Author(s): Munukka, Matti; Waller, Benjamin; Rantalainen, Timo; Häkkinen, Arja; Nieminen, Miika T.; Lammentausta, Eveliina; Kujala, Urho; Paloneva, Juha; Sipilä, Sarianna; Peura, Arttu; Kautiainen, Hannu; Selänne, Harri; Kiviranta, Ilkka; Heinonen, Ari

Title: Efficacy of Progressive Aquatic Resistance Training for Tibiofemoral Cartilage in Postmenopausal Women with Mild Knee Osteoarthritis : A Randomised Controlled Trial

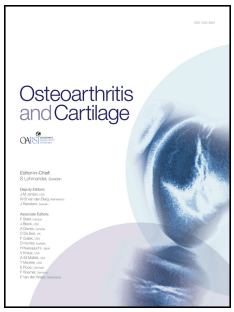
Year: 2016

Version:

Please cite the original version:

Munukka, M., Waller, B., Rantalainen, T., Häkkinen, A., Nieminen, M. T., Lammentausta, E., Kujala, U., Paloneva, J., Sipilä, S., Peura, A., Kautiainen, H., Selänne, H., Kiviranta, I., & Heinonen, A. (2016). Efficacy of Progressive Aquatic Resistance Training for Tibiofemoral Cartilage in Postmenopausal Women with Mild Knee Osteoarthritis : A Randomised Controlled Trial. Osteoarthritis and Cartilage, 24(10), 1708-1717. https://doi.org/10.1016/j.joca.2016.05.007

Accepted Manuscript



Efficacy of Progressive Aquatic Resistance Training for Tibiofemoral Cartilage in Postmenopausal Women with Mild Knee Osteoarthritis: A Randomised Controlled Trial

Matti Munukka, MSc, Benjamin Waller, MSc, Timo Rantalainen, PhD, Arja Häkkinen, PhD, Miika T. Nieminen, PhD, Eveliina Lammentausta, PhD, Urho M. Kujala, MD, PhD, Juha Paloneva, MD, PhD, Sarianna Sipilä, PhD, Arttu Peuna, Hannu Kautiainen, BA, Harri Selänne, MD, PhD, Ilkka Kiviranta, MD, PhD, Ari Heinonen, PhD

PII: S1063-4584(16)30084-X

DOI: 10.1016/j.joca.2016.05.007

Reference: YJOCA 3759

To appear in: *Osteoarthritis and Cartilage*

Received Date: 21 January 2016

Revised Date: 19 April 2016

Accepted Date: 9 May 2016

Please cite this article as: Munukka M, Waller B, Rantalainen T, Häkkinen A, Nieminen MT, Lammentausta E, Kujala UM, Paloneva J, Sipilä S, Peuna A, Kautiainen H, Selänne H, Kiviranta I, Heinonen A, Efficacy of Progressive Aquatic Resistance Training for Tibiofemoral Cartilage in Postmenopausal Women with Mild Knee Osteoarthritis: A Randomised Controlled Trial, *Osteoarthritis and Cartilage* (2016), doi: 10.1016/j.joca.2016.05.007.

Efficacy of Progressive Aquatic Resistance Training for Tibiofemoral Cartilage in Postmenopausal Women with Mild Knee Osteoarthritis: A Randomised Controlled Trial

Matti Munukka, MSc[1*], Benjamin Waller, MSc[1*], Timo Rantalainen, PhD[2], Arja Häkkinen, PhD[1,3], Miika T. Nieminen, PhD[4,5], Eveliina Lammentausta, PhD[4], Urho M. Kujala, MD, PhD[1], Juha Paloneva, MD, PhD[6], Sarianna Sipilä, PhD[1,7], Arttu Peuna[4,5], Hannu Kautiainen, BA[8,9], Harri Selänne, MD, PhD[10], Ilkka Kiviranta, MD, PhD[11], Ari Heinonen, PhD[1]

1) Department of Health Sciences, University of Jyväskylä, Jyväskylä, Finland
2) Centre for Physical Activity and Nutrition Research, School of Exercise and Nutrition Sciences, Deakin University, Melbourne, Australia
3) Department of Physical Medicine and Rehabilitation, Central Finland Central Hospital, Jyväskylä, Finland
4) Department of Diagnostic Radiology, Oulu University Hospital, Oulu, Finland
5) Centre for Medical Imaging, Physics and Technology research, University of Oulu, Oulu, Finland
6) Department of Surgery, Central Finland Central Hospital, Jyväskylä, Finland
7) Gerontology Research Center, University of Jyväskylä, Finland
8) Department of General Practice and Primary Health Care, University of Helsinki, Helsinki, Finland
9) Unit of Primary Health Care, Kuopio University Hospital, Kuopio, Finland
10) LIKES Research Centre, Jyväskylä, Finland
11) Department of Orthopaedics and Traumatology, University of Helsinki, and Helsinki University Hospital, Helsinki, Finland

Email addresses:
matti.munukka@jyu.fi
ben.waller@jyu.fi
t.rantalainen@deakin.edu.au
arja.hakkinen@jyu.fi
miika.nieminen@oulu.fi
eveliina.lammentausta@oulu.fi
urho.m.kujala@jyu.fi
juha.paloneva@ksshp.fi
sarianna.sipila@jyu.fi
Arttu.Peuna@ppshp.fi
hannu.kautiainen@medcare.fi
Harri.Selanne@likes.fi
ilkka.kiviranta@helsinki.fi
ari.o.heinonen@jyu.fi

**Both authors have equal contribution.*
Corresponding authors: MSc Matti Munukka and MSc Benjamin Waller. University of Jyvaskyla, Department of Health Sciences, P.O. Box 35, FI-40014 University of Jyvaskyla, Finland. Phone: +358408053606 and +358408053569. E-mail: matti.munukka@jyu.fi and ben.waller@jyu.fi

Running title: The impact of aquatic training on tibiofemoral cartilage

Abstract

Objective: To study the efficacy of aquatic resistance training on biochemical composition of tibiofemoral cartilage in postmenopausal women with mild knee osteoarthritis (OA). **Design:** 87 volunteer postmenopausal women, aged 60-68 years, with mild knee OA (Kellgren Lawrence grades I/II and knee pain) were recruited and randomly assigned to an intervention (n=43) and control (n=44) group. The intervention group participated in 48 supervised aquatic resistance training sessions over 16 weeks while the control group maintained usual level of physical activity. The biochemical composition of the medial and lateral tibiofemoral cartilage was estimated using single-slice transverse relaxation time (T2) mapping and delayed gadolinium-enhanced magnetic resonance imaging of cartilage (dGEMRIC index). Secondary outcomes were cardiorespiratory fitness, isometric knee extension and flexion force and knee injury and osteoarthritis outcome questionnaire. **Results:** After 4-months aquatic training, there was a significant decrease in both T2 -1.2ms (95% CI: -2.3 to -0.1, p=0.021) and dGEMRIC index -23ms (-43 to -3, p=0.016) in the training group compared to controls in the full thickness posterior region of interest (ROI) of the medial femoral cartilage. Cardiorespiratory fitness significantly improved in the intervention group by 9.8% (p=0.010). **Conclusions:** Our results suggest that, in postmenopausal women with mild knee OA, the integrity of the collagen-interstitial water environment (T2) of the tibiofemoral cartilage may be responsive to low shear and compressive forces during aquatic resistance training. More research is required to understand the exact nature of acute responses in dGEMRIC index to this type of loading. Further, aquatic resistance training improves cardiorespiratory fitness.

Keywords: Osteoarthritis; Aquatic Exercise; Magnetic Resonance Imaging (MRI), Cartilage, Randomised Controlled Trial

Trial registration number: ISRCTN65346593

INTRODUCTION

Knee osteoarthritis (OA) is a common cause of pain and limitations in physical function globally and represents a significant burden on healthcare costs[1]. The development of knee OA progresses slowly over years[2]. In the early phase of OA development changes are seen in the biochemical composition of the cellular matrix of the cartilage. These include a decrease in glycosaminoglycan (GAG) content, responsible for hydrophilic properties of collagen matrix, and loss of integrity of the collagen matrix, responsible restraining hydrostatic pressure and maintaining cartilage stiffness[3]. As this degeneration progresses the biomechanical properties of the cartilage are altered, reducing its ability to resist and distribute tensile, shear and compressive forces, causing further degradation and joint failure[4].

There is no known cure or treatment that prevents or reverses the biochemical changes in the cartilage, therefore, the current management of OA focuses on reducing the symptoms and decreased function associated with the disease[1]. Exercise, irrespective of modality (land or water) or type (strength or aerobic), has been shown to be effective in achieving these aims[5,6]. Moreover, an active life style with participation in exercise has been shown to be beneficial for maintenance of the biochemical properties of cartilage in both animals[7,8] and humans[9,10]. Further, exercise has been shown to reverse cartilage atrophy seen in disuse and immobilisation studies[11,12] and slow down progression of OA in animals[13]. Therefore, exercise could be an effective intervention for the maintenance of cartilage health. However, studies investigating the effect of exercise interventions on healthy and degenerated human cartilage are sparse[14-17]. Only two previous studies have investigated the effects of land based exercise on the biochemical composition of cartilage in postmenopausal women with mild knee OA, i.e. Kellgren-Lawrence grades I/II and knee pain[15,16]. We found an improvement in

the collagen matrix in the patella cartilage of women with mild knee OA following a one-year, three time a week, high-impact exercise intervention[15] while we did not see any worsening or improvement in the collagen matrix or GAG concentration of the tibiofemoral cartilage in the same study[16]. Therefore, there is sufficient evidence to show cartilage health is maintained by appropriate mechanical stimulus and environment[9,18].

Pain is a major modulator for activity avoidance in people with knee OA[19]. Water is a facilitating environment in which persons with lower limb OA can safely and comfortably exercise at high intensities utilising full joint range of motions[20] . Our recent systematic review showed that aquatic exercise has a similar effect on pain and self-reported functioning compared to land-based training[6]. Moreover, in our previous studies Pöyhönen *et al.*[21] and Valtonen *et al.*[22] both showed significant benefits of a progressive aquatic resistance training program for physical functioning in healthy women and following knee arthroplasty, respectively. Regular cyclic movements performed during aquatic exercise may provide sufficient mechanical stimulus and facilitate improved exchange of nutrients thus increasing chondrocyte activity[4,18]. Therefore, the aim of this study was to investigate if progressive, intensive and high volume aquatic resistance training affects the biochemical composition of tibiofemoral cartilage in postmenopausal women with mild knee osteoarthritis.

MATERIALS AND METHODS

Study design

This study was a 4-month registered randomised controlled trial (ISRCTN65346593) with two experimental arms: 1) aquatic resistance training and 2) control. Recruitment and data collection took place between January 2012 and May 2013 and followed the published protocol without changes[23]. Included participants were women aged 60-68 years with mild knee OA. In this study we classify mild knee OA as radiographic changes in tibiofemoral joint grades I (possible osteophytes) or II (definite osteophytes, possible joint space narrowing) according to the Kellgren-Lawrence (K/L) classification and experiencing knee pain on most days[24]. The study protocol (Dnro 19U/2011) was approved by the Ethics Committee of the Central Finland Health Care District and conforms to the Declaration of Helsinki. Written informed consent was obtained from all participants prior to enrolment.

Subject recruitment

A multistage recruitment process was implemented (Figure 1). Initially, postmenopausal women from the Jyväskylä region in Central Finland were voluntarily recruited through advertisements in local newspapers. Preliminary eligibility was assessed using a structured telephone interview (n=323), followed by evaluation of osteoarthritis severity in the tibiofemoral joint with radiographs (n=180) and finally through medical screening (n=111). Inclusion criteria were: postmenopausal woman aged 60–68 years, experiencing knee pain on most days, participates in intensive exercise ≤ twice a week, radiographic changes in

tibiofemoral joint K/L I or II, no previous cancer or chemotherapy, no medical contraindications or other limitations to full participation in an intensive aquatic training program and complete T2 data. Exclusion criteria included a T-score <-2.5 (indicating osteoporosis)[25] measured from the femoral neck using dual-energy X-ray absorptiometry (DXA), resting knee pain visual analogue scale (VAS) >50/100, surgery of the knee due to trauma or knee instability, meniscectomy within the last 12 months, inflammatory joint disease, intra-articular steroid injections in the knee during the previous 12 months, contraindications to MRI and allergies to contrast agents or renal insufficiency. Due to confounding factors related to obesity, a body mass index (BMI) of >34 kg/m^2 was an exclusion criterion.

Figure 1 here.

Randomisation and blinding

After baseline measurements, all participants were randomly allocated with a three digit identification number (ID) to blind researchers to intervention allocation and provision. A blinded statistician, only provided with ID and K/L grade, performed a computer generated block randomisation of size of 10, stratified according to K/L grade I or II. The MRIs were performed by external radiographers and segmentation was performed blinded to intervention allocation.

Health questionnaire

At baseline, a researcher-designed questionnaire was used to record physical activity levels, general health, medical conditions, current medications, menopausal status and hormone therapy. Leisure time physical activity levels, i.e. activity type (e.g. walking or golf), duration and intensity, prior to the study inclusion were converted into metabolic equivalent task (MET)-hours per week[26].

Primary outcome measures

Primary outcomes for this study were T2 relaxation time (T2) mapping (milliseconds, ms) and delayed gadolinium-enhanced magnetic resonance imaging of cartilage (dGEMRIC index, ms). Images were taken using a Siemens Magnetom Symphony Quantum 1.5-T scanner (Siemens AG, Medical Solutions, Erlangen, Germany). Single sagittal slice images from the centre of the medial and lateral femoral condyles were taken from the knee with the highest K/L grade (affected knee). In cases of identical grading bilaterally, the right knee was imaged. Images were manually segmented using an in-house MATLAB application with built-in motion correction for dGEMRIC (Mathworks, Inc. Natick, MA, USA). In this study we divided the femoral cartilage into three ROIs; anterior, central and posterior (Figure 2). dGEMRIC indices were corrected for BMI[27]. Precision, scan-rescan, (CV_{RMS}) of dGEMRIC in asymptomatic subjects is 7% for full-thickness ROIs and 5% for bulk cartilage[28]. In our laboratory, the inter-observer error (CV_{RMS}) for T2 full-thickness ROIs was 1.3% to 3.3% and 2.8% to 4.0% for dGEMRIC index. The full MRI protocol and example images are provided in the online supplemental material.

Figure 2 here.

Secondary outcomes

Physical performance

Cardiorespiratory fitness (VO_2 peak, ml/kg/min) was estimated using the UKK 2 km walking test (UKK Institute, Tampere, Finland)[29]. Isometric knee extension and flexion force (N) of the affected knee was measured using an adjustable dynamometer chair (Good strength; Metitur Ltd, Jyväskylä, Finland)[30].

Self-reported symptoms

Self-assessed impact of OA on pain, other symptoms, activities of daily living, sports and recreation and knee related quality of life were assessed using the validated Finnish[31] Likert version of the knee injury and osteoarthritis outcome score (KOOS) questionnaire[32]. Scores for each domain range between 0 to 100, with a score of 0 indicating extreme and 100 no knee problems.

Daily physical activity

Daily physical activity, for the whole intervention period, of each participant was recorded using a leisure time physical activity diary from which metabolic equivalent task (MET-

hours) per week was calculated[26]. In week 13 of the intervention period the daily physical activity (excluding intervention) was measured for 3 consecutive days including one weekend day using an accelerometer (Hookie AM 20, Traxmeet Finland). Mean amplitude deviation (MAD) of the resultant acceleration signal for each 5-sec epoch were calculated and categorized according to Vähä-Ypyä *et al.*[33].

Exercise protocol

The participants in the intervention group received one hour of supervised lower limb aquatic resistance training three times a week for 16 weeks, for a total of 48 training sessions. Resistance of exercises was progressed with three different levels: barefoot, small fins and large resistance boots[21] and the training leg performed all the movements without contact with the pool walls or bottom i.e. non-weight bearing. The intervention was completed in small groups of 6-8 subjects in a pool heated to 30-32 degrees with two instructors: one ensuring intensity and the other full range of movement. Intensity of the training sessions was set at "as hard and fast as possible" to ensure maximal muscle contraction. Pöyhönen *et al.*[34] discovered that during maximal knee flexion and extension exercises in water with large resistance boots the drag forces produced were 80-85% (145 ± 30 N) of maximal isokinetic movements. Full range of motion was strictly controlled for to ensure optimal movement of synovial fluid and exposure of the whole cartilage to the low compressive and shear forces created by the muscle contraction and movement. Training intensity was monitored using heart rate monitors (Polar Electro Ltd, Kempele, Finland), rate of perceived exertion (RPE) using the Borg 6-20 scale[35] and number of repetitions achieved per movement. Full

description of exercises and training methodology can be found from the online supplemental material.

Control group

The control group maintained usual care and were asked to continue their usual leisure time activities. They were offered the possibility of participating in two sessions consisting of 1 hour of light stretching and relaxation during the 4-month intervention period.

Statistical analyses

The main outcome variables were analysed according to the intention-to-treat analysis principle. Changes in all outcomes were analysed using the bootstrap type analysis of covariance (ANCOVA); confidence interval were obtained by bias-corrected bootstrapping (5000 replications) due to violation of distributions assumptions. T2 was adjusted for baseline value, height and weight and dGEMRIC index was adjusted for baseline value only. Secondary outcomes were adjusted for baseline value. There are multiple endpoints in this study, and results have to be viewed with certain provisos. All p-values and confidence intervals are quoted, rather than introducing the problems and potential errors associated with formal adjustments for potential multiplicity issues. Between-group changes in all outcomes are reported in text as mean difference (95% confidence interval, adjusted p-value). Effect size (*d*) was calculated by using the method of Cohen[36] where an effect size of 0.20 is considered small, 0.50 moderate, and 0.80 large. Confidence intervals for the effect sizes

were obtained by bias-corrected bootstrapping (5000 replications). Statistical analyses were performed using statistical software (Stata, release 13.1, StataCorp, College Station, Texas).

Target sample size of 70 (35 per research arm) was required to ensure the power of at least 80% to detect a difference of 40 ms effect in dGEMRIC between the groups at two-side α=0.05. Predicting a dropout rate of about 10% we aimed to recruit at least 80 participants at baseline.

RESULTS

In total 87 participants met the inclusion criteria and were randomised into the aquatic training group (n=43) and control group (n=44) (Figure 1). The demographic and clinical characteristics of both groups were similar at baseline (Table 1).

Table 1 here.

Program Feasibility

Drop-out rate, during the 4-month intervention period, for each group was 2.3% (n=1 per group) (Figure 1). Training compliance was 88% and mean (SD) training frequency was 2.6 (0.5) per week (including dropouts). The average intensity of each training session was RPE 15 (range, 12-17) and average (SD) maximum heart rate was 144 (12) beats per minute. The mean (SD) number of repetitions completed per session with the affected leg was 481 (67), 416 (68) and 387 (58) for barefoot, small fins and large boots, respectively. 70.5% of these repetitions involved full knee active extension and flexion which was mean 134.4 (SD, 5.6) degrees (affected knee) as measured during baseline assessment.

Harms

There were 2 medical consultations (bilateral knee pain and dyspnoea) as a result of the aquatic training. One subject from the control group required a medical consultation for knee pain after the baseline physical performance measures. All three subjects continued their participation in the study and attended follow-up measurements.

Primary outcomes

To ensure accuracy, each MRI image was inspected for quality. One participant was excluded from the study due to corrupted data as a result of excessive movement artefact in T2 images (Figure 1). One complete baseline dGEMRIC index data set was missing due to lost images (at time of imaging). Additionally, from the dGEMRIC index 11 medial compartments had movement artefact, while in the lateral compartment, 14 had artery-flow pulsating artefact, one had movement artefact and one inaccurate location of the slice compared to baseline image. In total 72 and 68 complete dGEMRIC data sets for medial and lateral femoral condyles respectively were available for quantitative analysis.

T2 and dGEMRIC index baseline values, changes, group differences and effect sizes (Cohen's *d*) at the end of the 4-month intervention are given in table 2. There was a significant decrease in both T2, mean difference -1.2ms (95% CI: -2.2 to -0.2, p=0.021) and dGEMRIC index -23ms (-43 to -3, p=0.022) in the training group compared to controls in the full thickness posterior ROI of the medial femoral cartilage. Further, significant decreases in the training group compared to controls were only seen in the deep posterior and not superficial ROI of the medial femoral cartilage, -1.6ms (-3.0 to -0.3, p=0.016), and -26ms (-50 to -3, p=0.030), for T2 and dGEMRIC index respectively (Figure 3).Values for the deep and superficial posterior ROI (Figure 3) can be found from the online supplemental material.

Table 2 here.

Figure 3 here.

Secondary outcomes

Cardiorespiratory fitness VO_2 peak increased 9.8% in the training group and 4.4% in the control group (d=0.58, p=0.010). There were no between group differences in the knee extension or flexion muscle force or in any domains of KOOS (Table 3).

Table 3 here.

Daily physical activity

The total mean (SD) MET-hours per week, including the intervention, were 40 (13) and 26 (16) in the training and control group respectively (p<0.001). No between group differences were seen in MET activity once the intervention activity was removed (p=0.112). There was no significant difference between the groups in physical activity as measured with accelerometers, excluding the intervention. Sedentary behavior accounted for 80% (5.0) or 13,903 (869) MADs of daily activity. The remaining physical activity was divided into slow walking 3166 (821), normal walking 198 (175) and brisk walking jogging and running together 1.7 (1.4) MADs.

DISCUSSION

As far as we know, this is the first study to show a response in the biochemical composition of tibiofemoral cartilage following 4-months of progressive aquatic resistance training in postmenopausal women with mild knee OA. A small significant change was observed in the biochemical composition of the medial posterior femoral cartilage, which is less loaded during activities of daily living[37]. Additionally, the training significantly improved cardiorespiratory capacity but had no significant effect on muscle force and self-reported symptoms.

This is the first study to show concurrent changes in both T2 and dGEMRIC index in an exercise intervention study. However, both MRI techniques have only been previously implemented once in the same study. In the study by Multanen *et al.*[16] we investigated the effects of a land-based impact intervention on the biochemical composition tibiofemoral cartilage in postmenopausal women with mild knee OA. No positive or negative effect was observed with either MRI technique, however, the posterior ROIs were not reported. In this study[16] the degree of knee motion during the land-based intervention was 0-65 degree and therefore the posterior ROI was not directly loaded. Knee flexion of over 90 degrees is required to produce contact between the posterior ROI of the femur and central tibia[38] which was achieved with our intervention at high frequency. Therefore, our results suggest that the chondrocytes in the posterior region of the femoral cartilage in persons with mild knee OA may have a lower threshold for adaption compared to the central and might be more responsive to the high repetition low shear and compressive cyclic forces produced in the aquatic resistance training. In contrast, the chondrocytes in the central region of the femur and tibia cartilage may require a higher or atypical load to stimulate an adaptive response.

Further, the response was limited to the medial femoral cartilage possibly due to anatomical differences. The medial tibial plateau is concave compared to the convex surface of the lateral side, thus on the medial tibiofemoral joint there is greater contact between the cartilage surfaces[39].

After 4-months of aquatic resistance training, T2 in the posterior region of the medial femoral condyle significantly decreased, with no change in the central femur and tibia regions. A decrease in T2 values is indicative of improved integrity and orientation of the collagen fibres and a decrease in hydration of articular cartilage[40,41]. In more detailed analysis we found that decrease in T2 occurred in deep posterior region of medial femoral cartilage which is in line with our previous study[15]. This study[15] showed a similar response in T2 in patella cartilage in women with mild knee OA following a one year intervention. While the intervention was different, the mechanical forces in the patella cartilage during the progressive impact exercises were shear with moderate compression in the patellofemoral joint i.e. forces were not directly compressive as in the tibiofemoral joint[16]. Therefore, our findings support the notion that the collagen-interstitial water environment in the tibiofemoral cartilage may respond to exercise.

We found a corresponding significant decrease in dGEMRIC index in the posterior region of medial femoral cartilage and again more specifically in its deep region. A lower dGEMRIC index is associated with a lower GAG concentration, thus, a decrease in dGEMRIC index may indicate degeneration of cartilage[42,43]. Our results suggest that the aquatic resistance training may have produced a decrease in GAG concentration within the cartilage matrix or faster contrast agent diffusion in to the cartilage through increased permeability of the cartilage surface[44]. These are characteristics of OA progression[4]. These results conflict with

the findings of Roos and Dahlberg[14] who found an increase in the dGEMRIC index following a 4-month neuromuscular training intervention. However, they measured only one ROI from the medial femoral cartilage and dGEMRIC values were not corrected for BMI, also their population was younger people at high risk of developing knee OA following surgery for meniscal injury. Alternatively, in a previous cross sectional study[45], similar associations i.e. lower T2 and dGEMRIC index was seen in the central ROI of the patella cartilage in young people with repetitive patella dislocation[45]. This finding was speculated to be due to a reparative process within the cartilage. Additionally, faster diffusion of the contrast agent into the medial tibiofemoral cartilage after intravenous injection may have been a combined result of improved contrast agent delivery through vascular changes i.e. increased blood flow in the subchondral bone and synovium with possible improvements in lower limb biomechanics. Further, an improved diffusion of the contrast agent could be explained by a decrease in cartilage thickness i.e. reversal of the cartilage swelling characterised in early OA[4,46]. Cartilage thickness was not measured in our study leaving this issue to speculation and open for further investigation in the future. Therefore, we could hypothesise that while our results indicate the integrity of the collagen-interstitial water environment may be responsive to shear/compressive forces during aquatic exercise, further research is required to understand the exact nature of acute responses in dGEMRIC index to this type of loading.

In line with the findings of our recent systematic review[6], we did not see a significant change in muscle force. However, we used isometric muscle testing whereas, the muscle contraction during isokinetic strength testing mimics closer the true muscle work performed during aquatic resistance training and could have been more sensitive to change. The improvements in cardiorespiratory fitness are in line with other studies investigating the effects of aerobic aquatic training[47]. An aquatic exercise program which also includes neuromuscular exercises

e.g. partial weight bearing exercises might produce better improvements in neuromuscular performance and possibly stimulate GAG production[14] as it is possible to speculate that the loading mechanics in our study may have been ineffective for this purpose.There were no between group changes in any of the domains of the KOOS, this lack of significance is not a surprise given the high values reported at baseline. In combination with the results from the measures of physical performance and KOOS there is no indication that aquatic exercises had a harmful effect on clinical findings in this population. Therefore, aquatic resistance training of sufficient intensity to improve cardiorespiratory function is well tolerated, has high compliance and does not increase pain in women with mild knee OA. Further research should focus on the efficacy of aquatic resistance training for people with more severe stages of OA progression.

The strengths of this study include the high adherence to a highly intensive aquatic training program. This study fulfilled all the important quality criteria of an RCT, except for blinding the participants to exercise therapy, which is common in exercise therapy studies[48]. Strict imaging procedure and segmentation rules ensured good stability and repeatability of the T2 and dGEMRIC indices. This limits, but does not rule out, the possibility that the results of this study are affected by the magic angle (particularly T2) and partial volume effects. The long imaging time in dGEMRIC mapping might result in motion artefact which was controlled for in our study by using a motion correction technique built into the in-house software, as well as strict inclusion/exclusion criteria for image quality. Minor limitations include: MRI imaging performed with a 1.5 tesla scanner, whereas a 3.0 tesla scanner would have produced better spatial resolution and higher signal-to-noise ratio. The mean changes seen in T2 and dGEMRIC index fall within the upper limits of our measurement error for both techniques therefore we cannot exclude measurement error as a possible explanation for

our findings. Further, this study had multiple endpoints and therefore results have to be viewed with caution. In some cases, occasionally thinned and deteriorated cartilage and movement or pulsating artery artefact prevented reliable segmentation of cartilage resulting in lost data. Also, we used single-slice segmenting method assessing articular cartilage, whereas multi-slice method might have produced more a comprehensive view of the knee cartilage. The MRI analysis application divided cartilage to deep and superficial compartments (50%/50%) and due to the 1.5T scanner used, segmented cartilage thickness was from two to five voxels reducing the spatial accuracy and therefore care should be taken when interpreting these results. Pre-contrast T1 imaging was not used in this study however its importance has been questioned and it is felt this omission does not affect our conclusions[46,49]. Classification of OA severity was performed using a combination of pain and Kellgren-Lawrence classification (weight-bearing) and therefore it was not possible to differentiate between healthy and biomechanically altered cartilage between ROIs and condyles[50]. It is still unknown if an aquatic training program of longer than 4-months would have created a global response throughout the cartilage. It is plausible to hypothesise that as cartilage health in one ROI improves it may cause a positive response in adjacent ROI's. Due to the strict inclusion criteria, our results cannot be directly applied to people with later stage OA, older or obese women and men. Finally, the authors acknowledge that the different qMRI parameters and their interactions are not yet fully understood and further investigations about the interaction between exercise and these parameters are warranted.

Our results suggest that, in postmenopausal women with mild knee OA, the integrity of the collagen-interstitial water environment (T2) of the tibiofemoral cartilage may be responsive to low shear and compressive forces during aquatic resistance training. Further research is required to understand the exact nature of acute responses in dGEMRIC index to this type of

loading. Clinical relevance of our findings remains unclear but strongly warrants further research. Additionally, aquatic resistance training of sufficient intensity to improve cardiorespiratory function is well tolerated, has high compliance and low risk of harm amongst women with mild knee OA.

Author contributions

Munukka, Matti: Analysis and interpretation of the data, drafting of the article, critical revision of the article for important intellectual content, final approval of the article, obtaining of funding, collection and assembly of data.
Waller, Benjamin: Analysis and interpretation of the data, drafting of the article, critical revision of the article for important intellectual content, final approval of the article, obtaining of funding, collection and assembly of data.
Rantalainen, Timo: Analysis and interpretation of the data, critical revision of the article for important intellectual content, final approval of the article, administrative, technical, or logistic support, collection and assembly of data.
Häkkinen, Arja: Conception and design, analysis and interpretation of the data, drafting of the article, critical revision of the article for important intellectual content, final approval of the article.
Nieminen, Miika : Conception and design, analysis and interpretation of the data, critical revision of the article for important intellectual content, final approval of the article, administrative, technical, or logistic support.

Lammentausta, Eveliina: Conception and design, analysis and interpretation of the data, critical revision of the article for important intellectual content, final approval of the article, administrative, technical, or logistic support.

Kujala, Urho: Conception and design, analysis and interpretation of the data, drafting of the article, critical revision of the article for important intellectual content, final approval of the article.

Paloneva, Juha: Analysis and interpretation of the data, critical revision of the article for important intellectual content, final approval of the article.

Sipilä, Sarianna: Conception and design, analysis and interpretation of the data, critical revision of the article for important intellectual content, final approval of the article.

Peuna, Arttu: Analysis and interpretation of the data, drafting of the article, critical revision of the article for important intellectual content, final approval of the article, administrative, technical, or logistic support, collection and assembly of data.

Kautiainen, Hannu: Conception and design, analysis and interpretation of the data, drafting of the article, critical revision of the article for important intellectual content, final approval of the article, statistical expertise, collection and assembly of data.

Selänne, Harri: Conception and design, analysis and interpretation of the data, critical revision of the article for important intellectual content, final approval of the article.

Kiviranta, Ilkka: Conception and design, analysis and interpretation of the data, critical revision of the article for important intellectual content, final approval of the article.

Heinonen, Ari: Conception and design, analysis and interpretation of the data, drafting of the article, critical revision of the article for important intellectual content, final approval of the article, obtaining of funding.

Role of funding sources

Financial supporters were: Academy of Finland (ref: 253198), The Social Insurance Institution of Finland (KELA) (ref: 34/26/2011), Finnish Cultural Foundation and Yrjö Jahnsson foundation. None of these had a role in study design, collection, analysis and interpretation of data; in the writing of the manuscript; and in the decision to submit the manuscript for publication.

Conflict of interest

There is no conflict of interest for any authors.

517 References

518 1. McAlindon T, Bannuru R, Sullivan M, Arden N, Berenbaum F, Bierma-Zeinstra S, *et al.*
519 OARSI guidelines for the non-surgical management of knee osteoarthritis. Osteoarthritis
520 Cartilage 2014;22:363-88.

521 2. Roos EM and Arden NK. Strategies for the prevention of knee osteoarthritis. Nat Rev
522 Rheumatol 2016;12:92-101. doi: 10.1038/nrrheum.2015.135 [doi].

523 3. Goldring MB and Goldring SR. Articular cartilage and subchondral bone in the
524 pathogenesis of osteoarthritis. Ann N Y Acad Sci 2010;1192:230-7.

525 4. Arokoski J, Jurvelin J, Väätäinen U, Helminen H. Normal and pathological adaptations of
526 articular cartilage to joint loading. Scand J Med Sci Sports 2000;10:186-98.

527 5. Fransen M, McConnell S, Harmer AR, Van der Esch M, Simic M, Bennell KL. Exercise
528 for osteoarthritis of the knee. Br J Sports Med 2015;10.1136/bjsports-2015-095424: doi:
529 10.1002/14651858.CD004376.pub3 [doi].

530 6. Waller B, Ogonowska-Slodownik A, Vitor M, Lambeck J, Daly D, Kujala UM, *et al*.
531 Effect of therapeutic aquatic exercise on symptoms and function associated with lower limb
532 osteoarthritis: systematic review with meta-analysis. Phys Ther 2014;94:1383-95. doi:
533 10.2522/ptj.20130417 [doi].

534 7. Kiviranta I, Tammi M, Jurvelin J, Saamanen AM, Helminen HJ. Moderate running
535 exercise augments glycosaminoglycans and thickness of articular cartilage in the knee joint of
536 young beagle dogs. J Orthop Res 1988;6:188-95. doi: 10.1002/jor.1100060205 [doi].

537 8. Kiviranta I, Tammi M, Jurvelin J, Arokoski J, Saamanen AM, Helminen HJ. Articular
538 cartilage thickness and glycosaminoglycan distribution in the canine knee joint after
539 strenuous running exercise. Clin Orthop Relat Res 1992;302-8.

540 9. Teichtahl AJ, Wluka AE, Forbes A, Wang Y, English DR, Giles GG, *et al*. Longitudinal
541 effect of vigorous physical activity on patella cartilage morphology in people without clinical
542 knee disease. Arthritis Rheum 2009;61:1095-102.

543 10. Tiderius CJ, Svensson J, Leander P, Ola T, Dahlberg L. dGEMRIC (delayed gadolinium-
544 enhanced MRI of cartilage) indicates adaptive capacity of human knee cartilage. Magn Reson
545 Med 2004;51:286-90.

546 11. Kiviranta I, Tammi M, Jurvelin J, Arokoski J, Säämänen A, Helminen HJ. Articular
547 cartilage thickness and glycosaminoglycan distribution in the young canine knee joint after
548 remobilization of the immobilized limb. J Orthop Res 1994;12:161-7.

549 12. Souza RB, Baum T, Wu S, Feeley BT, Kadel N, Li X, *et al*. Effects of unloading on knee
550 articular cartilage T1rho and T2 magnetic resonance imaging relaxation times: a case series. J
551 Orthop Sports Phys Ther 2012;42:511-20.

552 13. Brismar BH, Lei W, Hjerpe A, Svensson O. The effect of body mass and physical activity
553 on the development of guinea pig osteoarthrosis. Acta Orthop Scand 2003;74:442-8.

554 14. Roos EM and Dahlberg L. Positive effects of moderate exercise on glycosaminoglycan
555 content in knee cartilage: a four-month, randomized, controlled trial in patients at risk of
556 osteoarthritis. Arthritis Rheum. 2005;52:3507-14. doi: 10.1002/art.21415 [doi].

15. Koli J, Multanen J, Kujala UM, Häkkinen A, Nieminen MT, Kautiainen H, *et al*. Effect of Exercise on Patellar Cartilage in Women with Mild Knee Osteoarthritis. Med Sci Sports Exerc 2015;47:1767-74. doi: 10.1249/MSS.0000000000000629 [doi].

16. Multanen J, Nieminen MT, Häkkinen A, Kujala UM, Jämsä T, Kautiainen H, *et al*. Effects of High-Impact Training on Bone and Articular Cartilage: 12-Month Randomized Controlled Quantitative MRI Study. J Bone Miner Res 2014;29:192-201.

17. Hawezi ZK, Lammentausta E, Svensson J, Roos EM, Dahlberg LE, Tiderius CJ. Regional dGEMRIC analysis in patients at risk of osteoarthritis provides additional information about activity related changes in cartilage structure. Acta Radiol. 2015;0284185115591237: doi: 0284185115591237 [pii].

18. Wang Y, Wei L, Zeng L, He D, Wei X. Nutrition and degeneration of articular cartilage. Knee Surg Sports Traumatol Arthrosc 2013;21:1751-62.

19. Holla JF, Sanchez-Ramirez DC, van der Leeden M, Ket JC, Roorda LD, Lems WF, *et al*. The avoidance model in knee and hip osteoarthritis: a systematic review of the evidence. J Behav Med 2014;37:1226-41.

20. Roper JA, Bressel E, Tillman MD. Acute aquatic treadmill exercise improves gait and pain in people with knee osteoarthritis. Arch Phys Med Rehabil 2013;94:419-25. doi: 10.1016/j.apmr.2012.10.027 [doi].

21. Pöyhönen T, Sipilä S, Keskinen KL, Hautala A, Savolainen J, Mälkiä E. Effects of aquatic resistance training on neuromuscular performance in healthy women. Med Sci Sports Exerc 2002;34:2103-9. doi: 10.1249/01.MSS.0000039291.46836.86 [doi].

578 22. Valtonen A, Pöyhönen T, Sipilä S, Heinonen A. Effects of aquatic resistance training on
579 mobility limitation and lower-limb impairments after knee replacement. Arch Phys Med
580 Rehabil 2010;91:833-9. doi: 10.1016/j.apmr.2010.03.002 [doi].

581 23. Waller B, Munukka M, Multanen J, Rantalainen T, Pöyhönen T, Nieminen MT, *et al*.
582 Effects of a progressive aquatic resistance exercise program on the biochemical composition
583 and morphology of cartilage in women with mild knee osteoarthritis: protocol for a
584 randomised controlled trial. BMC Musculoskelet.Disord. 2013;14:10.1186/1471-2474-14-82.
585 doi: 10.1186/1471-2474-14-82 [doi].

586 24. Kellgren JH and Lawrence JS. Radiological assessment of osteo-arthrosis. Ann Rheum
587 Dis 1957;16:494-502.

588 25. Kanis JA, McCloskey EV, Johansson H, Oden A, Melton LJ, Khaltaev N. A reference
589 standard for the description of osteoporosis. Bone 2008;42:467-75.

590 26. Ainsworth BE, Haskell WL, Herrmann SD, Meckes N, Bassett DR,Jr, Tudor-Locke C, *et*
591 *al*. 2011 Compendium of Physical Activities: a second update of codes and MET values. Med
592 Sci Sports Exerc 2011;43:1575-81. doi: 10.1249/MSS.0b013e31821ece12 [doi].

593 27. Tiderius C, Hori M, Williams A, Sharma L, Prasad PV, Finnell M, *et al*. dGEMRIC as a
594 function of BMI. Osteoarthritis Cartilage 2006;14:1091-7. doi: S1063-4584(06)00154-3 [pii].

595 28. Multanen J, Rauvala E, Lammentausta E, Ojala R, Kiviranta I, Häkkinen A, *et al*.
596 Reproducibility of imaging human knee cartilage by delayed gadolinium-enhanced MRI of
597 cartilage (dGEMRIC) at 1.5 Tesla. Osteoarthritis Cartilage 2009;17:559-64.

598 29. Laukkanen R, Oja R, Pasanen M, Vuori I. Criterion validity of a two-kilometer walking
599 test for predicting the maximal oxygen uptake of moderately to highly active middle-aged
600 adults. Scand J Med Sci Sports 1993;3:267-72.

601 30. Sipilä S, Multanen J, Kallinen M, Era P, Suominen H. Effects of strength and endurance
602 training on isometric muscle strength and walking speed in elderly women. Acta Physiol
603 Scand 1996;156:457-64. doi: 10.1046/j.1365-201X.1996.461177000.x [doi].

604 31. Koli J, Multanen J, Häkkinen A, Kiviranta I, Kujala U, Heinonen A. Reliability of the
605 Finnish versions of WOMAC and KOOS forms for knee osteoarthritis. Physiotherapy
606 2011;97:D203-4.

607 32. Roos EM, Roos H, Ekdahl C, Lohmander L. Knee injury and Osteoarthritis Outcome
608 Score (KOOS)□validation of a Swedish version. Scand J Med Sci Sports 1998;8:439-48.

609 33. Vähä-Ypyä H, Vasankari T, Husu P, Suni J, Sievänen H. A universal, accurate intensity-
610 based classification of different physical activities using raw data of accelerometer. Clin
611 Physiol Funct Imaging 2015;35:64-70.

612 34. Pöyhönen T, Keskinen KL, Kyröläinen H, Hautala A, Savolainen J, Mälkiä E.
613 Neuromuscular function during therapeutic knee exercise under water and on dry land. Arch
614 Phys Med Rehabil 2001;82:1446-52.

615 35. Borg GA. Psychophysical bases of perceived exertion. Med Sci Sports Exerc
616 1982;14:377-81.

617 36. Faraone SV. Interpreting estimates of treatment effects: implications for managed care. P
618 T 2008;33:700-11.

619 37. Walker CR, Myles C, Nutton R, Rowe P. Movement of the knee in osteoarthritis. The use
620 of electrogoniometry to assess function. J Bone Joint Surg Br 2001;83:195-8.

621 38. Goyal K, Tashman S, Wang JH, Li K, Zhang X, Harner C. In vivo analysis of the isolated
622 posterior cruciate ligament-deficient knee during functional activities. Am J Sports Med
623 2012;40:777-85. doi: 10.1177/0363546511435783 [doi].

624 39. Neumann DA. Kinesiology of the musculoskeletal system: foundations for physical
625 rehabilitation. St. Louis: USA: Mosby 2002: 435-476.

626 40. Lammentausta E, Kiviranta P, Nissi M, Laasanen M, Kiviranta I, Nieminen M, *et al*. T2
627 relaxation time and delayed gadolinium-enhanced MRI of cartilage (dGEMRIC) of human
628 patellar cartilage at 1.5 T and 9.4 T: Relationships with tissue mechanical properties. J
629 Orthop Res 2006;24:366-74.

630 41. Nieminen MT, Rieppo J, Töyräs J, Hakumäki JM, Silvennoinen J, Hyttinen MM, *et al*.
631 T2 relaxation reveals spatial collagen architecture in articular cartilage: a comparative
632 quantitative MRI and polarized light microscopic study. Magn Reson Med 2001;46:487-93.

633 42. Bashir A, Gray M, Hartke J, Burstein D. Nondestructive imaging of human cartilage
634 glycosaminoglycan concentration by MRI. Magn Reson Med 1999;41:857-65.

635 43. Nissi M, Töyräs J, Laasanen M, Rieppo J, Saarakkala S, Lappalainen R, *et al*.
636 Proteoglycan and collagen sensitive MRI evaluation of normal and degenerated articular
637 cartilage. J Orthop Res 2004;22:557-64.

638 44. Li W, Scheidegger R, Wu Y, Edelman RR, Farley M, Krishnan N, *et al*. Delayed
639 contrast-enhanced MRI of cartilage: Comparison of nonionic and ionic contrast agents. Magn
640 Reson Med 2010;64:1267-73.

641 45. Bengtsson Moström E, Lammentausta E, Finnbogason T, Weidenhielm L, Janarv P,
642 Tiderius CJ. Pre-and postcontrast T1 and T2 mapping of patellar cartilage in young adults
643 with recurrent patellar dislocation. Magn Reson Med 2014;74:1363-9.

644 46. Hawezi ZK, Lammentausta E, Svensson J, Dahlberg LE, Tiderius CJ. In vivo transport of
645 Gd-DTPA2− in human knee cartilage assessed by depth-wise dGEMRIC analysis. J Magn
646 Reson Imaging 2011;34:1352-8.

647 47. Lim J, Tchai E, Jang S. Effectiveness of aquatic exercise for obese patients with knee
648 osteoarthritis: a randomized controlled trial. PM R 2010;2:723-31.

649 48. Furlan AD, Pennick V, Bombardier C, van Tulder M, Editorial Board, Cochrane Back
650 Review Group. 2009 updated method guidelines for systematic reviews in the Cochrane Back
651 Review Group. Spine 2009;34:1929-41. doi: 10.1097/BRS.0b013e3181b1c99f [doi].

652 49. Li W, Du H, Scheidegger R, Wu Y, Prasad PV. Value of precontrast T(1) for dGEMRIC
653 of native articular cartilage. J Magn Reson Imaging 2009;29:494-7. doi: 10.1002/jmri.21658
654 [doi].

655 50. Multanen J, Heinonen A, Hakkinen A, Kautiainen H, Kujala UM, Lammentausta E, *et al*.
656 Bone and cartilage characteristics in postmenopausal women with mild knee radiographic
657 osteoarthritis and those without radiographic osteoarthritis. J Musculoskelet Neuronal Interact
658 2015;15:69-77.

659

Tables

Table 1. Baseline demographic and clinical characteristics of the participants

	Exercise group (n=43)	Control group (n=44)
Age (years)	64 (2)	64 (2)
Height (cm)	162 (5)	162 (5)
Body mass (kg)	69.6 (10.3)	71.0 (11.3)
Body mass index (kg/m^2)	26.6 (3.8)	27.1 (3.5)
Time from menopause (years)	14 (6)	14 (6)
Pain killers for knee pain, *n* (%) of users	11 (25.6)	9 (20.5)
Glucosamine use occasionally, *n* (%)	12 (28)	8 (18)
Kellgren Lawrence grade, *n* (%)		
Grade 1	23 (53.5)	24 (54.5)
Grade 2	20 (46.5)	20 (45.5)
Knee pain during last week, (VAS, mm)[a]		
• Affected leg	28 (25)	24 (19)
• Non-affected leg	24 (19)	23 (18)
Habitual physical activity (METh/week)	29 (31)	36 (33)

Values are means (SD) or n (%)
METh = metabolic equivalent task hour.
[a] Range, 0-100 mm

Table 2. Effects of aquatic training on T2 relaxation time and dGEMRIC index in full-thickness ROIs.

	Baseline, mean (SD)		Change to month 4, mean (95% CI)		Effect Size (95% CI)	P-value	
	Training (n=42)	Controls (n=42)	Training (n=42)	Controls (n=42)		Crude	Adjusted
T2, ms							
Femur ‡							
Lateral condyle							
Central	52.6 (4.9)	53.4 (4.1)	-0.18 (-1.05 to 0.59)	-0.03 (-0.95 to 0.91)	0.05 (-0.38 to 0.48)	0.81	0.58[a]
Posterior	49.6 (4.6)	48.8 (3.6)	-0.23 (-1.26 to 0.97)	0.74 (0.01 to 1.40)	0.31 (-0.12 to 0.74)	0.15	0.30[a]
Medial condyle							
Central	52.8 (4.5)	52.0 (4.4)	-0.20 (-1.17 to 0.83)	0.48 (-0.71 to 1.68)	0.18 (-0.25 to 0.61)	0.40	0.47[a]
Posterior	52.0 (4.7)	51.9 (4.5)	-1.16 (-1.85 to -0.50)	0.10 (-0.72 to 0.94)	0.48 (0.05 to 0.91)	0.028	0.021[a]
Tibia							
Lateral plateau							
Central	41.0 (8.3)	42.9 (8.1)	-0.66 (-1.86 to 0.45)	0.05 (-1.67 to 1.68)	0.15 (-0.28 to 0.58)	0.50	0.30[a]
Medial plateau							
Central	44.5 (5.0)	42.7 (4.2)	-0.02 (-1.45 to 1.41)	-0.02 (-0.85 to 0.79)	-0.00 (-0.43 to 0.43)	1.00	0.41[a]
dGEMRIC*, ms							
Femur							
Lateral condyle [+]							
Central	433 (70)	424 (44)	-4 (-16 to 7)	-1 (-11 to 8)	0.07 (-0.40 to 0.55)	0.77	0.93[b]
Posterior	422 (60)	428 (57)	2 (-10 to 16)	6 (-8 to 22)	0.08 (-0.40 to 0.57)	0.72	0.71[b]
Medial condyle [§]							
Central	411 (61)	410 (65)	-19 (-32 to -6)	-6 (-17 to 7)	0.34 (-0.12 to 0.80)	0.14	0.13[b]
Posterior	453 (60)	448 (61)	-23 (-39 to -8)	1 (-14 to 16)	0.53 (0.06 to 0.99)	0.022	0.022[b]
Tibia							
Lateral plateau							
Central	424 (76)	419 (82)	-1 (-20 to 17)	1 (-15 to 17)	0.03 (-0.46 to 0.52)	0.91	0.95[b]
Medial plateau							
Central	382 (75)	386 (50)	-20 (-35 to -6)	-6 (-20 to 8)	0.32 (-0.15 to 0.78)	0.19	0.09[b]

669 T2 = transverse relaxation time; [a]ANCOVA: adjusted for baseline value, height and weight.
670 dGEMRIC = delayed gadolinium-enhanced magnetic resonance imaging of cartilage; [b]ANCOVA: adjusted for baseline value.
671 In T2 low values correspond to improved integrity and orientation of the collagen fibres and a decrease in hydration of articular cartilage.
672 In dGEMRIC, high values correspond to high glycosaminoglycan concentration.
673 *Missing data for dGEMRIC [+]n=16, [§]n=12

Table 3. Effects of aquatic training on physical performance and clinical symptoms

	Baseline, mean (SD)		Change to month 4, mean (95% CI)		Effect Size (95% CI)	P-value	
	Training (n=42)	Controls (n=42)	Training (n=42)	Controls (n=42)		Crude	Adjusted[a]
Cardiorespiratory fitness (ml/kg/min)							
Estimated VO_2 peak	24.6 (5.6)	24.9 (4.9)	2.4 (1.8 to 3.1)	1.1 (0.5 to 1.8)	0.58 (0.15 to 1.06)	0.006	0.010
Force (N)							
Extension	335 (64)	343 (70)	20 (8 to 33)	9 (-5 to 23)	0.27 (-0.17 to 0.70)	0.22	0.28
Flexion	164 (52)	165 (40)	20 (9 to 30)	17 (7 to 27)	0.07 (-0.36 to 0.51)	0.74	0.71
KOOS(0-100)							
Pain	80 (10)	82 (12)	4 (1 to 7)	1 (-2 to 4)	0.30 (-0.14 to 0.74)	0.17	0.25
Other symptoms	74 (13)	75 (14)	7 (3 to 10)	2 (-1 to 6)	0.37 (-0.07 to 0.81)	0.09	0.09
ADL	84 (10)	85 (11)	4 (1 to 7)	0 (-2 to 3)	0.39 (-0.03 to 0.78)	0.08	0.10
Sport	63 (20)	65 (22)	8 (2 to 14)	3 (-3 to 8)	0.27 (-0.16 to 0.73)	0.21	0.24
QOL	65 (17)	71 (20)	7 (3 to 11)	3 (-1 to 8)	0.26 (-0.18 to 0.71)	0.24	0.38

677 ADL= activites of daily living; Sport = sports and recreation; QOL = knee related quality of life, KOOS = Knee injury and osteoarthirtis
678 outcome score
679 [a]ANCOVA: adjusted for baseline

FIGURE LEGENDS

Figure 1. Flow chart showing enrolment, allocation and four month end measurements.

Figure 2. Illustration of the region of interests (ROIs) in the full-thickness femoral and tibial cartilage. Midlines split both femoral and tibial cartilage into superficial and deep sections.

Figure 3. Magnitude of effect (Cohen's *d* and 95% CI) at superficial and deep layers of T2 and dGEMRIC cartilage ROIs from medial and lateral condyles. T2 = transverse relaxation time; dGEMRIC = delayed gadolinium-enhanced magnetic resonance imaging of cartilage; sPF = superficial posterior femur; dPF = deep posterior femur.