Low Molecular Weight Heparin impairs tendon repair

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Introduction: Thromboprophylaxis inhibits thrombin activity. Thrombin has many biological properties similar to growth factors. It can activate matrix-bound mitogenic factors by catalysis, but also activate cell surface receptors. Finally, thrombin can stimulate mitogenic cell surface receptors independent of its enzymatic function (1). Thrombin is activated by factor Xa, as a result of the coagulation cascade.

We have previously shown that locally injected bovine thrombin improves the healing of transected rat Achilles tendons (3). How about the opposite: Would decreased thrombin activity have an adverse effect? Low molecular weight heparins (LMWHs) inhibit thrombin. We therefore hypothesized that LMWH inhibits tendon repair in rats. Because we needed to give the LMWH at a dose with some relevance to the human situation, we measured anti-factor Xa activity, to ascertain that we reached a thromboprophylactic level. In order to exclude that any effects were the consequences of a possible increase in bleeding, continuous LMWH treatment was compared with controls that received preoperative LMWH treatment only. Having seen that continuous LMWH treatment impaired tendon healing, we tested a second hypothesis: that daily periods of normalized factor Xa activity would allow sufficient thrombin stimulation to allow normal repair.

Materials and Methods: 20 rats were randomized to continuous LMWH treatment versus control (experiment 1). Then 30 rats were randomized to continuous treatment, a single LMWH injection during the operation, or untreated control (experiment 2). To mimic a clinical dosing regime, 20 rats were then randomized to LMWH or saline as subcutaneous injections twice daily (experiment 3). Results were evaluated by mechanical testing at 7 days. Finally, anti-factor Xa activity was analyzed in 16 rats (experiment 4). Blood was taken 2 and 12 hours after LMWH injections and from controls.

A 3 mm segment was removed from the Achilles tendon. The tendon was left unsutured, and there was no post-operative immobilisation. Osmotic mini pumps were implanted subcutaneously to deliver 1.6 IE dalteparin sodium (Fragmin®) per hour. Subcutaneous injections consisted of 40 IE (200 IE/kg/24 h). The surgeon was not aware of treatment until after the tendon surgery was completed and it was time to insert the minipump.

Mechanical parameters measured were force at failure, stiffness and energy uptake. Ultimate stress was calculated. Blood collection was performed by heart puncture. Measurement of anti-Xa was performed using the reagent Coamtic®Heparin purchased from Haemochrom Diagnostica (Mölndal, Sweden) on an automated coagulation analyzer. Force at failure was chosen as the primary outcome variable. Experiments were evaluated with Student’s t-test or Anova.

Results: There were no bleeding complications. All specimens ruptured in the tendon callus that had formed in the gap. Comparing continuous treatment with untreated controls, LMWH caused a 27 % decrease in force at failure. Stiffness and energy uptake were also decreased Effect of continuous LMWH

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<thead>
<tr>
<th></th>
<th>Force</th>
<th>Str</th>
<th>Energy</th>
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<tbody>
<tr>
<td>Control</td>
<td>1</td>
<td>2</td>
<td>3</td>
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<tr>
<td>LMWH</td>
<td>0.008</td>
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Comparing injections twice daily with controls, no significant differences were seen, and differences of an important magnitude can be excluded with confidence.

Discussion: Continuous LMWH treatment has a detrimental effect on tendon repair. The tendon callus became thinner. Material properties were unchanged. This suggests that most of the detrimental effect was related to proliferation and growth of the early callus, whereas tissue differentiation (and probably the ability to respond to mechanical stimulation of differentiation) was undisturbed. This suggests that periods of mitogenic stimulation are enough to trigger the processes necessary for early tendon callus formation, whereas periods of decreased fibrin formation might be sufficient to reduce thrombus formation or growth. There is a cause for concern regarding new drugs, developed to have a continuous effect enabling once-weekly doses. It is not unlikely that these drugs will have detrimental effects like the continuous dosing in this study.

In conclusion, this study confirms indirectly the importance of thrombin signalling for repair, and suggests that thromboprophylaxis might hamper healing via interference with factor Xa; but intermittent treatment might be harmless.

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The regime with two daily injections lead to a significant increase in antifactor Xa activity of a relevant magnitude 2 hours after the injection, but the activity had normalized after 12 hours. Thus we had mimicked the human situation, in which the antifactor Xa activity has time to normalize between injections.

References: