Short Communication

Gross Morphological Changes of the Mouse Brain Exposed Prenatally to Ionizing Radiation

TAKERU MINAMISAWA, KOUICHI HIROKAGA AND SHUNSAKU SASAKI*

Department of Biology, Yamanashi Medical College, Tamaho, Nakakoma, Yamanashi-ken, 409-38
*Division of Physiology and Pathology, National Institute of Radiological Sciences, Chiba, 260
(Received December 27, 1989)
(Revised version, accepted February 23, 1990)

Prenatal irradiation/Mouse/Brain/Morphology

Gross morphological changes of the brains of first generation (F₁) C57BL/6 × C3H hybrid male mice irradiated prenatally were studied at 6 months of age. Gravid female mice were irradiated with 1.0 Gy, 2.0 Gy or 3.0 Gy of gamma radiation on the 14th day of gestation. The brain in dorsal view was photographed. The measurements of the area and the distance of parts of the brain were made from photographs, using a computer system for biological image analysis. The body and brain weight and the area and length of the cerebral hemispheres (CHs) showed dose-related decrease. There was a dose related increase in the area of the superior colliculi (SC), the distance (DP) from the point which the medial walls of the CHs begin to slope away from the midline to the posterior end of CHs, and the proportion of the DP to the CH length. From the results of the dose-response curve, it appears that the area of the SC and the proportion of DP to CH length are affected by small doses of less than 1.0 Gy.

INTRODUCTION

Recently there has been considerable increasing interest in the effects of prenatal exposure to low levels of ionizing radiation on central nervous system morphology and physiology¹,². Much of the evidence has been concerned with fine structural changes, but there are few quantitative studies of gross morphological alterations. The cerebral hemispheres (CHs) of rats irradiated with X-rays during fetal life were smaller than normal³. The development of computer imaging techniques provides a powerful tool for the study of bio-medicine. Using such a technique, we have studied gross morphological alterations of the brains of mice exposed prenatally to ionizing radiation. In this short report we evaluated the dose related consequence of moderate gamma irradiation of less than 3.0 Gy. The 14th day of gestation was chosen for irradiation since this time has been reported to be in the period when behavior is altered⁴.

南澤 武, 広利浩一：山梨医科大学生物響教室, 山梨県中巨摩郡玉穂町下河東1110 〒409－38
佐々木俊作：放射線医学総合研究所生理病理研究部, 千葉市穴川4－9－1 〒260

NII-Electronic Library Service
ANIMALS AND METHODS

Three experimental groups and one control group of first generation (F₁) C57BL/6 × C3H hybrid male mice were used for this study. All the mice were bred and reared under Specific Pathogen Free (SPF) conditions. The C57BL/6 females, 10–14 weeks old, were caged nightly

![Image of brains](image)

**Fig. 1.** The fixed brains of two adult mice, C and I, that were irradiated with 0 Gy (control) and 3.0 Gy on the 14th day of gestation. The millimeter rule was photographed with the brains.

![Diagram of mouse brain](image)

**Fig. 2.** Schematic outline of mouse brain in dorsal view, with method of measuring areas of olfactory bulbs (OBs), cerebral hemispheres (CHs), superior colliculi (SC), inferior colliculi (IC), the length of cerebral hemispheres and the distance from the point where the medial walls of the cerebral hemispheres begin to slope away from the midline to the posterior end of the cerebral hemispheres (DP).
with C3H males of the same age. The presence of sperm in the vagina the next morning was taken as presumptive evidence of pregnancy, and the day on which this was determined was designated as 0 day of gestation. The pregnant mice were irradiated with 1.0, 2.0 and 3.0 Gy of $^{137}$Cs gamma rays on the 14th day of gestation. The offspring were fostered by normal mothers. All offspring were weaned at four weeks of age, and separated according to sex. The mice were housed individually and were kept for periods of from 2 to 6 months of age. The ambient regulated temperature was $22 \pm 2^\circ$C. A 12 h day-night cycle was maintained with light on at 8:00 a.m. and off at 8:00 p.m. each day. Food and water were given ad libitum.

The mice, at 6 months of age, were anaesthetized with intraperitoneal injections of pentobarbital sodium. The body weight was determined. The mice were decapitated, and the brains were removed immediately and placed in 10% neutral formalin. The brain, separated from the spinal cord near the cervico-medullary junction, was weighed several days after decapitation. The brain in dorsal view was photographed (Fig. 1).

The areas of the olfactory bulbs (OBs), CHs, superior colliculi (SC) and inferior colliculi (IC) were measured. The CH length and the distance (DP) from the point where the medial walls of CHs begin to slope away from the midline to the posterior end of CHs were also measured (Fig. 2). Area and distance measurements were made from photographs, using a computer system for biological image analysis (NEC 9801VM and EPSON GT-4000).

RESULTS AND DISCUSSION

Table 1 shows the body and brain weight, the areas of the OBs, CHs, SC and IC, CH length, and the DP of the 3 irradiated and control groups. The irradiated groups showed significantly lower body weight than the control group. The decrease in body weight was dose related. The control group had greater brain weight than the irradiated groups. The brain weight also showed the decrease related to irradiation dose. The mice that received the greatest exposure were about 40% lighter than the controls. The studies of Rugh et al. in the monkey, of Sikov et al. in the rat, and of Minamisawa and Sasaki in the mouse reported similar results.

The area of CHs of the irradiated groups was significantly smaller than that of the control group. The area was decreased in a dose related way. Changes in the area of OBs were similar to those of CHs, but the difference between the 1.0 Gy and control groups was not significant. The area of SC was significantly greater in the irradiated groups than in the control group. There was a dose related increase in the area of SC of irradiated groups. Changes in the area of IC of the irradiated groups resembled those of SC, but the differences were not significant in the 1.0 Gy and 2.0 Gy groups. Similar qualitative results have been reported by Cowen and Geller and D'Amato and Hicks for the CHs and OBs of rats irradiated with 1.5 Gy or 2.5 Gy on the 14th to 17th day of gestation.

Irradiated groups showed that the medial walls of CHs sloped away from the midline, especially posteriorly, where the midbrain was exposed. The CH length of the irradiated groups was significantly shorter than that of the control group. The decrease in the CH length was dose related. The cerebral hemispheres of the rat irradiated with 2.0 Gy on the 17th day of gestation
PRENATALLY IRRADIATED MOUSE BRAIN

Table 1. The body and brain weight, the area of the OBs, CHs, SC and IC, the CH length and the DP of the 3 irradiated and control groups.

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>1.0 Gy</th>
<th>2.0 Gy</th>
<th>3.0 Gy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of litters</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>Litter size (male)</td>
<td>3.7±0.6</td>
<td>3.7±0.6</td>
<td>3.0±1.0</td>
<td>2.8±1.0</td>
</tr>
<tr>
<td>Number of animals</td>
<td>9</td>
<td>8</td>
<td>7</td>
<td>9</td>
</tr>
<tr>
<td>Body weight (g)</td>
<td>40.4±1.2</td>
<td>37.9±1.4*</td>
<td>32.3±2.5*</td>
<td>25.0±1.4*</td>
</tr>
<tr>
<td>Brain weight (mg)</td>
<td>495.2±19.4</td>
<td>449.9±19.2*</td>
<td>373.3±12.3*</td>
<td>315.3±21.9*</td>
</tr>
<tr>
<td>OB area (mm²)</td>
<td>11.5±1.4</td>
<td>11.2±0.6</td>
<td>8.2±1.1*</td>
<td>7.1±1.0*</td>
</tr>
<tr>
<td>CH area (mm²)</td>
<td>73.6±5.0</td>
<td>62.6±2.5*</td>
<td>52.8±2.4*</td>
<td>45.6±2.9*</td>
</tr>
<tr>
<td>SC area (mm²)</td>
<td>2.6±0.5</td>
<td>3.8±0.4*</td>
<td>4.7±0.6*</td>
<td>5.4±0.7*</td>
</tr>
<tr>
<td>IC area (mm²)</td>
<td>3.2±0.5</td>
<td>3.6±0.2</td>
<td>3.6±0.3</td>
<td>4.4±0.8*</td>
</tr>
<tr>
<td>CH length (mm)</td>
<td>8.9±0.3</td>
<td>8.2±0.2*</td>
<td>7.6±0.2*</td>
<td>7.2±0.3*</td>
</tr>
<tr>
<td>DP (mm)</td>
<td>1.9±0.3</td>
<td>2.2±0.1*</td>
<td>2.3±0.2*</td>
<td>2.5±0.3*</td>
</tr>
<tr>
<td>DP/CH (%)</td>
<td>20.9±3.1</td>
<td>26.6±1.8*</td>
<td>30.7±2.9*</td>
<td>34.4±4.1*</td>
</tr>
</tbody>
</table>

All values are shown as an average ± standard deviations.
Significant differences from control: *p<0.05.
See Fig. 2 for abbreviations.

were reduced by 26% in length in comparison to the controls\textsuperscript{3). In the present study, the irradiated groups had significantly longer DP than did the control group. The DP was increased as a function of dose delivered. The proportion of DP to CH length was significantly higher in the irradiated groups than in the controls, and was proportional to dose increase. The qualitative studies of Hicks et al.\textsuperscript{10} and D’Amato\textsuperscript{11} reported that the CHs in the rat irradiated with 1.5 Gy or 2.0 Gy on days 12 to 18 of gestation, sloped laterally from the midline, especially posteriorly, leaving the colliculi exposed.

The results reported here indicate that body and brain weight, area of CHs and SC, and proportion of DP to CH length are more sensitive indicators of the effects of prenatal exposure to ionizing radiation than the area of OBs and IC. Examination of the dose-response curve suggests that area of SC and proportion of DP to CH length are affected by small doses of less than 1.0 Gy.

ACKNOWLEDGMENTS

We wish to thank Prof. T. Nakazawa for his helpful comments, and Prof. A. Hyman for his careful review of the manuscript. This work was supported in part by a grant for a research project (1988) at Yamanashi Medical College.
REFERENCES