HISTOPATHOLOGIC STUDIES OF THE DISRUPTION OF MURINE TUMOR VASCULATURE BY MILD-INTENSITY ULTRASOUND

A.K.W. Wood,[‡] R.M. Bunte,[*] S. Ansaloni,[†] W.M-F. Lee,[§] and C.M. Sehgal.[†] University Laboratory Animal Resources,[*] Depts of Radiology [†] and Medicine, [§] University of Pennsylvania Medical Center, and Dept Clinical Studies (Phila), [‡] School of Veterinary Medicine, University of Pennsylvania, 3900 Delancey St, Philadelphia, PA 19104, USA.

Introduction

Angiogenesis provides the vascular supply to a growing neoplasm, however, the new blood vessels are fragile, leaky and not fully functional. They are disrupted by mild-intensity ultrasound. This study was aimed at defining the histopathologic changes that could explain the antivascular effects of mild-intensity ultrasound on tumor neovasculature.

Methods

In 27 mice (C3HV/HeN strain) a subcutaneous melanoma ($K1735^{22}$) was insonated (1, 2 or 3 min) with continuous mild-intensity, physiotherapy ultrasound (1 MHz; I_{SATA} = 2.3 W cm^{-2}). B-mode ultrasonographic observations were made of the neoplasm before and after its insonation. Following each B-mode study, a contrast agent (0.1 mL Optison) was intravenously injected and the enhancement of power Doppler images was recorded on a videotape for quantitative analysis. The mice were euthanized either immediately or 24h after insonation and the tumor was removed for histopathology. A linear regression analysis was performed to establish whether there was a relationship between the % area of histological change and the % increase in tumor avascularity detected in the contrast enhanced power Doppler images.

Results

Analyses of contrast enhanced power Doppler observations showed that insonation significantly ($p < 0.005$) increased the avascular area in the neoplasm. Histologically, the untreated neoplasm was characterized by spindle shaped cells arranged in streams and bundles with unapparent vasculature. The predominant acute effect of insonation was an apparently irreparable dilation of the tumor capillaries with associated intercellular edema; other immediate effects were hemorrhage, and increased intercellular fluid. Liquefactive necrosis of neoplastic cells was a delayed effect. Pre-existing arterioles and venules were unaffected by the insonation. There was a high correlation ($R^2 = 0.91$) between the % area affected on histologic examination and the % increase in avascularity of the neoplasm in the Doppler study.

Discussion

Other workers have insonated endothelial cells growing in tissue culture and noted the presence of nitric oxide, a vasodilator, in the culture medium. Whether an ultrasound induced endothelial synthesis of nitric oxide caused the capillary dilation in our *in vivo* study requires further investigation. It appeared that the necrosis of the neoplastic cells was not related to a direct effect of ultrasound on the neoplastic cells, but rather to a generalized tumor ischemia following the acute effects of insonation on the neoplasm's capillaries. The observed bioeffects following insonation may be thermal in origin, however, other mechanisms including cavitation, radiation pressure and other non-linear effects should also be considered. Supported by NIH grant no. EB001713.