BRIEF COMMUNICATIONS

Breast Cancer Screening Using Small-Angle X-ray Scattering Analysis of Human Hair

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In a double-blind study, James et al. (1) reported consistent distinctive changes in the x-ray diffraction patterns of pubic hair from all eight patients with breast cancer compared with those of pubic hair from four healthy women. Results obtained from samples of scalp hair were almost as clear-cut; all 15 of the patients with breast cancer but only three (19%) of 16 of the healthy women showed an aberrant finding. Samples from women with a hereditary predisposition for breast cancer because they carried a BRCA1 gene mutation also showed either a full or a partial aberrant x-ray pattern (2). The aberrant finding was described as a superimposed ring corresponding to a molecular spacing of around 4.44 nm, probably arising from randomly orientated lipid bilayers of the plasma membrane of the hair cells. The x-ray diffraction studies were performed with synchrotron radiation according to previously published protocols (3–5).

James et al. (1) proposed the use of x-ray diffraction analysis of pubic hair as a reliable method to screen for the presence of or increased risk for breast cancer. However, a repetition of this study by Briki et al. (6) using scalp hair revealed conflicting results. The diffraction ring around 4.44 nm that James et al. reported as specific for patients with breast cancer was observed in all 10 samples from healthy control women, but it was observed in only eight of 10 samples from patients with breast cancer. Other groups (7–9) have also reported results that conflict with those of James et al. (1).

Routine investigation of hair specimens by synchrotron radiation would be difficult because the apparatus is available only at specialized centers. We, therefore, decided to repeat the investigations with a recently developed small-angle x-ray scattering (SAXS) system (10) (Nanostar System; Bruker AXS GmbH, Karlsruhe, Germany). The SAXS system offers high reliability and fast measuring time without the requirement for synchrotron radiation. It consists of an ordinary sealed-tube x-ray generator powering a standard fine-focus Cu tube. The collimation system, a pair of cross-coupled multilayer mirrors (Goebel mirrors), produces a nearly perfect parallel beam. Measurements are done in the laboratory by a high-resolution SAXS camera with a pinhole system. Samples are measured in transmission, and the scattering signal is detected at a distance of 65 cm from the specimen by a high-sensitivity, two-dimensional area detector.

We obtained samples of scalp and pubic hairs from nine breast cancer patients and from eight healthy control women after they gave written informed consent. The research protocol was approved by the Human Subjects Review Committee, University of Heidelberg, Germany. Two of the breast cancer patients, previously screened for BRCA1 and BRCA2 mutations, showed the common BRCA1 mutation 5382insC, which is associated with hereditary breast cancer. Bundles of about 10 hairs were tied and fixed in a parallel manner as tightly as possible and then subjected to a monochromatic Cu-Kα (i.e., copper-potassium α) radiation beam for 30,000 seconds. Silver benenate, a typically used calibration standard in SAXS, was used to accurately determine the sample-to-detector distance, which is needed for determination of the exact reciprocal spacing (peak position in nanometers).

Fig. 1. Left—typical small-angle x-ray scattering spectrum of samples from a patient with breast cancer. Top: scalp hair. Bottom: pubic hair. Ten-nanometer diffuse maxima at smaller angles were seen in both hair types (arrows 1), but the 4.44-nm ring was present only in scalp hair (arrows 2). Right—intensity distribution along the vertical axis showing the two maxima (arbitrary units).

Fig. 1 shows typical measured SAXS spectra from scalp and pubic hairs. In agreement with previous investigations (1,6–9) with synchrotron radiation, two maxima were generally found, one at 10 nm and the other at 4.44 nm (Fig. 1, labeled 1 and 2). Both sample types (scalp and pubic hairs) demonstrated anisotropy in the scattering signal at the 10-nm maximum. Evaluation of the azimuthal distribution indicated that the intensity of all observed rings at 4.44 nm was also quite oriented, thus forming sharp maxima rather than the “diffuse ring” pattern described by James et al. (1). The SAXS spectrum was isotropic in only the following two cases: 1) one pubic hair sample from a patient with breast cancer who also carried the BRCA1 mutation and 2) one scalp hair sample from a healthy subject showing a very diffuse scattering signal at lower angles (but no maximum at 4.44 nm).

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This change is not visible in the pubic hair sample of the second affected patient with a BRCA1 mutation.

Our results for the 4.44-nm maximum and a comparison with the results obtained by James et al. (1) are shown in Table 1. The most striking difference was the lack of 4.44-nm diffracting rings in pubic hair samples in our population of patients compared with the 100% reported by James et al. (1). Furthermore, there was no statistically significant difference between our groups of breast cancer patients and control women for scalp or pubic hair, in contrast to the results of James et al. (1). Rather, our results mirror those of other groups (6–9).

In conclusion, standard SAXS using the Nanostar System is a suitable method for characterization of human hair diffraction patterns. However, our results did not indicate specific differences in the x-ray diffraction patterns of scalp or pubic hair samples from healthy women and from women with breast cancer. Although the number of patients was small, there were no obvious differences to substantiate the findings reported previously by James et al. (1). Thus, in agreement with other studies of synchrotron radiation (6–9), the presence of the 4.44-nm maximum in the x-ray diffraction pattern is not an indication of either the presence of breast cancer or the susceptibility to breast cancer.

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NOTE

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