The effect of prosthesis-related loading on soft tissue health

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Worldwide, one individual loses a limb every 30 seconds because of complications of diabetes [1]. Considering our aging population, this is expected to worsen over the decades to come [2]. To mitigate the negative impact of an amputation on the individual, restoring mobility is key. Often a prosthesis is fitted that replaces the lost limb. One of its most important parts is the socket, which forms the direct connection between the prosthesis and the leg. However, designing and fitting a good prosthetic socket is a considerable challenge: Most amputees report discomfort and pain when using their prosthesis, which often leads to reduced functionality and low acceptance rates. A major reason is our limited understanding of how compressing soft tissues like muscle between the rigid socket and the bone affects soft tissue health. Whilst researchers have explored how prolonged, static loading, for example when standing with a prosthesis, impacts on soft tissue health [3], dynamic loading scenarios like walking and running, have been neglected [4]. We therefore aim to compare the effect of static and dynamic loading representative of prosthetic use on muscle tissue.

We developed an *ex vivo* experiment that uses two different muscles from the hindlimb of Sprague Dawley rats (extensor digitorum longus and soleus). The muscles were dissected and compressed with a pressure of 100kPa for 1 hour either statically, representing standing and sitting, or dynamically, representing walking and running (frequencies of 1.42Hz and 4Hz). Subsequently, the tissues were stained with Procion Yellow MX4R (ProY) for 1 hour before being processed for microscopy. ProY is a fluorescent stain that enters cells that have lost their membrane integrity. Accordingly, we could detect mechanically damaged cells under the microscope (Leica SP8 system). The number of dead cells was counted across five samples for each experimental group in a semi-automated process with ImageJ software. The results were tested for statistically significant differences ($p \leq 0.05$) with Mann-Whittney-tests.

Both dynamic loading scenarios induced more cellular damage than static loading (median number of dead cells: 79.5 (static), 90 (1.42Hz), 135.5 (4Hz)). However, results were only statistically significant between the static and 4Hz ($p=0.011$), and the 1.42Hz and 4Hz group ($p=0.044$). Interpreting these results in a clinical context, sitting, standing, and walking seem to bare a similar risk of damage for the muscle tissue in the residual limb. Fast walking or running however is potentially more harmful to the tissues, which could result in pain and tissue damage. To avoid these issues, adjusting socket fit to minimise soft tissue deformation is crucial. Additionally, identifying threshold levels for damage-inducing loading levels related to the activity performed by an amputee would be helpful. This information can not only inform clinical guidelines on prosthetic socket fit and use, but also be integrated into home-monitoring systems to allow for patient-specific risk assessment beyond the hospital.

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