A Randomized, Double-blind, Placebo-controlled Study of Proteoglycan and Phosphatidylserine for Management of Knee and Hip Pain

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Authors' contributions

This work was carried out in collaboration among all authors. All authors approved the protocol, performed the statistical analysis, reviewed the manuscript for intellectual content, and read and approved the final manuscript. All authors read and approved the final manuscript.

Original Research Article

ABSTRACT

Aims: This study was designed to evaluate if combined oral administration of proteoglycan and phosphatidylserine (PG+PS) for 30 days would improve osteoarthritis symptoms in adults with moderate to severe knee and hip pain.
Study Design: Randomized, double-blind, placebo-controlled study.

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Place and Duration of Study: Well-Come Fitness & Health Center (Copenhagen, Denmark), between September 2023 to November 2023.

Methodology: A total of 96 participants (65 female, 31 male, mean age 51 years, mean body mass index 25 kg/m^2) were randomized to take 1 tablet/day of placebo (N=31), 12.5 mg proteoglycan (PG, N=32), or 12.5 mg proteoglycan + 60 mg phosphatidylserine (PG+PS, N=33) orally for 30 days. The change from baseline in physical performance was evaluated by stair climb test (SCT) and self-paced walk test (SPWT). Joint (knee and hip) pain prior to physical performance tests was evaluated by the change in visual analog scale (VAS) score (range 0-10).

Results: Statistically significant improvements were observed in the change from baseline to Day 30 for the SCT and SPWT for PG+PS vs placebo and PG+PS vs PG (all P < .05). Statistically significant improvements in joint pain (VAS Score) were also observed for PG+PS vs placebo and vs PG. Improvements in physical performance and joint pain with PG+PS generally occurred during the first week of the study.

Conclusion: Combination administration of PG+PS for 30 days reduced joint pain and improved physical performance in adults with moderate to severe knee and hip pain. Additional studies to further validate these findings are warranted.

1. INTRODUCTION

Osteoarthritis is a leading cause of disability worldwide [1-3]. Osteoarthritis is characterized by deterioration of articular cartilage, the connective tissue that covers the bone surface in joints and facilitates movement while withstanding joint loading. Permanent degradation of the extracellular matrix (ECM) and joint tissue remodeling eventually leads to joint dysfunction, reduced mobility, and pain.

The ECM is primarily composed of water, collagen, and proteoglycan. PGs make up 10 to 15% of cartilage, provide compressive strength, attract water, and are critical for cartilage maintenance and repair [4]. PG degradation and deficiency are features of osteoarthritis. Loss of aggrecan, the most abundant PG in articular cartilage, is associated with onset of osteoarthritis [5]. Conversely, increased aggrecan levels were observed concomitant with improvements in pain and mobility in a study of resveratrol in individuals with knee osteoarthritis [6]. Chondroitin sulfate (a PG) and glucosamine sulfate, which have a well-documented beneficial effect on pain and mobility in osteoarthritis, have been shown to increase PG synthesis [7]. PG administration in animals is associated with a reduction in arthritis [8]. Oral administration of PGs in humans has also been demonstrated to have a beneficial effect on mobility, pain, and joint health [4,9,10].

Phosphatidylserine (PS) is a glycerophospholipid that predominantly occurs on the inner leaflet of eukaryotic cell membranes. PS is involved in biological processes including enzyme activation, apoptosis, neurotransmission, and synaptic refinement [11,12]. PS has been shown to reduce inflammation and bone loss in animal models of arthritis [12-15]. PS has well documented anti-inflammatory effects and studies of PS in humans have primarily focused on potential benefits on cognitive function in elderly individuals [11]. Dysregulation of PS is associated with central nervous system disorders associated with inflammation, including Alzheimer's disease, Parkinson's disease, depression, and stroke [11]. Studies in humans support a positive effect of oral PS administration on cognition, mood, and stress [11,16-21]. PS is present in high amounts in soy and sunflower lecithin as well as meat and fish, and oral administration of PS is well tolerated. We hypothesized that PS would have a positive impact on mood and motivation that may lead to improved training ability.

To our knowledge, no other studies have evaluated combined administration of PG and PS on osteoarthritis in humans. This study was designed to evaluate if combined administration of PG and PS for 30 days would improve osteoarthritis symptoms in adults with moderate to severe knee and hip pain.

2. MATERIALS AND METHODS

2.1 Study Design and Setting

This randomized, double-blind, placebo-controlled study evaluated once-daily administration of PG+PS for 30 days on knee

and hip pain in adults with moderate to severe knee and hip pain. A placebo group and a positive control (PG only) group were included. The study was conducted from September 2023 to November 2023 at Well-Come Fitness & Health Center (Copenhagen, Denmark).

2.2 Participants

Participants were recruited from health clinics and sports centers in Copenhagen and were evaluated for eligibility by a specially trained nurse and a physiotherapist. Generally healthy male and female (not pregnant or breastfeeding) adults between the ages of 30 to 75 (inclusive) with BMI<30 kg/m^2 and moderate to severe knee and hip pain (individuals self-reported their knee and hip pain according to the following scale: none, mild, moderate, or severe) were eligible to participate. Participants were ineligible if they were taking prescription pain medication or supplements containing any of the following within 90 days prior to the start of the study: chondroitin, glucosamine, methylsulfonylmethane (MSM), Boswellia, turmeric, type II collagen, or PS. Other exclusion criteria included a history of any medical or arthritic conditions that could interfere with evaluation of the index knee joint including fibromyalgia, rheumatoid arthritis, or other inflammatory arthropathies affecting the knee joint, arthroscopic or open surgery to the knee within the previous 6 months, or knee injections with corticosteroids within the previous 30 days or hyaluronic acid within the previous 3 months.

2.3 Intervention

Participants were randomized 1:1:1 to 1 tablet/day of placebo, 12.5 mg proteoglycan (PG), or 12.5 mg proteoglycan + 60 mg phosphatidylserine (PG+PS) orally for 30 days. Tablets were manufactured by John M. Petersen, M.Sc. (Almega A/S, Ringsted, Denmark). Simple randomization of participants was performed by using a random number generator. Participants were provided coded containers containing 30 tablets of either PG+PS, PG, or matching placebo. Participants were randomly assigned a coded container by the recruiting nurse. The sealed list of container numbers and corresponding contents was kept by an independent auditor until the end of the study. The participants, recruiting nurse, and investigators remained blinded throughout the study. Participants were instructed to take 1 tablet every morning for the 30-day study.

Participants could take 200 mg Ibuprofen up to 3 times a day during the study. All participants were provided with daily reminders of study procedures via SMS text or virtual meetings.

2.4 Outcomes and Assessments

Study outcomes include the change from baseline in the number of stairs climbed in the stair climb test (SCT), the distance walked in the self-paced walk test (SPWT), and joint pain.

Assessments were conducted at baseline (Day 0) and on Days 4, 7, 14, and 30. At the beginning of each assessment, participants rated their joint pain by placing their finger on a blank 100 mm Visual Analog Scale (VAS), with 0 defined as no pain and 100 defined as worst possible pain. The blank ruler was then compared with a 100 mm lined ruler and divided by 10. Thus, the *VAS Score* ranged from 0 to 10 and served as an assessment of joint pain prior to the SCT and SPWT.

The SCT assessed the ability to ascend a flight of stairs, as well as lower extremity strength, power, and balance [22]. The SCT was conducted using a Stairmaster (Life Fitness, Rosemont, IL, USA) and participants could adjust the level and speed. The number of floors achieved over a test period of 5 minutes was recorded. The SPWT assessed the distance (km) each participant walked over a test period of 5 minutes [22]. The SPWT was conducted using a treadmill (Life Fitness, Rosemont, IL, USA). Participants were instructed to walk quickly and safely without overexerting themselves and could adjust the speed on the treadmill as needed during the test.

Participants were asked to report any adverse effects at each assessment. Participants could also contact study personnel at any time for questions about the study or to report adverse events. Compliance was assessed on Day 7, 14 and 30 by measuring the number of tablets remaining in the bottle.

2.5 Data Analysis

Continuous variables were summarized using descriptive statistics. The Kruskal-Wallis test was used to compare the continuous variables across the three groups with post-hoc Dwass-Steel-Critchlow-Fligner pairwise comparison test for between group differences. The statistical significance level was set at $p < .05$. Statistical analyses were conducted using RStudio (Version 4.1) and jamovi (Version 2.3).

3. RESULTS

3.1 Study Population

A total of 127 individuals were assessed for eligibility. Overall, 106 participants were included in the study (allocated to treatment groups). Of these, 96 participants completed the study and were included in the analysis (Fig. 1). The majority of study participants were female (n=65, 68%). The mean ±SD age was 50.6 ±9.5 years and BMI was 24.9 ±1.8 kg/m^2. Age and BMI were similar across groups.

3.2 Measures of Physical Performance

3.2.1 Stair climb test results

A statistically significant improvement from baseline to Day 30 was observed for PG+PS vs placebo and PG in the SCT (both $P < .001$, Table 1, Fig. 2). Notably, a statistically significant difference for PG+PS vs both placebo and PG was observed at the earliest timepoint assessed (Day 4), and this difference persisted for all subsequent assessments through Day 30 (Fig. 1).

3.2.2 Self-paced walk test results

The Day 30 change from baseline in the SPWT was statistically significant for PG+PS vs placebo and PG (both $P < .05$, Table 1, Fig. 2). Similar to the SCT, statistically significant differences were observed for PG+PS vs placebo and PG at the earliest timepoint assessed (Day 4); these differences persisted for the duration of the study.

3.3 Joint Pain Results

3.3.1 VAS Score

A statistically significant improvement from baseline to Day 30 was observed for PG+PS vs placebo and PG in VAS Score (both $P < .05$, Table 1, Fig. 2). Additionally, a statistically significant difference for PG+PS vs placebo was observed at Day 7 that persisted for the duration of the study (Fig. 2). The difference between PG+PS and PG was statistically significant at Day 30 (Fig. 2).

3.4 Safety and Tolerability

There were no adverse events or tolerability issues in any treatment group during the study.
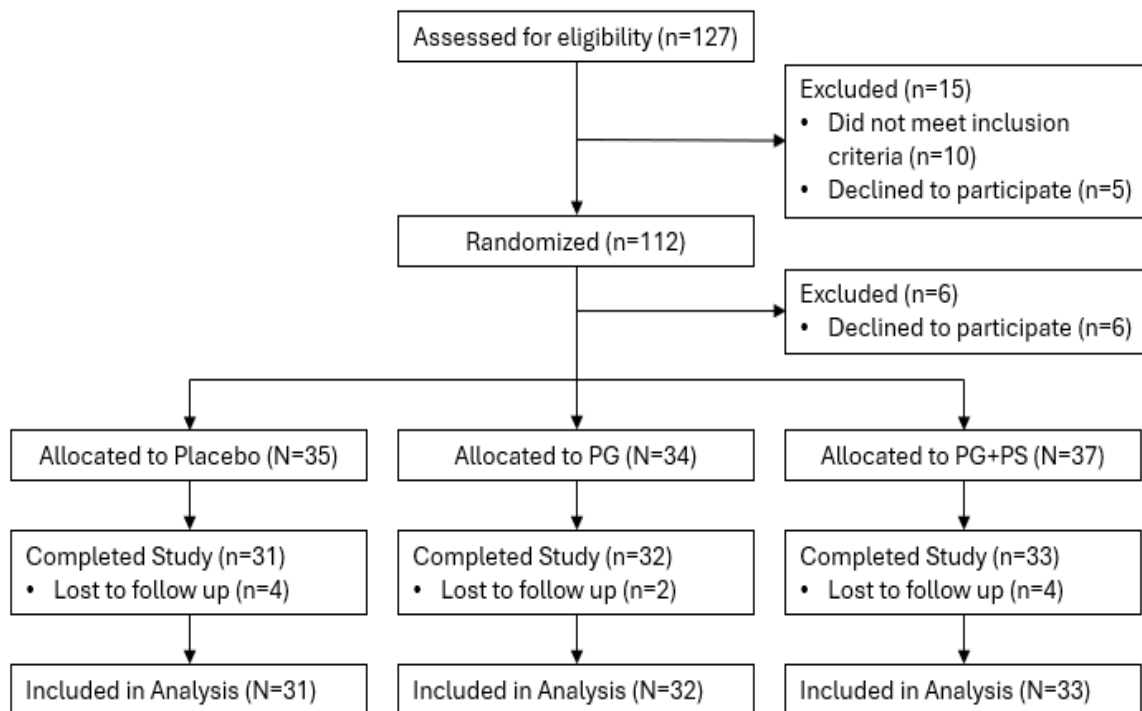

Fig. 1. Study population

Table 1. Efficacy measures: Comparison between baseline and Day 30

	Placebo (N=31)	PG (N=32)	PG+PS (N=33)	*P* value vs Placebo	*P* value vs PG
SCT					
Baseline	19.0 ±2.9	16.5 ±1.5	17.7 ±3.2		
Day 30	24.4 ±4.2	21.9 ±3.3	25.7 ±5.9		
Change	5.3 ±0.5	5.3 ±0.4	8.0 ±0.5	*P* < .001	*P* < .001
% Change	28 ±15	32 ±14	44 ±10		
SPWT					
Baseline	0.41 ±0.08	0.33 ±0.05	0.37 ±0.09		
Day 30	0.61 ±0.12	0.52 ±0.05	0.60 ±0.12		
Change	0.20 ±0.01	0.19 ±0.01	0.22 ±0.01	*P* = .034	*P* = .025
% Change	49 ±17	60 ±26	62 ±13		
VAS Score					
Baseline	4.3 ±1.1	4.4 ±1.2	4.8 ±1.6		
Day 30	3.7 ±0.9	3.4 ±0.9	3.2 ±1.0		
Change	-0.6 ±0.1	-1.0 ±0.1	-1.6 ±0.2	*P* < .001	*P* = .020
% Change	14 ±12	22 ±15	31 ±16		

SCT=Stair Climb test. Numbers indicate floors completed during the 5-minute test. SPWT=Self-Paced Walk Test. Numbers indicate distance in km covered during the 5-minute test. VAS Score indicates level of joint pain on a scale of 0-10 where 0=no pain. Baseline and Day 30 data are mean and standard deviation (SD). Change data are mean and standard error of the mean (SEM) for the change from baseline to Day 30. P values are indicated for comparison vs PG+PS
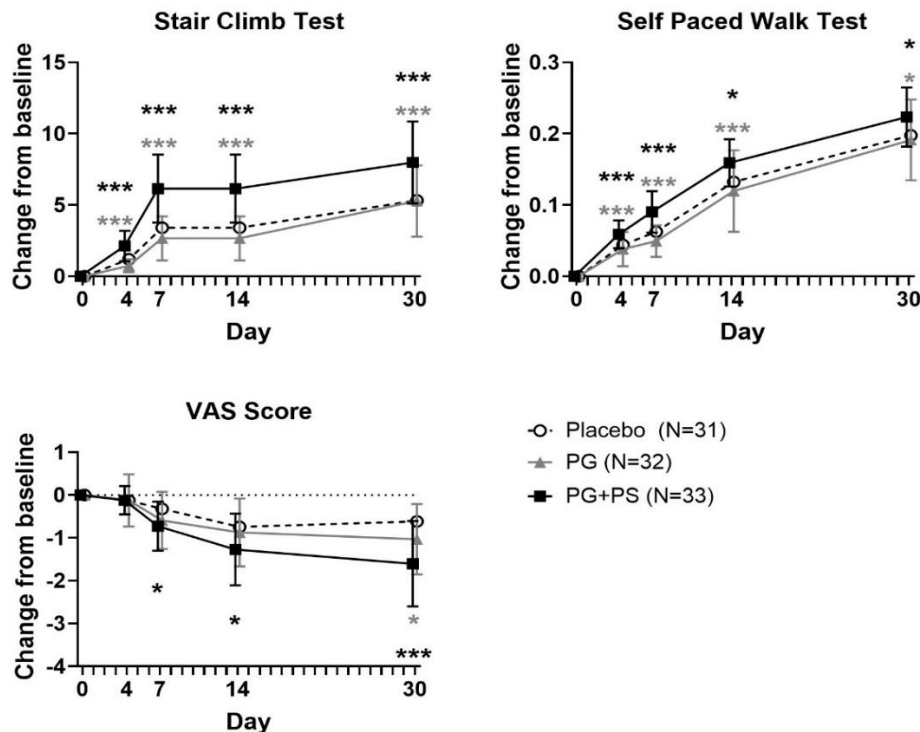

Fig. 2. Change from baseline in efficacy measures

*Stair Climb test numbers indicate floors completed during the 5-minute test. Self-Paced Walk test numbers indicate distance in km covered during the 5-minute test. VAS Score indicates level of joint pain on a scale of 0-10 where 0=no pain. Data are mean and standard deviation (SD) for the change from baseline (day 0) for each group (N=96). * P < .05, ** P < 0.01, *** P < 0.001 for comparison vs PG+PS. P values for comparison vs PG are indicated by grey stars; P values for comparisons vs Placebo are indicated by black stars*

4. DISCUSSION

PG+PS consistently resulted in improvements in physical performance measures and joint pain vs placebo for the 30-day study. Improvements for PG+PS were observed vs both placebo and PG in the SCT and SPWT at the earliest timepoint measured (Day 4) and persisted for the duration of the study. Improvements in joint pain were statistically significant for PG+PS vs placebo as early as Day 7. These results suggest that the addition of PS to PG resulted in earlier improvements in physical performance within 1 week of administration. Early improvements in joint pain were also observed with PG+PS vs placebo but not PG. The improvement in VAS Score with PG+PS at Day 30 reflects a clinically meaningful improvement in joint pain that may have contributed to improved physical performance.

In the PS+PG group, the percent change from baseline to Day 30 in the SCT and SPWT was 44% and 62%, respectively. The difference in the change from baseline to Day 30 was statistically significant for PG+PS compared to both the PG and placebo groups. The SPWT and SCT are reliable measures of physical performance in studies of knee and hip osteoarthritis and are generally responsive to improvements in mobility, however, an MCID for both measures has not yet been established [22]. The consistent results for the two different tests strengthen the validity of the current findings. Additionally, both the SCT and SPWT are brief measures of physical performance that would not be expected to improve with limited repeated assessments (eg, performance would not improve due to training) [22]. However, any effects of repeated testing on performance would be expected to occur across all groups (including placebo). These results indicate that the addition of PS to PG produced a superior improvement to PG alone.

A reduction in knee and hip pain was observed in the PG+PS group, demonstrated by an improvement from baseline to Day 30 of 31% in the VAS Score. Additionally, statistically significant differences for PG+PS vs placebo were observed for the change from baseline to Day 30. Moreover, differences for PG+PS vs placebo were observed as early as Day 7 for both measures. VAS Score was also improved for PG+PS vs PG at Day 30.

VAS measures of pain on a 100 mm scale such as the one used in this study are commonly used measures for assessing pain and the change in pain [23]. The minimal clinically important difference (MCID) for 100-mm VAS measures has been shown to rage from 11 to 14 mm and does not depend on the severity of the pain experienced at baseline [23]. Thus, the mean change from baseline to Day 30 in VAS Score in the PG+PS group of -1.6 (or -16 on a 100-mm scale) reflects a clinically important improvement in joint pain.

The mechanism for the improvement in mobility and reduction in knee and hip pain observed is likely due to improvement in joint integrity but other benefits have yet to be evaluated. PGs are critical to maintenance and repair of articular cartilage [4] and loss of PG occurs in osteoarthritis [5]. In humans, oral administration of PGs has been shown to improve integrity of articular cartilage and to have a beneficial effect on mobility, pain, and joint health [4,7,9,10]. Additionally, a positive effect of PS has been observed on cognition, mood, and stress in elderly individuals [11,16-21]. Although the mechanism for the benefits of PG+PS on mobility and joint pain requires further study, we hypothesize that PS has a positive impact on mood and motivation that may lead to improved training ability. Future studies of the potential benefits of PS in similar populations is needed to better understand the contribution of PS.

Strengths of the study are the randomized, placebo-controlled study design. Furthermore, participants were blinded to the nature of the treatment. These study design qualities strengthen the validity of the findings. The early superiority of PG+PS vs PG on measures of physical performance was not observed in the measures of joint pain. This may be attributed to smaller between group differences that were not detectable with the sample size in this study. Additional studies on the impact of PG+PS would be useful to better understand the impact of PG+PS on physical performance and joint pain in this population.

5. CONCLUSION

This study is the first to evaluate the impact of daily supplementation with PS plus PG on measures of physical performance and joint pain in adults with moderate to severe knee and hip pain. Study results indicate that the combination of PG+PS produced a beneficial effect on

physical performance and joint pain within 1 week of administration. Additionally, administration of PG+PS for 30 days resulted in clinically meaningful improvements in joint pain at Day 30. We hypothesize that the combination of PS with PG may have beneficial effects on mood/motivation as well as reduced knee and hip pain that likely contributed to the beneficial effects of PG+PS in this study. Overall, these results indicate that PG+PS improves mobility and reduces joint pain in adults with moderate to severe joint pain.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

CONSENT AND ETHICAL APPROVAL

All experiments were examined and approved by the appropriate committee represented by D.M.Sc.,MD Eli Kassis (University of Copenhagen, Gentofte, Denmark, Protocol PGPS-101-01, Approved 14 April 2023) and were performed in accordance with ethical requirement and current standard as indicated by European Good Clinical Practice Guidelines and the 1964 Declaration of Helsinki. Informed and written consent was obtained from all participants prior to participation in the study.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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