Morphology and distribution of focal hemorrhage following primary blast injury

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Introduction

Understanding injury from blast overpressure is a major challenge facing modern military and civilian medicine. Mechanisms behind blast-induced brain injury are not yet fully understood. We have developed a model of primary blast injury that results in focal blood-brain-barrier disruption.

Methods

Using a .308 bolt action rifle and a modified steel casing fitted for a .50 caliber primer, we have developed a benchtop shock tube capable of producing overpressures of up to 440 kPa. Male Sprague Dawley rats were anesthetized with isoflurane and immobilized in a Kevlar hammock offset from the muzzle to eliminate loading by the exiting gas jet and burning propellant. Injury severity from lateral exposure was tested at three levels of blast overpressure: 150, 280, and 440 kPa, determined by distance from the shock tube. Animals were then transcardially perfused with PBS and 4% paraformaldehyde. Brains were cut into 50 micron coronal sections and examined immunohistochemically. Primary markers were Biotinylated GtxRt IgG1 and RbxMs anti-laminin IgG1 to detect displaced blood and to visualize vasculature, respectively. Sections were imaged with an Olympus E600 upright microscope and then analyzed with NI Vision Assistant to measure lesion size and location.

Results

Pressure curves were characterized at varying distances to find appropriate levels of blast overpressure outside of the gas jet. In animals exposed to blast, small focal regions of high fluorescence were found in the parenchymal brain tissue, indicating the presence of rat IgG. Lesions were too small for detection via high resolution MRI. Lesion size and frequency increased with blast exposure. Initial study at the 280 kPa blast level (N=2) resulted in lesion densities of 0.25 and 0.14 injuries/mm² of brain tissue. A total of 137 separate injuries were found in the brain of one animal in which every section was examined. Many of these lesions persisted through multiple sections. Just 79 injuries were identified in a second animal where every fifth section was sampled. Lower levels of blast injury were also examined (50, 100, 125 kPa), but no significant injury was found in these cases. Further study is ongoing. Lesion distribution was measured from the midline of each section. In one animal the center of the distribution was found to shift towards the hemisphere ipsilateral to the site of blast exposure, experiencing a 5.4% (0.9 mm) shift from the center of the brain. Injuries were most commonly found in the deep dorsal ipsilateral cortex, at the boundary of the cortex and the hypothalamus on both sides, and at the contralateral ventral surface of the cortex. Laminin staining revealed vascular structure in the brain and surrounding injury sites. Knowing vessel structure helped filter suspected areas of injury, as only sites with clearly associated vasculature were considered lesions. A common feature that many vessels shared was a branch point close to the injury site.

Conclusions

Despite widespread lesions, general trends for injury location have emerged. The boundary between the cortex and deeper brain structures seems to be particularly vulnerable, possibly due to mechanical impedance mismatches between gray and white matter. Lesion distribution favored the ipsilateral hemisphere. Another common injury zone was the contralateral ventral surface of the cortex, suggesting possible coup-contrecoup injury resulting from the blast wave. Laminin staining helped identify lesion sites and revealed information about microscale injury factors. In standard engineering applications, corners and joints are sites of stress concentration. Blood vessels appear to follow the same rules, as branch points were subject to tension during tissue deformation. Blast brain injury remains a topic of intense research. Understanding mechanisms behind its mechanical and physiological effects on both neural and vascular components of the brain will aid in developing therapies to treat this important problem.

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