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NASHA hyaluronic acid vs methylprednisolone for knee osteoarthritis: a prospective, multi-centre, randomized, non-inferiority trial

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SUMMARY

Objective: To compare NASHA hyaluronic acid gel as single-injection intra-articular (IA) treatment for knee osteoarthritis (OA) against methylprednisolone acetate (MPA).
Design: This was a prospective, multi-centre, randomized, active-controlled, double-blind, non-inferiority clinical trial. A unique, open-label extension phase (OLE) was undertaken to answer further important clinical questions. Subjects with painful unilateral knee OA were treated and followed for 26 weeks (blinded phase). All patients attending the clinic at 26 weeks were offered NASHA treatment, with a subsequent 26-week follow-up period (extension phase). The primary objective was to show non-inferiority of NASHA vs MPA in Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) pain responder rate (percentage of patients with ≥40% improvement from baseline in WOMAC pain score and an absolute improvement of ≥5 points) at 12 weeks.
Results: In total, 442 participants were enrolled. The primary objective was met, with NASHA producing a non-inferior response rate vs MPA at 12 weeks (NASHA: 44.6%; MPA: 46.2%; difference [95% CI]: 1.6% [−11.2%; +7.9%]). Effect size for WOMAC pain, physical function and stiffness scores favoured NASHA over MPA from 12 to 26 weeks. In response to NASHA treatment at 26 weeks, sustained improvements were seen in WOMAC outcomes irrespective of initial treatment. No serious device-related adverse events (AEs) were reported.
Conclusions: This study shows that single-injection NASHA was well tolerated and non-inferior to MPA at 12 weeks. The benefit of NASHA was maintained to 26 weeks while that of MPA declined. An injection of NASHA at 26 weeks conferred long-term improvements without increased sensitivity or risk of complications.
Study identifier: NCT01209364 (www.clinicaltrials.gov).

Introduction

Intra-articular hyaluronic acid (IA HA) is an established treatment for knee osteoarthritis (OA), offering a useful option that lies between analgesic treatment and joint replacement. The options for pharmacological analgesic therapy include simple analgesics, non-steroidal anti-inflammatory drugs (NSAIDs) and cyclo-oxygenase type 2 (COX-2) inhibitors. However, these treatments are associated with potential safety concerns[1–4]. IA corticosteroids, another treatment option for knee OA, are believed to have a faster onset of action than HA but a shorter duration of effect[5,6], and repeated use of corticosteroids may be associated with increased side effects[7,8].

NASHA is a unique HA product whose production process begins with bacterial synthesis of HA. This is followed by purification and then a carefully controlled stabilization process involving a small degree of molecular cross-linking (~1%)[9]. The resulting NASHA gel

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retains the biocompatibility of HA, but it has a high molecular mass and a prolonged IA residence time: the half-life in both the human knee and a pre-clinical model is around 4 weeks[10,11].

NASHA is administered as a single IA injection. Previous studies in knee OA have demonstrated that NASHA provides significant post-treatment benefits, lasting for at least 6 months[12,13]. NASHA has also been shown to provide similar benefit to the corticosteroid triamcinolone acetonide (TA) in a 12-week study[14].

The corticosteroid methylprednisolone acetate (MPA), commonly used for IA treatment of OA[15], has lower solubility than unmodified methyprednisolone[16]. Few high-quality clinical studies have compared the effects of different corticosteroid preparations in knee OA. However, there is evidence that MPA may have a longer duration of effect than other corticosteroids[17].

We performed a 26-week study to compare the effectiveness and safety of a single IA injection of NASHA with MPA, which may be considered as a standard-of-care treatment. The study hypothesis was that NASHA is at least as effective as MPA for the treatment of knee OA. A non-inferiority study design was chosen because the only previous comparison of NASHA with IA corticosteroid (study duration: 12 weeks), showed lack of statistically significant efficacy differences between the two treatments. Although IA corticosteroids may be perceived as having a short duration of action (4 weeks or less)[6], there is evidence that they can provide symptomatic improvement for 16–24 weeks[18]. At the end of the double-blind comparison, all study participants were offered unblinded NASHA treatment.

Methods

Study design

This was a prospective, multi-centre, randomized (1:1), corticosteroid-controlled, double-blind, non-inferiority clinical study with an open-label extension phase (OLE), performed in patients with knee OA. The initial blinded phase included a 26-week follow-up period. This was followed by unblinded NASHA treatment and a further 26-week follow-up period (OLE). The study was performed in accordance with the Declaration of Helsinki, the International Conference on Harmonization Guidelines for Good Clinical Practice, and applicable regulatory requirements. The results are presented in line with guidelines from Consolidated Standards of Reporting Trials (CONSORT)[19] and International Committee of Medical Journal Editors (ICMJE)[20]. The study protocol was reviewed and approved by Independent Ethics Committees/Institutional Review Boards at each participating site. Signed informed consent was obtained from each patient.

Study objectives

The primary objective was to determine whether NASHA is non-inferior to MPA in Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) pain responder rate (defined as an improvement from baseline of at least 40% and an absolute improvement of five points), at 12 weeks post-treatment. A pre-defined margin for non-inferiority was implemented (lower bound of the two-sided 95% confidence interval (CI) for between-group difference above −15%). Secondary objectives were to assess the safety and effectiveness of NASHA compared with MPA, during both the blinded phase and the OLE.

Participants

Men and women aged 35–80 with a body mass index of $\leq$40 kg/m^2, the ability to walk 50 m unaided, unilateral knee pain meeting the American College of Rheumatology criteria for the diagnosis of OA[21], WOMAC pain score of 7–17 in the study knee, and radiographically verified OA of the study knee (Kellgren–Lawrence grade II or III; severity consistent with several trials demonstrating the efficacy of IA corticosteroids[6]), were recruited for the study. Key exclusion criteria were: clinically detectable knee effusion, clinically significant contralateral knee OA (WOMAC pain score >3), clinically significant pain in joints other than the knee, IA steroid injection into the study knee within the preceding 3 months, IA HA injection into the study knee within the preceding 9 months, use of systemic glucocorticosteroids (excluding inhaled steroids) within the preceding 3 months and arthroscopy or other surgical procedure in the study knee within the preceding 12 months. Study participants were enrolled at each centre by staff other than the treating investigator (usually it was the evaluating investigator). A total of 442 participants were enrolled at sites in Canada (15 sites, 284 participants), UK (four sites, 36 participants) and Sweden (five sites, 122 participants). The first patient was screened on March 27, 2007 and the study was stopped when the last patient completed the 52-week visit on November 12, 2008.

Study treatments

For the blinded phase, eligible patients were randomized 1:1 to receive a single IA injection of either NASHA (Q-Med AB, Uppsala, Sweden; DUROLANE 60 mg in 3 mL) or MPA (1 mL, 40 mg; dose consistent with several trials demonstrating the efficacy of MPA[6]). The randomization scheme was computer-generated by a contract research organisation using a block-size of 4 stratified by site. Identities of both the study and reference product were concealed by packaging them in identical boxes labelled with the subject number. Patient numbers were assigned sequentially as subjects were randomized at each site. Prior to IA injection, synovial fluid was aspirated as needed and IA injection of lidocaine (2 mL, 1% solution) was performed. IA injection was performed using one of three techniques without fluoroscopic or ultrasound guidance: lateral upper-patellar, lateral mid-patellar or medial IA. The treating investigator opened the treatment package before treatment, but kept the identity of the treatment concealed from other study staff (e.g., evaluating investigator). Blinding of study participants was ensured by using drapes to prevent them from viewing the injection procedure. The treating investigator was allowed to perform some of the screening assessments, as well as all of the injection-related procedures, but they were not allowed to perform any effectiveness evaluations or assessments after the patient had been randomized and treated. The assessment schedule consisted of a screening visit (Visit 1), a baseline visit during which the IA injections were administered (Visit 2), telephone calls at 2 and 4 weeks post-injection to evaluate safety and concomitant medication use, and clinic visits at 6, 12, 18 and 26 weeks post-injection to conduct effectiveness and safety assessments (Visits 3–6). Rescue medication with acetaminophen was allowed at up to 3 g per day.

The OLE was uniquely designed to answer two questions: can one injection cause sensitivity to the second injection, and what are the benefits of a second injection? Subjects attending the 26-week visit were offered a single, unblinded IA injection of NASHA, administered using the same method as in the blinded phase. Clinic visits for the OLE were scheduled at 28 weeks, 39 weeks and 52 weeks after the initial injection. The patient, treating investigator and assessing investigator remained unaware of which treatment was administered at the first injection. Patients not requesting a second injection were also followed to 52 weeks if their WOMAC pain score was improved from baseline at the 26-week visit.

Effectiveness assessments

The WOMAC index (Likert format, version 3.1) was administered with three domains: pain (five items, score 0–20), stiffness (two items, score 0–8) and physical function (17 items, score 0–68); items were assessed for the preceding 48 h. Patients' assessment of global status was recorded on a five-point scale. Physical activity was assessed by each patient at baseline using a four-point scale; subsequent assessments were made relative to baseline using a five-point scale. Two additional functional assessments were performed. Firstly, for 'Get up and go', the patient was timed while rising from a chair, walking four paces, returning, and sitting down again. Secondly, for '10-m timed walk', the patient was timed while walking 10 m. Rescue medication was recorded as the number of tablets taken since the last study visit and the number of tablets taken during the preceding 48 h.

WOMAC pain responder rate was defined as an improvement in WOMAC pain score of at least 40% vs baseline and an absolute improvement of at least five points[22]. Patients were classified as an OMERACT-OARSI responder (proposition D)[23] if they achieved a 50% reduction and an absolute improvement of 4 (for WOMAC pain) or 14 (for WOMAC physical function), or if they demonstrated improvement in two out of three parameters as follows: a 20% reduction and an absolute improvement of 2 (WOMAC pain), 7 (WOMAC physical function) or 1 (for patient assessment of global status).

Safety analysis

The number and percentage of patients who reported adverse events (AEs) was summarized using MedDRA terminology. AEs were also classified by intensity (mild, moderate or severe) and relationship to the study treatment.

Statistical methods

All statistical analyses were performed using SAS® software, with two-sided statistical tests and a significance level of 5%.

Analysis populations and imputation methods

The safety population comprised all subjects who received any randomized study treatment. The primary population for all effectiveness analyses was the 'full analysis population' (FAP) which comprised subjects from the safety population with a baseline and at least one post-treatment WOMAC pain score assessment. The evaluable population (EP) consisted of subjects from the FAP with no major protocol violations and not withdrawn prematurely. Two EPs were defined: one at 12 weeks (EP12) and one at 26 weeks (EP26).

Multiple imputation (MI) was used to predict missing WOMAC pain values for the FAP. Treatment group, prior WOMAC pain scores and all baseline factors significantly related to post-treatment WOMAC pain score (Kellgren–Lawrence score and WOMAC physical function score) were included in the imputation regression model. Last observation carried forward (LOCF) and baseline observation carried forward (BOCF) imputation methods were used for sensitivity purposes, for WOMAC pain score analyses. LOCF was the primary imputation method for all other endpoints. All EP results and data for the OLE were based on observed cases (OC) only.

Primary effectiveness analysis

The primary effectiveness endpoint was WOMAC pain responder rate, an outcome not used in trials demonstrating the efficacy of MPA[6]. Non-inferiority of NASHA to MPA was assessed at 12 weeks using a two-sided 95% CI; the criterion for non-inferiority was the lower bound of the CI for treatment difference (NASHA − MPA) being above −15%.

Secondary effectiveness analyses

The superiority of NASHA to MPA was to be assessed if non-inferiority was demonstrated. Superiority was to be declared if the lower bound of the CI for treatment difference in responder rate was greater than zero. WOMAC pain score was analysed for the FAP with an MI approach.

Further secondary effectiveness outcomes included: WOMAC pain responder rates (at time-points other than 12 weeks), WOMAC pain score, WOMAC physical function score, WOMAC stiffness score, patient assessment of global status, physical activity assessment, functional mobility assessment ('Get up and go' and '10-m timed walk'), OMERACT-OARSI responder rate and use of rescue medication. Effect-sizes were calculated for the three WOMAC domains as product difference in mean change from baseline divided by the pooled standard deviation (SD).

Sample size estimation

The sample size calculation was based on a non-inferiority margin of 15% and an assumed percentage of subjects responding to each treatment of 45%. The number of subjects required for 80% power to show that the two-sided 95% CI of the treatment difference lies above the non-inferiority margin was 180 per treatment group. Assuming a drop-out rate of 20%, a minimum of 432 patients were to be included.

Results

Subjects

442 participants were enrolled into the study; 221 were treated with NASHA and 221 with MPA. At 26 weeks, 342 patients elected to receive open-label NASHA, 163 from the NASHA group and 179 from the MPA group (Fig. 1).

For the blinded phase, more patients in the NASHA group than in the MPA group had synovial fluid aspirated at the time of injection (NASHA: 15.6%; MPA: 8.8%, $P = 0.04$) and there was a statistically significant 2.3 cm difference in the height of female subjects between the two groups (NASHA: 164.8 cm, MPA: 162.5 cm; $P = 0.019$). As shown in Table I, there were no other statistically significant differences in demographic or other baseline characteristics between the two study groups. Also, there were no significant baseline differences between patients who did or did not receive NASHA at 26 weeks.

Effectiveness

Blinded phase (0–26 weeks)

The primary effectiveness objective was met, with WOMAC pain responder rates at 12 weeks in the FAP demonstrating NASHA to be non-inferior to MPA (NASHA: 44.6%; MPA: 46.2%; 95% CI of difference: −11.2%; +7.9%). WOMAC pain responder rates at weeks 6, 12 and 18 remained comparable between NASHA and MPA. Between weeks 18 and 26, the WOMAC pain responder rate remained stable in the NASHA group, while there was a decrease in the MPA group over this period [Fig. 2(A)]. Sensitivity analyses of WOMAC responder rates (LOCF, BOCF, OC, EP) all showed NASHA to be non-inferior to MPA at all time-points [Fig. 2(B)]. Superiority was not demonstrated at any time.

Secondary analysis of WOMAC pain score over the first 26 weeks demonstrated that MPA provided early improvements in pain, with the improvement reaching a maximum at 6 weeks and declining thereafter until 26 weeks. In comparison, NASHA was associated

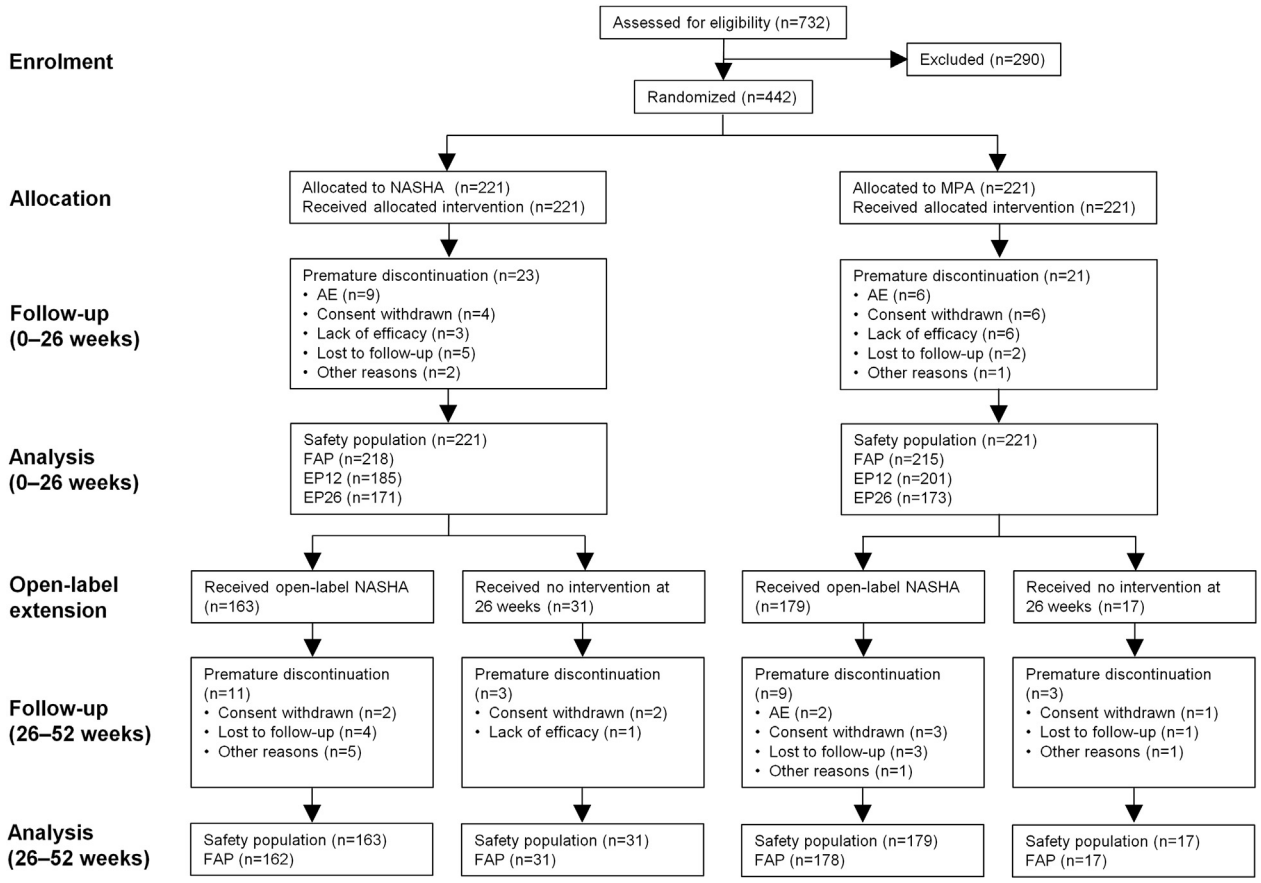


Fig. 1. Patient disposition. EP12/26, evaluable population at 12/26 weeks.

Table I
Patient demographics and baseline characteristics for the blinded phase (FAP)

Variable	NASHA ($n = 218$)	MPA ($n = 215$)
Age in years [mean (SD)]	61.9 (9.6)	61.5 (9.9)
Female:male [n (%)]	111:107 (51:49)	102:113 (47:53)
Height [mean (SD)]		
Female*	164.8 (7.1)	162.5 (7.1)
Male	176.6 (7.5)	177.8 (8.7)
BMI [mean (SD)]	28.2 (4.2)	28.3 (4.1)
Race [n (%)]		
Caucasian	208 (95.4)	208 (96.7)
Black	4 (1.8)	1 (0.5)
Asian	4 (1.8)	5 (2.3)
Hispanic	0 (0)	1 (0.5)
Other	2 (0.9)	0 (0)
Duration of OA in years [mean (SD)]	4.7 (5.4)	4.9 (6.3)
Kellgren–Lawrence score‡		
II [n (%)]	71 (32.6)	85 (39.5)
III [n (%)]	147 (67.4)	130 (60.5)
WOMAC pain score [mean (SD)]‡	10.1 (2.2)	10.0 (2.3)
WOMAC physical function score [mean (SD)]	31.0 (9.7)	29.5 (9.8)
Get-up-and-go test [mean (SD)]	9.9 (2.7)	10.0 (3.3)
Timed 10-m walk test [mean (SD)]	9.8 (3.5)	10.2 (4.5)
Synovial fluid aspirated† [n (%)]	34 (15.6%)	19 (8.8%)
NSAID or analgesics for OA at baseline [n (%)]	139 (63.8)	119 (55.3)
Previous treatment with IA steroids [n (%)]‡	62 (28.4)	60 (27.9)
Previous treatment with IA HA [n (%)]‡	18 (8.3)	27 (12.6)
Previous arthroscopy/knee surgery [n (%)]‡	74 (34.1)	63 (29.3)

* Significant difference was noted between groups ($P = 0.019$, two-sample t-test).
† Fluid aspirated from the study knee immediately before injection of study medication, a significant difference was noted between groups ($P = 0.04$, Fisher's exact test).
‡ Relating to the study knee.

with lower initial reductions, but no substantial decline was observed between 6 and 26 weeks. When analysing WOMAC pain score using the pre-specified repeated measures model, the improvement from baseline was larger for NASHA than MPA at week 26 ($P = 0.034$) [Fig. 2(C)]. As shown in [Fig. 3(A)], the effect size favoured NASHA from 12 to 26 weeks.

Results for the remaining secondary effectiveness variables over the 26-week double-blind follow-up period were similar, with NASHA eliciting greater improvements than MPA at weeks 12, 18 and 26 in WOMAC physical function [Fig. 3(B)] and WOMAC stiffness [Fig. 3(C)]. For 'Get-up-and-go', '10-m timed walk', and patient global assessment, improvements from baseline were evident in both treatment groups throughout the 26-week blinded phase, with no significant between-group differences. For physical activity, no significant differences were evident between groups. However, OMERACT-OARSI responder rates were significantly different at 6 weeks (higher response rate in the MPA group, $P = 0.0138$) and at 26 weeks (higher response rate in the NASHA group, $P = 0.0237$) (Fig. 4). There were no significant between-group differences in the use of rescue medication.

Open-label extension (26–52 weeks)

Patients who received NASHA at 26 weeks showed higher WOMAC pain responder rates at 39 and 52 weeks than at 26 weeks [Fig. 5(A)]. The extent of improvement, approximately 10 percentage points, was similar whether patients initially received NASHA or MPA. The responder rate was lower in the MPA group than the NASHA group at 26 weeks and this difference was maintained at 39 and 52 weeks, with statistical significance at 39 weeks. Similar results were observed with WOMAC pain scores [Fig. 5(B)]. Mean

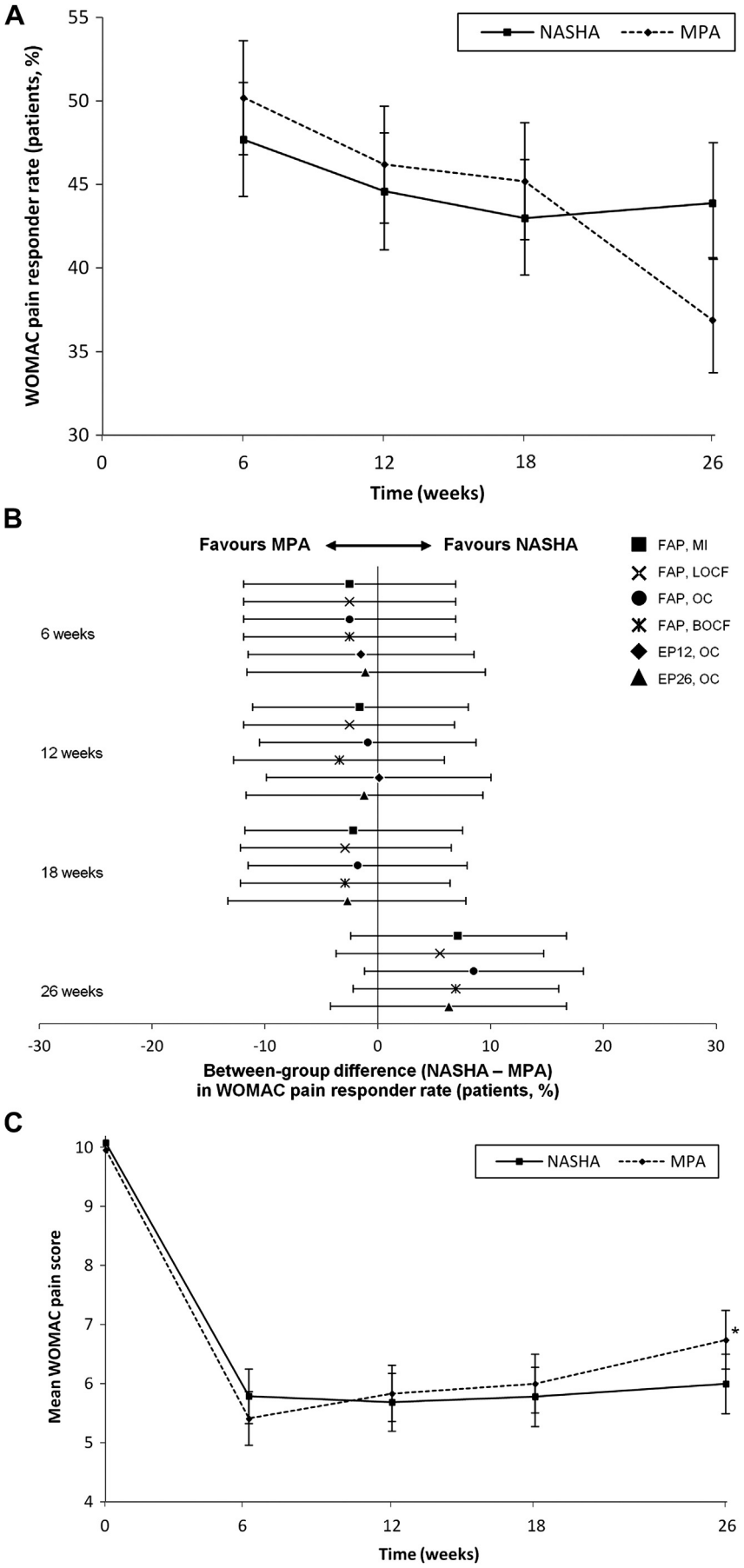


Fig. 2. **A)** WOMAC pain responder rates (±standard error) for the blinded phase of the study (FAP, MI model). **B)** Difference (NASHA − MPA) in WOMAC pain responder rate (±95% CIs): primary (FAP, MI) and secondary analyses (FAP, LOCF; FAP, OC; FAP, BOCF; evaluable population at 12 weeks [EP12], OC and EP26, OC). **C)** WOMAC pain scores (±95% CIs) following double-blind treatment with either NASHA or MPA for each visit to 26 weeks (FAP, MI, repeated measures model). *$P = 0.034$. EP12/26, evaluable population at 12/26 weeks.

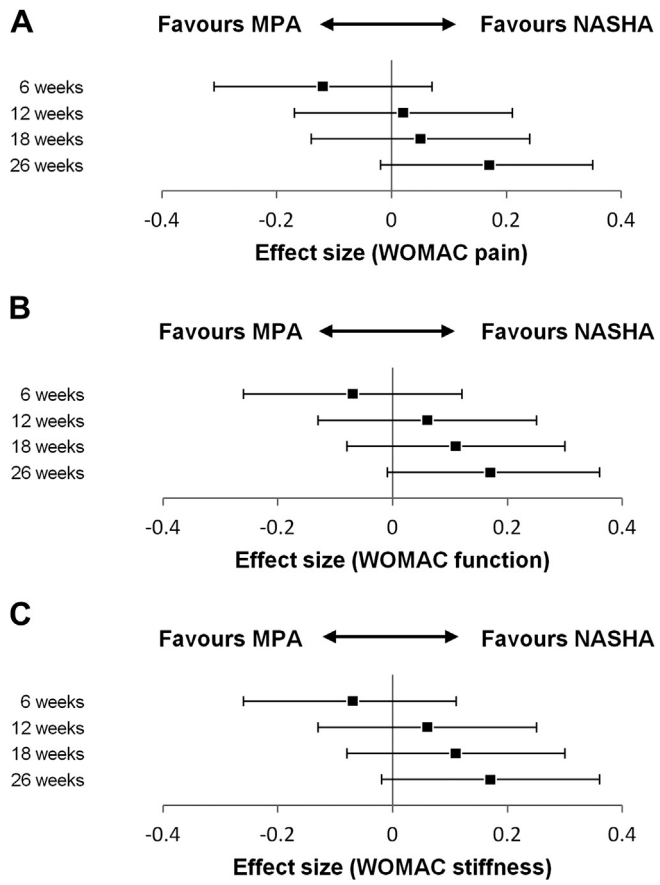
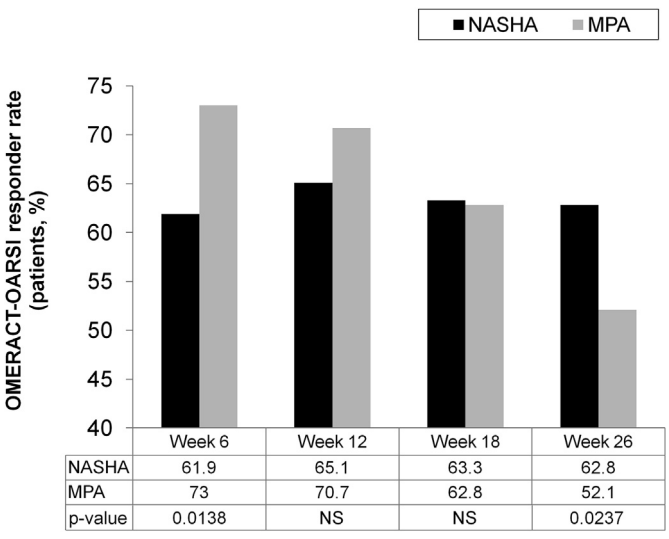


Fig. 3. Effect-sizes for WOMAC domains with 95% CIs during the blinded phase of the study. **A)** WOMAC pain. **B)** WOMAC physical function. **C)** WOMAC stiffness.

WOMAC pain score among recipients of NASHA at 26 weeks decreased by approximately one point between 26 and 39 weeks in both the NASHA–NASHA and MPA–NASHA groups, and both groups showed a small additional decrease between 39 and 52 weeks. WOMAC physical function [Fig. 5(C)] and WOMAC stiffness [Fig. 5(D)] demonstrated similar improvement following the injection of NASHA at 26 weeks. Sustained increases in OMERACT-OARSI responder rates were observed in both of the groups that received the 26-week NASHA injection, with 52-week responder rates of 74.0% in the NASHA–NASHA group and 65.7% in the MPA–NASHA group (Fig. 6).

A minority of patients chose not to receive a second injection at 26 weeks (NASHA: $n = 31$; MPA: $n = 17$). Continuing improvement from baseline was observed among these patients, reflecting the fact that they did not feel the need for a second IA treatment.



	Week 6	Week 12	Week 18	Week 26
NASHA	61.9	65.1	63.3	62.8
MPA	73	70.7	62.8	52.1
p-value	0.0138	NS	NS	0.0237

Fig. 4. OMERACT-OARSI responder rates during the blinded phase of the study.

Safety results

Blinded phase (0–26 weeks)

During the blinded phase, 894 AEs were reported: 462 events among patients in the NASHA group (172/221 patients) and 432 events among patients in the MPA group (156/221 patients). There were 15 serious AEs (nine in the NASHA group, six in the MPA group), none of which were considered related to study treatment. The number of treatment-related AEs was 64 in the NASHA group (48/221 patients), and 15 in the MPA group (15/221 patients). Arthralgia was the most common treatment-related AE in both study groups, with injection site pain, joint stiffness and joint swelling also occurring in more than 1% of patients (Table II). Most treatment-related AEs were reported within 3 days of injection (NASHA: 48/64 cases, MPA: 8/15 cases) and resolved within 2–3 weeks (NASHA: mean 17.5 days, MPA: mean 15.5 days).

Open-label extension (26–52 weeks)

During the OLE, 328 AEs were reported: 160 in the NASHA–NASHA group (96/163 patients) and 168 in the MPA–NASHA group (94/179 patients). The nature of AEs was similar to that in the blinded phase: no allergic sensitivities were observed, arthralgia was the most common treatment-related AE, and joint stiffness, joint swelling and musculoskeletal discomfort were observed in more than 1% of patients (Table II). Consistent with the double-blind phase, there were no treatment-related serious AEs. Also, no allergic reactions to the second injection were observed.

Discussion

In this comparison of IA NASHA with a corticosteroid, NASHA was well tolerated and demonstrated to be non-inferior to MPA throughout the 26-week, double-blind phase of the study. Little deterioration in WOMAC responder rate was observed between 12 and 26 weeks in the NASHA group, whereas significant deterioration was observed in the MPA group between 18 and 26 weeks. Open-label treatment with NASHA at 26 weeks provided improvements lasting to 52 weeks and demonstrated that NASHA provides benefit in patients previously treated with corticosteroid.

By demonstrating non-inferiority vs MPA, this study provides level one evidence that NASHA should be considered as a treatment option for knee OA. Most evidence supporting the efficacy of IA corticosteroids for knee OA has been obtained in patients with Kellgren–Lawrence grade II or III[6]. For the present study, patients with the same severity of knee OA were recruited to ensure that the outcome would be applicable to patients most likely to benefit from IA corticosteroid therapy. With respect to WOMAC pain responder rate, MPA showed activity to 18 weeks post-treatment. This result is consistent with high-quality studies showing that IA injection of corticosteroid provides statistically significant improvement in symptoms at 16–24 weeks[18], although this treatment is generally perceived to have a shorter duration of action. In a meta-analysis of corticosteroids and HA, corticosteroids demonstrated earlier effectiveness with a short duration, while HA demonstrated a slower time to onset of effectiveness but longer lasting pain relief[5]. The WOMAC pain data from this study indicate the same trends [Fig. 2(C) and Fig. 3(A)]. The first evaluation was at 6 weeks and it is

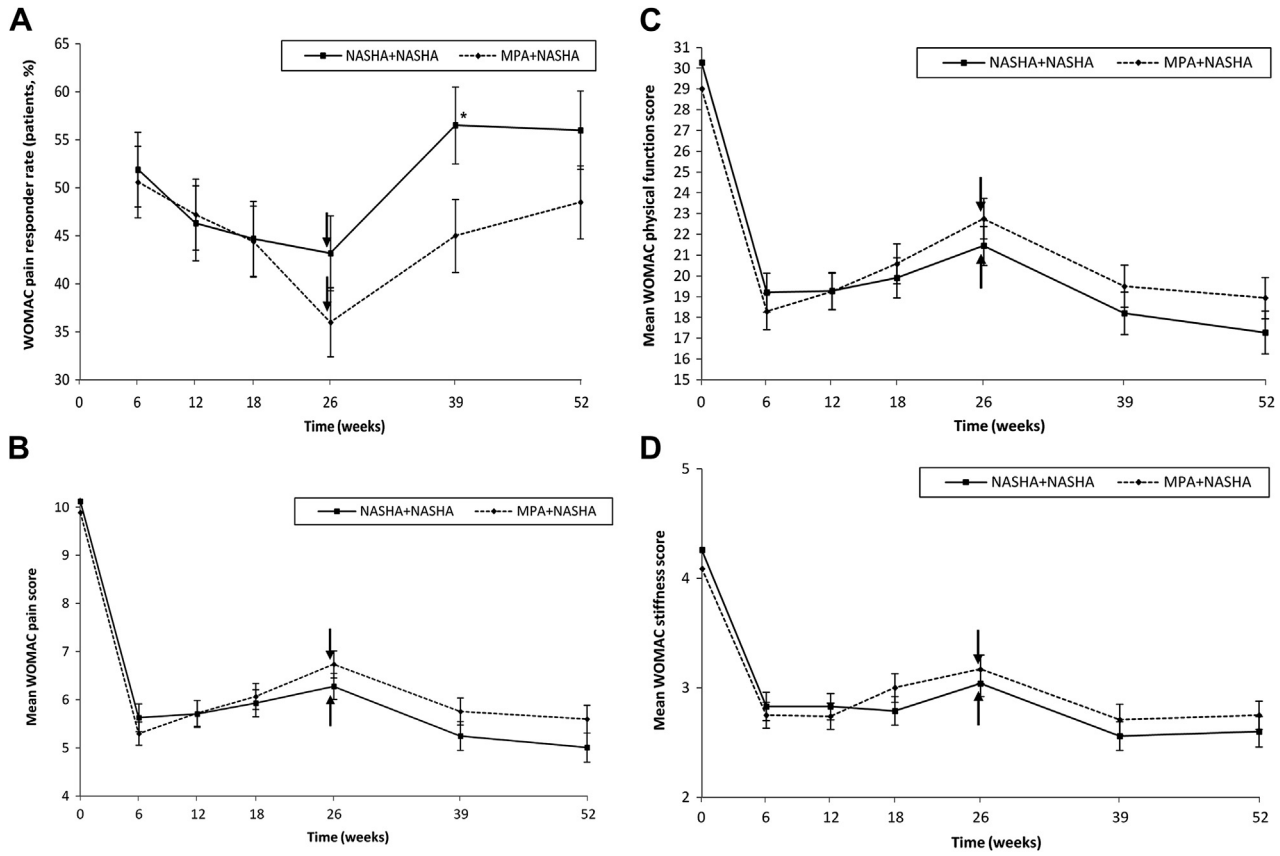
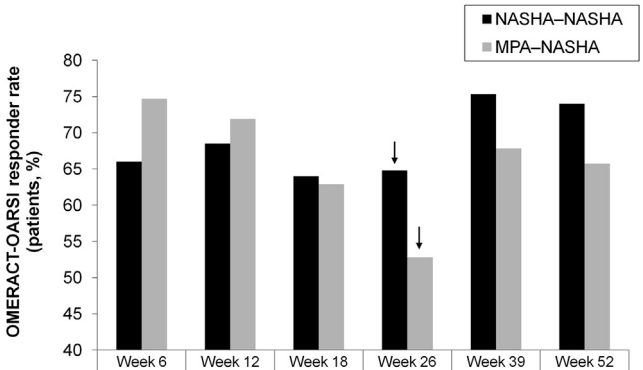


Fig. 5. Outcomes for each visit to 52 weeks among patients receiving NASHA treatment at 26 weeks (OLE analysis population, observed cases). Arrows indicate the 26-week timepoint for open-label treatment with NASHA. **A)** WOMAC pain responder rates (±standard error). Number of observed values (NASHA–NASHA/MPA–NASHA) at 6 weeks and 12 weeks: 162/178; 18 weeks: 161/178; 26 weeks: 162/178; 39 weeks: 154/171; 52 weeks: 150/169. **B)** WOMAC pain scores (±standard error). Number of observed values (NASHA–NASHA/MPA–NASHA) at 6 weeks and 12 weeks: 162/178; 18 weeks: 161/178; 26 weeks: 162/178; 39 weeks: 154/171; 52 weeks: 150/169. **C)** WOMAC physical function scores (±standard error). Number of observed values (NASHA–NASHA/MPA–NASHA) at 6 weeks and 12 weeks: 161/177; 18 weeks: 161/178; 26 weeks: 162/176; 39 weeks: 154/171; 52 weeks: 150/168. **D)** WOMAC stiffness scores (±standard error). Number of observed values (NASHA–NASHA/MPA–NASHA) at 6 weeks and 12 weeks: 162/178; 18 weeks: 161/178; 26 weeks: 162/178; 39 weeks: 153/171; 52 weeks: 150/169. *$P = 0.039$.

possible that the effectiveness of MPA peaked before this time. The effectiveness cross-over point for HA and corticosteroid in the meta-analysis was at 4 weeks, vs ~12 weeks in the current study. The size and quality of the current study suggest that it is likely to provide a more realistic view.



	Week 6	Week 12	Week 18	Week 26	Week 39	Week 52
NASHA–NASHA	66	68.5	64	64.8	75.3	74
MPA–NASHA	74.7	71.9	62.9	52.8	67.8	65.7

Fig. 6. OMERACT-OARSI responder rates for each visit to 52 weeks among patients receiving NASHA treatment at 26 weeks (OLE analysis population, observed cases). Arrows indicate the 26-week timepoint for open-label treatment with NASHA. Number of observed values (NASHA–NASHA/MPA–NASHA) at 6 weeks and 12 weeks: 162/178; 18 weeks: 161/178; 26 weeks: 162/178; 39 weeks: 154/171; 52 weeks: 150/169.

Lack of allergic reactions to the second injection shows a lack of increased sensitivity to the device; this answered the first of the two queries relating to the OLE. Regarding the second query, improvements were observed in both groups following the second injection. This suggests that further injections of NASHA may be administered according to clinical need arising after the initial treatment, and that symptomatic improvement may be expected. The post-NASHA improvement in WOMAC pain responder rate was smaller after the 26-week OLE injection than the increase after initial treatment. This is in line with expectation, given the residual effect of initial treatment. However, duration of response to the second treatment appeared longer than after the initial injection. Among patients receiving two NASHA injections, considerable reductions from baseline in WOMAC pain score were evident at 26 weeks (~40%) and 52 weeks (nearly 50%). Sustained reduction from baseline between 26 and 52 weeks was also observed among patients receiving NASHA after MPA. These findings, together with the WOMAC pain responder rate of 56% at 52 weeks in the NASHA–NASHA group (OMERACT-OARSI responder rate of 74%), represent a meaningful change in OA symptoms for most patients.

Unlike previous studies of NASHA for knee OA, the present trial recruited patients without end-stage OA (Kellgren–Lawrence grade IV), without clinical effusion at baseline and without significant pain from other joints. It is now recognised that HA is not generally effective for end-stage OA or in patients with large clinical effusions[24–26]. The presence of poly-articular pain appears to be a

Table II
AEs, related to the product or injection procedure, occurring in >1% of patients (safety population)

	NASHA*		MPA†		
	Patients	%	Patients	%	*P*-value‡
Blinded phase					
Arthralgia	38	17.2	7	3.2	<0.0001
Injection site pain	3	1.4	1	0.5	0.623
Joint stiffness	4	1.8	0	0	0.123
Joint swelling	5	2.3	1	0.5	0.216
Open-label extension					
Arthralgia	30	18.4	31	17.3	NC
Joint stiffness	1	0.6	3	1.7	NC
Joint swelling	2	1.2	1	0.6	NC
Musculoskeletal discomfort	3	1.8	0	0.0	NC

NC, not calculated.
* Blinded phase, $n = 221$; open-label extension, NASHA–NASHA group, $n = 163$.
† Blinded phase, $n = 221$; open-label extension, MPA–NASHA group, $n = 179$.
‡ Fisher's exact test.

confounding factor in knee OA pain studies[27]. When these factors were eliminated in a post-hoc analysis, the results were consistent with the current study[28]. Single-injection NASHA was shown in the present study to provide a stable, long-term improvement in OA symptoms. The duration of effect was consistent with the 4-week half-life of NASHA and its prolonged residence time within the joint. Although the precise mechanism of action of HA has yet to be fully described, supplementation of HA through IA injection of NASHA appears to offer the potential for sustained patient benefits.

AEs were largely anticipated in nature, with a lack of differences between NASHA and MPA regarding the type, onset and duration of AEs. Arthralgia was the most common treatment-related AE, and this is anticipated to occur among recipients of IA HA[12,29,30]. The occurrence of arthralgia in the NASHA group did not affect the WOMAC pain responder rate at 12 weeks. No systemic AEs or acute flare reactions were seen with NASHA, and a second injection of the product at 26 weeks appeared to be at least as well tolerated as the initial injection.

The strengths of this study were the use of MPA control as a pragmatic standard-of-care for knee OA, and the excellent conduct of the study as shown by the very low drop-out rate (~90% completion of the 26-week blinded phase). Similarity of patients' baseline characteristics in the two study groups confirms that the 1:1 randomization process was effective, and that post-treatment between-group differences may be attributed to study medication. Additionally the WOMAC pain responder rate, a clinically meaningful primary endpoint, has been used in few other studies. The definition of the responder rate is an important consideration. For example, responder rates based on absolute improvement are likely to be higher among patients with more severe symptoms than responder rates based on percentage improvement. In accordance with a report from the Osteoarthritis Research Society International Standing Committee for Clinical Trials, the primary endpoint in this study was based on both percentage and absolute improvement[22]. In a post-hoc analysis using a WOMAC pain responder definition of 40% improvement and no requirement for absolute change, the 26-week data showed a statistically significant benefit with NASHA (responder rates: NASHA, 54%; MPA, 44%; $P = 0.043$). The lack of a saline control arm may be considered as a limitation of this study but, at the time of designing the study, the inclusion of such a control group was considered unethical. We consider that the results are applicable to knee OA patients presenting with mild to moderate structural changes and pain in the absence of clinical effusion.

In conclusion, this study showed that NASHA is a valuable treatment for knee OA, providing effectiveness that was non-inferior to MPA. It also indicated that the effect of NASHA is longer lasting, with significantly improved pain response at 26 weeks compared to MPA. NASHA is well tolerated in relation to both primary and secondary injections, with most AEs being anticipated and non-allergenic in nature.

Contributions

- Dr Ross Leighton: (1) Conception and design, (2) Drafting the article, (3) Final approval of the article
- Dr Christian Åkermark: (1) Acquisition of data, (2) Critical revision of the article for important intellectual content, (3) Final approval of the article
- Prof René Therrien: (1) Acquisition of data, (2) Critical revision of the article for important intellectual content, (3) Final approval of the article
- Prof James Richardson: (1) Acquisition of data, (2) Critical revision of the article for important intellectual content, (3) Final approval of the article
- Mats Andersson: (1) Statistical expertise, (2) Critical revision of the article for important intellectual content, (3) Final approval of the article
- Martin Todman: (1) Analysis and interpretation of data, (2) Drafting the article, (3) Final approval of the article
- Prof Nigel Arden: (1) Conception and design, (2) Drafting the article, (3) Final approval of the article

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Conflict of interest

- Dr Ross Leighton is a paid consultant for Etex Corporation; he has participated in speaker bureaux for Stryker, Smith and Nephew, Synthes, Depuy, Biomet and Zimmer; and he receives a royalty from a plate and screw set designed and marketed with Zimmer USA.
- Dr Christian Åkermark reports no conflicts of interest.
- Prof René Therrien reports no conflicts of interest.
- Prof James Richardson reports no conflicts of interest.
- Mats Andersson was, at the time of the study, a full time employee of Q-Med, AB.
- Martin Todman (PhD) was, at the time of the study, a full-time employee of Smith & Nephew, UK Ltd.
- Prof Nigel Arden is a paid consultant for Q-Med, AB and Smith & Nephew, Inc.

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References

1. Jones R, Rubin G, Berenbaum F, Scheiman J. Gastrointestinal and cardiovascular risks of nonsteroidal anti-inflammatory drugs. Am J Med 2008;121:464–74.
2. Naesdal J, Brown K. NSAID-associated adverse effects and acid control aids to prevent them: a review of current treatment options. Drug Saf 2006;29:119–32.
3. Schilling A, Corey R, Leonard M, Eghtesad B. Acetaminophen: old drug, new warnings. Cleve Clin J Med 2010;77:19–27.
4. Trelle S, Reichenbach S, Wandel S, Hildebrand P, Tschannen B, Villiger PM, *et al*. Cardiovascular safety of non-steroidal anti-inflammatory drugs: network meta-analysis. BMJ 2011;342:c7086.
5. Bannuru RR, Natov NS, Obadan IE, Price LL, Schmid CH, McAlindon TE. Therapeutic trajectory of hyaluronic acid versus corticosteroids in the treatment of knee osteoarthritis: a systematic review and meta-analysis. Arthritis Rheum 2009;61:1704–11.
6. Bellamy N, Campbell J, Robinson V, Gee T, Bourne R, Wells G. Intraarticular corticosteroid for treatment of osteoarthritis of the knee. Cochrane Database Syst Rev 2006;(2). CD005328.
7. Habib GS. Systemic effects of intra-articular corticosteroids. Clin Rheumatol 2009;28:749–56.
8. Habib GS, Saliba W, Nashashibi M. Local effects of intra-articular corticosteroids. Clin Rheumatol 2010;29:347–56.
9. Agerup B, Berg P, Akermark C. Non-animal stabilized hyaluronic acid: a new formulation for the treatment of osteoarthritis. BioDrugs 2005;19:23–30.
10. Edsman K, Hjelm R, Larkner H, Nord LI, Karlsson A, Wiebensjo A, *et al*. Intra-articular duration of Durolane™ after single injection into the rabbit knee. Cartilage 2011;2:384–8.
11. Lindqvist U, Tolmachev V, Kairemo K, Astrom G, Jonsson E, Lundqvist H. Elimination of stabilised hyaluronan from the knee joint in healthy men. Clinical Pharmacokinet 2002;41:603–13.
12. Altman RD, Akermark C, Beaulieu AD, Schnitzer T. Efficacy and safety of a single intra-articular injection of non-animal stabilized hyaluronic acid (NASHA) in patients with osteoarthritis of the knee. Osteoarthritis Cartilage 2004;12:642–9.
13. Krocker D, Matziolis G, Tuischer J, Funk J, Tohtz S, Buttgereit F, *et al*. Reduction of arthrosis associated knee pain through a single intra-articular injection of synthetic hyaluronic acid. Z Rheumatol 2006;65:327–31.
14. Skwara A, Ponelis R, Tibesku CO, Rosenbaum D, Fuchs-Winkelmann S. Gait patterns after intraarticular treatment of patients with osteoarthritis of the knee–hyaluronan versus triamcinolone: a prospective, randomized, doubleblind, monocentric study. Eur J Med Res 2009;14:157–64.
15. Hochberg MC, Perlmutter DL, Hudson JI, Altman RD. Preferences in the management of osteoarthritis of the hip and knee: results of a survey of community-based rheumatologists in the United States. Arthritis Care Res 1996;9:170–6.
16. Cole BJ, Schumacher Jr HR. Injectable corticosteroids in modern practice. J Am Acad Orthop Surg 2005;13:37–46.
17. Pyne D, Ioannou Y, Mootoo R, Bhanji A. Intra-articular steroids in knee osteoarthritis: a comparative study of triamcinolone hexacetonide and methylprednisolone acetate. Clin Rheumatol 2004;23:116–20.
18. Arroll B, Goodyear-Smith F. Corticosteroid injections for osteoarthritis of the knee: meta-analysis. BMJ 2004;328:869.
19. Schulz KF, Altman DG, Moher D. CONSORT 2010 statement: updated guidelines for reporting parallel group randomised trials. PLoS Med 2010;7:e1000251.
20. International Committee of Medical Journal Editors (ICMJE). Uniform Requirements for Manuscripts Submitted to Biomedical Journals: Writing and Editing for Biomedical Publication. Available from: http://www.icmje.org/urm_full.pdf. (Accessed 27.03.13).
21. Recommendations for the medical management of osteoarthritis of the hip and knee: 2000 update. American College of Rheumatology Subcommittee on Osteoarthritis Guidelines. Arthritis Rheum 2000;43:1905–15.
22. Dougados M, Leclaire P, van der Heijde D, Bloch DA, Bellamy N, Altman RD. Response criteria for clinical trials on osteoarthritis of the knee and hip: a report of the Osteoarthritis Research Society International Standing Committee for Clinical Trials response criteria initiative. Osteoarthritis Cartilage 2000;8:395–403.
23. Pham T, Van Der Heijde D, Lassere M, Altman RD, Anderson JJ, Bellamy N, *et al*. Outcome variables for osteoarthritis clinical trials: the OMERACT-OARSI set of responder criteria. J Rheumatol 2003;30:1648–54.
24. Stephens MB, Beutler AI, O'Connor FG. Musculoskeletal injections: a review of the evidence. Am Fam Physician 2008;78:971–6.
25. Conrozier T, Mathieu P, Schott AM, Laurent I, Hajri T, Crozes P, *et al*. Factors predicting long-term efficacy of Hylan GF-20 viscosupplementation in knee osteoarthritis. Joint Bone Spine 2003;70:128–33.
26. Gossec L, Dougados M. Do intra-articular therapies work and who will benefit most? Best Pract Res Clin Rheumatol 2006;20:131–44.
27. O'Malley KJ, Suarez-Almazor M, Aniol J, Richardson P, Kuykendall DH, Moseley Jr JB, *et al*. Joint-specific multidimensional assessment of pain (J-MAP): factor structure, reliability, validity, and responsiveness in patients with knee osteoarthritis. J Rheumatol 2003;30:534–43.
28. Arden NK, Akermark C, Andersson M, Todman MG, Altman RD. A randomized saline-controlled trial of NASHA hyaluronic acid for knee osteoarthritis. Curr Med Res Opin 2013, in press.
29. Kemper F, Gebhardt U, Meng T, Murray C. Tolerability and short-term effectiveness of hylan G-F 20 in 4253 patients with osteoarthritis of the knee in clinical practice. Curr Med Res Opin 2005;21:1261–9.
30. Curran MP. Hyaluronic acid (Supartz(R)): a review of its use in osteoarthritis of the knee. Drugs Aging 2010;27:925–41.