bjh short report

Longitudinal changes in red blood cell distribution width decades after radiation exposure in atomic-bomb survivors

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Summary

Red blood cell distribution width (RDW), which generally increases with age, is a risk marker for morbidity and mortality in various diseases. We investigated the association between elevated RDW and prior radiation exposure by examining longitudinal RDW changes in 4204 atomic-bomb survivors over 15 years. A positive association was found between RDW and radiation dose, wherein RDW increased by 0·18%/Gy. This radiation-associated effect increased as the participants aged. Elevated RDW was also associated with higher all-cause mortality. The biological mechanisms underlying these observed associations merit further investigation.

Introduction

Red blood cell distribution width (RDW) is an index of the heterogeneity of circulating erythrocyte size. It gradually increases with age and is remarkably elevated in several types of anaemia.[1] Recently, RDW has been established as a strong predictor of prognosis and mortality due to various diseases, including cancer and cardiovascular disease.[1] In a meta-analysis using the data of seven population-based studies, all-cause mortality risk increased by 14% for every 1% increment in RDW.[2] The association between RDW and mortality remained or became stronger after exclusion of subjects with anaemia or other nutritional conditions that could potentially affect red blood cells (RBCs).[2] The magnitude and robustness of the association between RDW and mortality indicate that RDW is an integrative biomarker of several pathophysiological processes associated with ageing, particularly those involving bone marrow dysfunction and disturbed erythropoiesis.[3] Radiation exposure adversely impacts haematopoiesis, particularly by affecting erythroid progenitor cells.[4] The evaluation of RDW, an inexpensive and widely available marker, may therefore contribute to the assessment of radiation health risks.

In atomic-bomb (A-bomb) survivors whose mortality is associated with radiation dose,[5] the somatically mutated fraction of RBCs (i.e., the frequency of *glycophorin A* mutants) increases with ageing and radiation exposure.[6] Longitudinal trends of haemoglobin levels also indicate a decrement with radiation (especially >1 Gy).[7] Hence, radiation exposure can lead to long-term impairments in the maintenance of homeostasis in erythropoiesis. Such radiation-associated haematological damage and deficiency may be related to longitudinal changes in RDW. Biennial health examinations in the Adult Health Study (AHS) programme[8] have accumulated longitudinal RDW data for 4204 individuals over 15 years, allowing a quantitative assessment of the effects of acute radiation exposure on peripheral blood erythrocytes over time. Thus, the present study tested the hypothesis that prior radiation

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exposure is related to a longitudinal elevation of RDW. Additionally, we evaluated the association between longitudinal RDW change and mortality in A-bomb survivors.

Subjects and methods

Study design and subjects

This study was based on the AHS programme at the Radiation Effects Research Foundation (RERF),[8] which has conducted health examinations every 2 years. For this study, RDW values from 1996 to 2010 (21 540 RDW measurements in total) were obtained from 4554 AHS participants (1500 males and 3054 females) whose Dose System 2002 (DS02) estimated radiation doses[9] were available. We focussed on the measurements obtained from subjects who were aged <90 years at the time of RDW measurement. Among the 4554 participants, the participants whose first visits occurred later than the year 2000 (211 participants) and whose urine creatinine levels exceeded 2 mg/dl (37 participants) were also excluded from the analysis.

This study was approved by the RERF Institutional Review Board and was conducted according to the principles outlined in the Declaration of Helsinki.

Statistical analysis

In regression analyses, to avoid the potential for bias that could arise from informative censoring of RDW due to death, we formulated a joint model of the longitudinal trajectory of RDW and the survival process of the study participants. The joint model consisted of a linear mixed-effects model for the RDW trajectory and a proportional hazards regression model for the risk of all-cause mortality. Regression analysis was adjusted for the city of residence, sex, autoanalyser indicator, and smoking status. In addition, regression analyses with and without adjustment for haemoglobin levels were conducted to investigate their effect on the association between RDW and radiation. Age at the initial RDW measurement, or baseline, and number of years from the baseline were included in the analysis for the evaluation of longitudinal changes in RDW. Furthermore, the interaction terms between the radiation dose and the age at baseline or number of years from baseline were evaluated to test whether the association between RDW and radiation varied by age at study entry or by increasing age, respectively. All analyses were conducted using R 3.6.3 (R Foundation for Statistical Computing, Vienna, Austria), including the JM package.[10]

A more detailed description of the methods is provided in the Supporting Information (Data S1).

Results

A total of 4204 eligible participants were included. Overall, the median (interquartile range) follow-up time was 12·2 (10·9–13·1) years, and the median (range) number of health examinations was 5 (3–7). There were 1425 deaths (33·9%). The basic characteristics of the study subjects at the baseline are shown in Table SI. Both the age at study entry and smoking status significantly differed among the radiation dose groups and were therefore included in the statistical analysis.

Longitudinal changes in RDW and the association with radiation exposure and mortality

Table I summarises the association of age and radiation exposure with RDW. RDW increased with age [0·077%/year, 95% confidence interval (CI) 0·071–0·084], male sex, and radiation dose (0·18%/Gy, 95% CI 0·13–0·23). To descriptively assess radiation effects on RDW, residuals calculated from adjusted models that did not include the radiation dose were plotted *versus* the radiation dose (Fig 1, with unadjusted age trends shown in Figure S1). With the adjustment for haemoglobin level, the association between RDW and radiation was slightly attenuated, but the association between RDW increase and radiation remained (0·14%/Gy, 95% CI 0·09–0·18).

Radiation-dose slopes for RDW varied by the number of years from the baseline (an increase in the RDW value of 0·007%/year, 95% CI 0·002–0·013). In contrast, the association between RDW and radiation did not change with the age at baseline or with sex. RDW values in current and past smokers were higher than those in non-smokers (Table I), but the smoking status had little effect on the association between RDW and radiation dose (not shown).

The hazard ratio of all-cause death was 1·46 (95% CI 1·39–1·53) with a 1% increase in RDW. Both radiation and smoking were associated with increased mortality, which is consistent with previous studies.[5] Analyses excluding participants with a diagnosis or history of anaemia (115 participants) showed similar results (not shown). Model diagnostics did not indicate any systematic lack of fit.

Discussion

Associations of higher RDW with age, male sex, or smoking are consistent with those reported previously.[2] However, to our knowledge, this is the first study to demonstrate a change in RDW (0·18% increase in RDW/Gy) several decades after exposure to ionising radiation. The results support the notion that radiation exposure causes long-term impairment of the human haematopoietic system.[6,7]

Higher RDW is strongly associated with higher all-cause mortality among A-bomb survivors. The increased risk of mortality (46%, Table I) for a 1% increment in RDW was much higher than that found in a previous meta-analysis (14% increase in the mortality risk),[2] which may be partly due to the older ages of the participants in our present study. Our present result suggests that longitudinal RDW changes

Table 1. Associations with longitudinal trajectory of RDW and all-cause mortality.

	Outcome	
	Longitudinal RDW Estimate (95% CI)	All-cause mortality Estimate (95% CI)
RDW at age 70 years*, %	12·7 (12·69, 12·79)	1·46 (1·39, 1·53)
City difference	−0·035 (−0·097, 0·027)	0·94 (0·84, 1·06)
Male (vs. female)	0·15 (0·065, 0·229)	1·75 (1·51, 2·02)
Age at baseline, years†	0·031 (0·027, 0·035)	1·11 (1·10, 1·12)
Year from baseline, years	0·077 (0·071, 0·084)	N/A
Radiation dose, Gy	0·18 (0·13, 0·23)	1·19 (1·10, 1·29)
Current smoker‡	0·19 (0·099, 0·27)	1·34 (1·13, 1·59)
Past smoker‡	0·16 (0·085, 0·24)	1·09 (0·94, 1·28)
Smoking status unknown‡	−0·10 (−0·26, 0·054)	1·20 (0·85, 1·69)
Year from baseline × radiation dose	0·007 (0·002, 0·013)	N/A

*Hiroshima female non-smokers aged 70 years; RDW, red blood cell distribution width.
†Centred at age 70 years.
‡Compared to non-smoker.

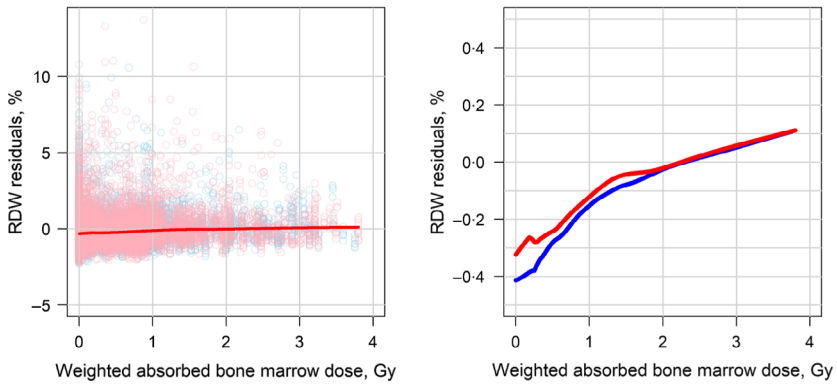

Fig 1. Relationships of radiation dose with RDW by sex (blue for males, red for females). The covariate-adjusted relationship between the radiation dose and RDW is shown. Residuals were calculated from adjusted linear mixed-effects models that did not include the radiation dose. The lines are LOWESS (locally weighted scatterplot smoothing)-smoothed curves. The right panel shows the shape of the smoothed curve across a restricted range (RDW residuals, −0·5 to 0·5%). RDW, red blood cell distribution width. [Colour figure can be viewed at wileyonlinelibrary.com]

in radiation-exposed individuals can contribute considerably to the assessment of disease morbidity and mortality risks, particularly in patients with haematological malignancies.[11]

The biological mechanisms underlying the association of RDW with radiation dose or even with ageing remain largely unclear. Inflammatory cytokines are known to inhibit erythropoietin production, primarily in renal tissue, while also impairing the proliferation of erythroid progenitor cells in response to erythropoietin, leading to a mixed population of RBC volumes.[1,2] In A-bomb survivors, elevations in the levels of peripheral blood inflammatory cytokines, T helper type 1 cells, and monocytes are significantly associated with increasing radiation dose,[12] suggesting that prolonged inflammation following radiation exposure may be a critical factor involved in increased RDW. Moreover, in the present study, the radiation-associated effect on RDW increased as the participants aged (Table I). An increase in RDW is often observed in individuals exhibiting the clonal expansion of haematopoietic stem and progenitor cells (HSPCs), that is, clonal haematopoiesis (CH), which is prevalent in those around the age of 70 years or older.[13] Clonal chromosome aberrations frequently observed in A-bomb survivors were in some cases derived from the clonal expansion of HSPCs,[14] implicating the emergence of radiation-related CH, which could accompany reduced HSPC pool sizes, induced somatic mutations, and stimulated inflammatory signalling.[15] An evolving hypothesis is that prior radiation exposure can cause an elevation of RDW through the expansion of particular HSPC clones with somatic mutations.

A limitation of the present study is that it was conducted only for participants who were alive until 1996, although the statistical analysis allowed for informative censoring during the follow-up through to 2010. The radiation effects on RDW and haematopoietic cells might be diluted over the long period (>50 years) since radiation exposure. Another limitation is the lack of clarity regarding the mechanistic

basis for the RDW increase and related mortality risk. Our present results therefore warrant further investigations, including molecular epidemiology studies and mouse model experiments, to elucidate the underlying mechanisms by which RDW increases with age and radiation exposure, and is associated with mortality.

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Author contributions

Kengo Yoshida, Munechika Misumi, Yoichiro Kusunoki and Michiko Yamada designed the research; Munechika Misumi analysed the data; Kengo Yoshida, Munechika Misumi, Yoichiro Kusunoki and Michiko Yamada wrote the paper.

Conflict of interest

The authors have no competing interests.

Supporting Information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Fig S1. The unadjusted age trends of RDW for the participants.

Table SI. Characteristics of study participants at baseline.

Data S1. Supplementary material.

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