Toll-like receptor 4 signalling in striated muscle during equine hyperinsulinaemia.

MA de Laat, CK Clement, KJ Gruntmeir, MN Sillence and VA Lacombe.
School of Earth, Environmental and Biological Sciences, Queensland University of Technology, Brisbane, Qld, Australia.


Introduction

Toll-like receptors (TLR) are integral to pathogen recognition by the innate immune system and trigger pro-inflammatory cytokine release. In addition, TLR4 signalling plays a lesser understood role in linking inflammation and metabolic disease by regulating glucose transporters (GLUT). In horses, hyperinsulinaemia is a key risk factor for laminitis. However, other metabolic disease sequelae recognised in humans, such as cardiovascular disease, are poorly elucidated in horses. The study aim was to investigate TLR4 signalling and GLUT regulation in equine striated muscle during hyperinsulinaemia.

Methods and materials

Horses were treated (48h) with a prolonged-euglycaemic hyperinsulinaemic clamp or balanced electrolytes (controls). Left ventricular and skeletal muscle protein extracts (total and cell-membrane fractions) were analysed by immunoblotting for TLR4, GLUT1, GLUT4, TNF-α and IL-6 expression.

Results

Hyperinsulinaemia induced acute laminitis in treated horses. Down-regulation of TLR4 from the cell membrane (by 77%, p<0.05) occurred in myocardium of treated horses, without a change in TNF-α or IL-6 protein content. In contrast, hyperinsulinaemia did not alter skeletal muscle TLR4 signalling. Myocardial GLUT isoforms were not altered by hyperinsulinaemia, either in cell-membrane or total lysates. However, GLUT1 content decreased in skeletal muscle of treated horses, while GLUT4 did not change, compared to controls.

Relevance to clinical equine practice

A lack of GLUT4 down-regulation in skeletal muscle during hyperinsulinaemia suggests that insulin resistance is not required for laminitis development. Further, hyperinsulinaemia may directly decrease TLR4 activation in the myocardium of non-obese individuals, potentially reducing cardiac immune system capability. Investigation of cardiac TLR4 signalling in obese insulin-resistant horses is required.