. Therefore cancer risks from low doses of ionizing radiations in those exposed workers are higher than the normal populations. However the issue is complicated due to individuals’ differences in radiosensitivity, general state of the health, nutrition, habit and life style.

Acknowledgments
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