JOURNAL OF THE AMERICAN COLLEGE OF CARDIOLOGY
VOL. 64, NO. 1, 2014

ISSN 0735-1097/$36.00
PUBLISHED BY ELSEVIER INC.

# Letters

## Antioxidants Prevent DNA Double-Strand Breaks From X-Ray–Based Cardiac Examinations

### A Randomized, Double-Blinded, Placebo-Controlled Trial

Ionizing radiation can induce deoxyribonucleic acid double-strand breaks (DSBs), which if not efficiently repaired can initiate carcinogenesis. In vivo experimental animal studies and in vitro human data (1) support the case for a protective effect of antioxidants against radiation-induced deoxyribonucleic acid damage. However, clinical data in humans are lacking. We tested in patients the hypothesis that antioxidant pre-treatment with vitamin C or *N*-acetylcysteine (NAC) may reduce DSB induction by x-ray-based cardiac examinations.

In this single-center, double-blinded, placebo-controlled study, we studied 29 controls who did not undergo x-ray-based examinations (group A), 30 patients exposed to low-dose (<3 mSv) coronary computed tomographic angiography (2) (group B), and 29 patients exposed to higher radiation doses (>9 mSv) in complex catheter-based cardiac interventions (group C). The subjects, who provided written informed consent, had an age range of 18 to 70 years. Smoking, leukemia or lymphoma, radio- or chemotherapy, and radiation-based examinations within the past 3 days were exclusion criteria. Because concerns about increased DSBs also have been raised for exposure to nonionizing radiation, such as that produced by cardiac magnetic resonance or ultrasound imaging (3,4), we only included controls who did not undergo any such examination to avoid potential confounders. The study protocol was approved by the local institutional review board (KEK-2010-0352) and the Swiss authorities (Swissmedic-2012DR2031) and was registered at ClinicalTrials.gov (NCT01578395).

Peripheral blood lymphocyte DSBs were measured by immunofluorescence as previously reported (3) before the randomized intravenous infusion (saline vs. 1.2 g NAC vs. 3 g vitamin C) and repeated within minutes after the end of the x-ray-based examination (groups B and C) or without x-ray exposure (group A). An increase in median DSB values is expressed as excess foci, either per cell for immunofluorescence microscopy or in relative units for flow cytometry (fluorescence-activated cell sorting units).

For continuous data evaluation, the Kruskal-Wallis and Mann-Whitney *U* tests were applied. Chi-square and Fisher exact tests were used for categorical variables. Values of $p < 0.05$ were considered statistically significant.

The median effective radiation dose differed among groups A (0 mSv; interquartile range: 0 to 0 mSv), B (2.0 mSv; interquartile range: 1.4 to 2.4 mSv), and C (29.0 mSv; interquartile range: 15.5 to 52.6 mSv), consistent with the study design. Although in our high-radiation group, irradiation was fractionated during angiography, whereby considerable damage repair may take place, we found substantially higher DSB induction after high-dose exposure compared with low-dose exposure by coronary computed tomography angiography. In group A, antioxidants had no impact on DSBs, with negligible excess foci not differing ($p$ = NS) between placebo (0.0), NAC (−0.02), and vitamin C (0.01). Low x-ray radiation (group B) induced 0.15 excess foci with placebo and 0.07 ($p$ = NS) after antioxidant pre-treatment (NAC or vitamin C). In the high-radiation exposure group (C), antioxidant pre-treatment yielded a significant reduction (−66%) in excess foci compared with placebo (0.1 vs. 0.3, $p < 0.05$) (**Fig. 1**), whereby the effect of vitamin C (−87%) was more pronounced than the effect of NAC (−43%) ($p = 0.005$ vs. vitamin C).

Pre-treatment with antioxidants before exposure to high radiation dose reduced the amount of excess foci by two-thirds, ranging well within the values of 50% to 87% observed for protective effects of NAC and vitamin C in experimental animal and cell culture studies (1,5).

One must be prudent in extrapolating the results of our acute-effect study with one cell type to potential long-term effects and other cell types. Therefore, a firm conclusion regarding the reduction of carcinogenic risk by antioxidant pre-treatment may not be offered, and further evaluation in larger trials should be encouraged. However, in the context of the linear-no-threshold theory and in view of the fact that

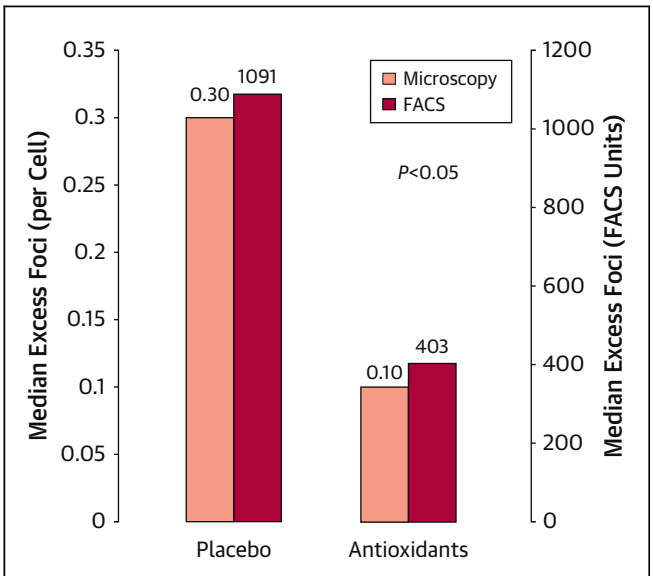


**FIGURE 1 Antioxidants Prevent X-Ray-Induced Double-Strand Breaks**

Pre-treatment with antioxidants before high radiation exposure (>9 mSv) reduced x-ray-induced deoxyribonucleic acid damage (excess foci). FACS = fluorescence-activated cell sorting.

DSBs are considered the most important genetic lesions because they can trigger deoxyribonucleic acid instability and exert tumorigenic effects, it seems reasonable to deduce that the observed substantial reduction of DSBs in lymphocytes by about two-thirds may translate into a reduction of carcinogenic risk.

**ACKNOWLEDGMENTS** The authors thank Christiane Koenig, Jose Maria Mateos, Stefano Ferrari, and Florian Mair (University of Zurich) and Arnold von Eckardstein (Institute for Clinical Chemistry, University Hospital Zurich) for their invaluable advice and technical support for the immunofluorescence analysis and Valerie Treyer for the excellent data management.

**Julia Stehli, MD**
**Tobias A. Fuchs, MD**
**Jelena R. Ghadri, MD**
**Oliver Gaemperli, MD**
**Michael Fiechter, MD, PhD**
***Philipp A. Kaufmann, MD**

*University Hospital Zurich
Ramistrasse 100
CH-8091 Zurich
Switzerland
E-mail: pak@usz.ch

http://dx.doi.org/10.1016/j.jacc.2014.04.024

Please note: This study was supported by grants from the Swiss National Science Foundation to Dr. Kaufmann and by a grant (320030-127604/1) from SwissLife to Dr. Ghadri. The authors have reported that they have no relationships relevant to the contents of this paper to disclose. Drs. Stehli, Fuchs, Ghadri, Fiechter, and Kaufmann contributed equally to this work.

## REFERENCES

**1.** Kuefner MA, Brand M, Ehrlich J, et al. Effect of antioxidants on X-ray-induced gamma-H2AX foci in human blood lymphocytes: preliminary observations. Radiology 2012;264:59–67.

**2.** Husmann L, Herzog BA, Gaemperli O, et al. Diagnostic accuracy of computed tomography coronary angiography and evaluation of stress-only single-photon emission computed tomography/computed tomography hybrid imaging: comparison of prospective electrocardiogram-triggering vs. retrospective gating. Eur Heart J 2009;30:600–7.

**3.** Fiechter M, Stehli J, Fuchs TA, et al. Impact of cardiac magnetic resonance imaging on human lymphocyte DNA integrity. Eur Heart J 2013;34:2340–5.

**4.** Picano E, Vano E, Rehani MM, et al. The appropriate and justified use of medical radiation in cardiovascular imaging: a position document of the ESC Associations of Cardiovascular Imaging, Percutaneous Cardiovascular Interventions and Electrophysiology. Eur Heart J 2014;35:665–72.

**5.** Reliene R, Pollard JM, Sobol Z, et al. *N*-acetyl cysteine protects against ionizing radiation-induced DNA damage but not against cell killing in yeast and mammals. Mutat Res 2009;665:37–43.