PALL Life Sciences
Animal Health

Field Trial Summary Report

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A Letter from Dr. Jeffrey Schaffer

Regenerative medicine, and platelet therapy in particular, is an exciting new field of medicine. As the body of peer reviewed literature on the subject continues to evolve, many manufacturers of platelet concentration devices, such the new SECUROS Canine Platelet Enhancement Therapy (C-PET) from Pall Corporation, have embarked on clinical trials to study the efficacy of their products.

Justifying the funding for these clinical trials is an interesting process. For our recent controlled clinical study of C-PET, our decision to move forward was based on a large, uncontrolled field trial of C-PET that we had conducted over the previous year. Our goals were to (1) assess what indications veterinarians were treating with platelet therapy, (2) provide a preliminary assessment of the effectiveness of C-PET, and (3) gain insight into variables that we might want to explore in controlled clinical trials.

With the growing interest in our C-PET platelet therapy, we have decided to make our field trial data available to the public. As an observational dataset there are obviously limitations to how far we can interpret this information. However, given how relatively little is known about platelet therapy today, we believe that providing this information in the context of a careful analysis, without overstating the case, could be of benefit to the broader conversation taking place about platelet therapy in the veterinary community.

Even now after successfully completing our first clinical trial, we find ourselves continuing to return to the field trial as a source of inspiration for new clinical studies and improvements to our technology. We hope that you find yourself equally inspired.

Sincerely,

Jeffrey Schaffer, DVM

Methods of the Field Trial

The SECUROS C-PET field trial was a collaborative effort between Pall Animal Health, MWI Veterinary Supply, Inc., and veterinarians identified as either using or interested in using platelet therapy to treat lameness in dogs.

C-PET was provided to 31 veterinarians at no charge who treated 105 dogs between August 2010 and March 2011 with a single injection[1] of C-PET platelet concentrate prepared as per the manufacturer's instructions. In exchange for treatment, veterinarians and owners were asked to complete a Hudson Visual Analog Scale (HVAS), an 11 question survey of lameness, just prior to, and again 3 months after, treatment.

HVAS has been shown to correlate with force plate kinetics by Hudson[2], and was used in our C-PET clinical trial recently reported at the Veterinary Orthopedic Society meeting.[3] In this study, C-PET improved symptoms of lameness similar in magnitude to results obtained using another subjective scoring system, the Canine Brief Pain Index (CBPI[4]) as well as from force plate kinetics.

Pre- and 3 month post-treatment assessments were received from 90 patients. Not all records were complete for all patients, and as a result the sample sizes for some analyses vary. However, all of the available data was used for each analysis with no outliers removed.

C-PET Platelet Concentrate and Administration

Cell counts for whole blood and C-PET platelet concentrates are available for 71 patients. On average, the C-PET system produced 6.5mls of platelet therapy with a 3x concentration of platelets and a 2x concentration of white blood cells (table 1).

Table 1: Average characteristics of the C-PET platelet concentrate.

Volume of Platelet Concentrate	Platelets	WBC
6.5ml	3x	2x

Patients received a single intra-articular (IA) injection of C-PET platelet concentrate per treatment site, and 70% of patients were treated at multiple sites (e.g. both stifles were treated

[1] Three patients returned to receive a second treatment of C-PET but are not included in this analysis. Two received 2nd injections for new conditions, and one received a 2nd injection for the same condition.
[2] Hudson *et al.* Assessing repeatability and validity of a visual analogue scale questionnaire for use in assessing pain and lameness in dogs. *Am J Vet Res.* 2004 Dec;65(12):1634-43
[3] Fahie M, *et al*. "Clinical Outcome using Canine Platelet Enhancement Therapy (C-PET) for Osteoarthritis: A Prospective, Double-blinded, Controlled, Multi-center Study." 2012 Veterinary Orthopedic Society. Crested Butte, CO. March 7 2012. Podium Presentation.
[4] Drown *et al.* Ability of the canine brief pain inventory to detect response to treatment in dogs with osteoarthritis. *JAVMA*. 2008 Oct; 233(8): 1278-83.

at the same time). In total 56 stifles, 28 hips, 16 elbows, 3 carpi, 3 shoulders, and 2 tarsi were treated. For IA injections, the volume of C-PET platelet concentrate administered varied widely, within a given joint type and even when normalized to body weight. Veterinarians reported that the primary drivers guiding their decision on how much volume to inject IA were (1) the desire to fill the joint space without distending the joint capsule, and (2) palpable changes in resistance when injecting, at which point they would stop. As a point of reference, the average volume injected IA per joint type per body weight is reported in Table 2.

Table 2: Average volume of C-PET platelet concentrate injected IA per joint type per body weight.

Joint Type	# of Joints Injected	Average µl/lb Injected (± stdev)
Stifle	56	39 (± 24)
Hip	28	33 (± 15)
Elbow	16	35 (± 18)
Carpus	3	19 (± 14)
Shoulder	3	46 (± 13)
Tarsus	2	56 (± 29)

Overall Results: 84% of patients improved

No adverse effects of any kind were reported as a result of treatment with C-PET. Average HVAS scores at 3 months post C-PET treatment improved for 76 of the 90 patients (table 3). 24% of patients saw improvement between 0.1 and 1 point on the 10 point scale, 19% saw improvement between 1.1 and 2 points, and 41% saw improvement in excess of 2 points. About 16% of dogs treated saw no change or saw a continued decline in their HVAS scores.

Table 3: Change in HVAS at 3 months, and expanded details on the level of improvement.

Change in HVAS at 3 months	% of Population (# of dogs)
Improved	**84%** (76 dogs)
No change	**7%** (6 dogs)
Declined	**9%** (8 dogs)

Improvement (HVAS Month 3 – HVAS time 0)	% of Population (# of dogs)
>2	**41%** (37 dogs)
1.1 to 2	**19%** (17 dogs)
0.1 to 1	**24%** (22 dogs)

Overall, owners and veterinarians reported significant improvements 3 months post C-PET treatment in 10 of the 11 questions of the HVAS survey (no change in question 5, *Amount Active*). Averaging the 11 questions together, we see that the overall population saw a 2.2 point change in the median HVAS score on the 10 point scale. (Figure 1)

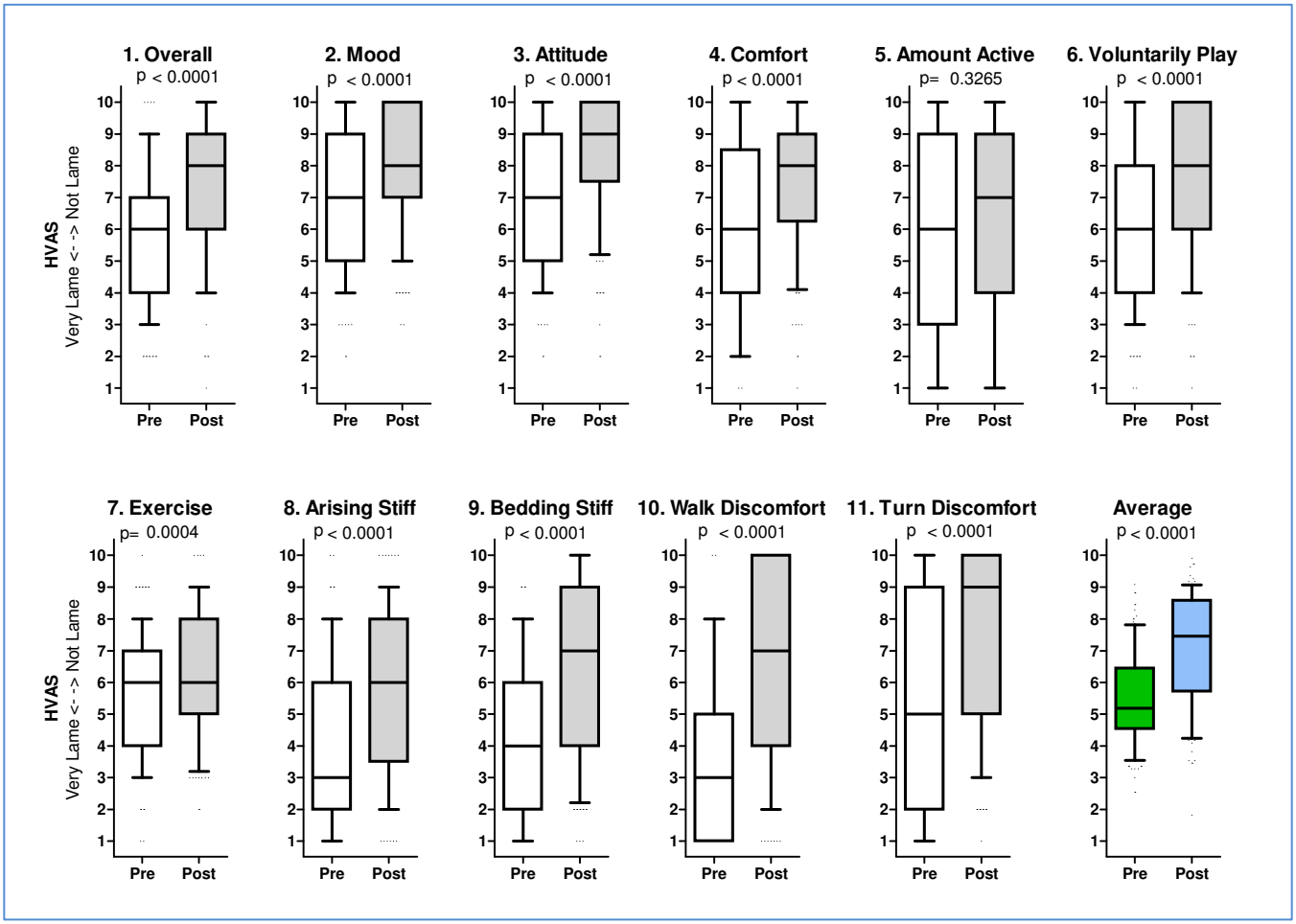

Figure 1: Pre-treatment and 3 months post-treatment scores for all 11 questions of the HVAS. N=90. Box plots represent median, 25th and 75th percentiles, and whiskers represent 10th and 90th percentiles. p-values for Wilcoxon sign ranked test.

There are two interesting trends in this dataset that make us more comfortable with the measurement tool. First, personality variables like *Mood* and *Attitude* tend to have higher pre-treatment starting values than the other metrics. This is consistent with our past experiences working with owners, who often remark that their canine companions are stubbornly-determined to enjoy life despite their pain. Second, variables that describe visual symptoms of lameness, like *Arising Stiff*, *Bedding Stiff*, *Walking Discomfort,* and *Turning Discomfort,* tend to have the lowest pre-treatment values. We would expect this as well, since visual signs of lameness are a major reason why dog owners would take their dog to the vet and subsequently have a chance to be enrolled in the trial.

Of all of the questions in the survey, we had hoped to see the largest improvement in the questions associated visual symptoms of lameness, and the results suggest that we do.

Indications Treated: compelling results in patients with OA

As an exploratory field trial there were recommendations, but no restrictions on the indications that veterinarians were allowed to treat with C-PET. The primary indication of interest was osteoarthritis (OA). As a degenerative disease we would not expect patients with OA to spontaneously improve, and this helps us interpret the OA field trial data in the absence of a treatment control. Interpreting the data from other indications can be complicated, and depends largely on whether or not the disease or injury is self limiting. With this understanding, we loosely classified the remaining indications as either cruciate ligament injuries (CL), or bone injuries (BI), and separated their data from patients with OA.

At face value, patients with OA and CL indications saw significant improvements in HVAS scores at 3 months. All 4 patients with BI indications saw improvement at 3 months, but the change did not reach a statistical level of significance (α=0.05) due to the low sample size.

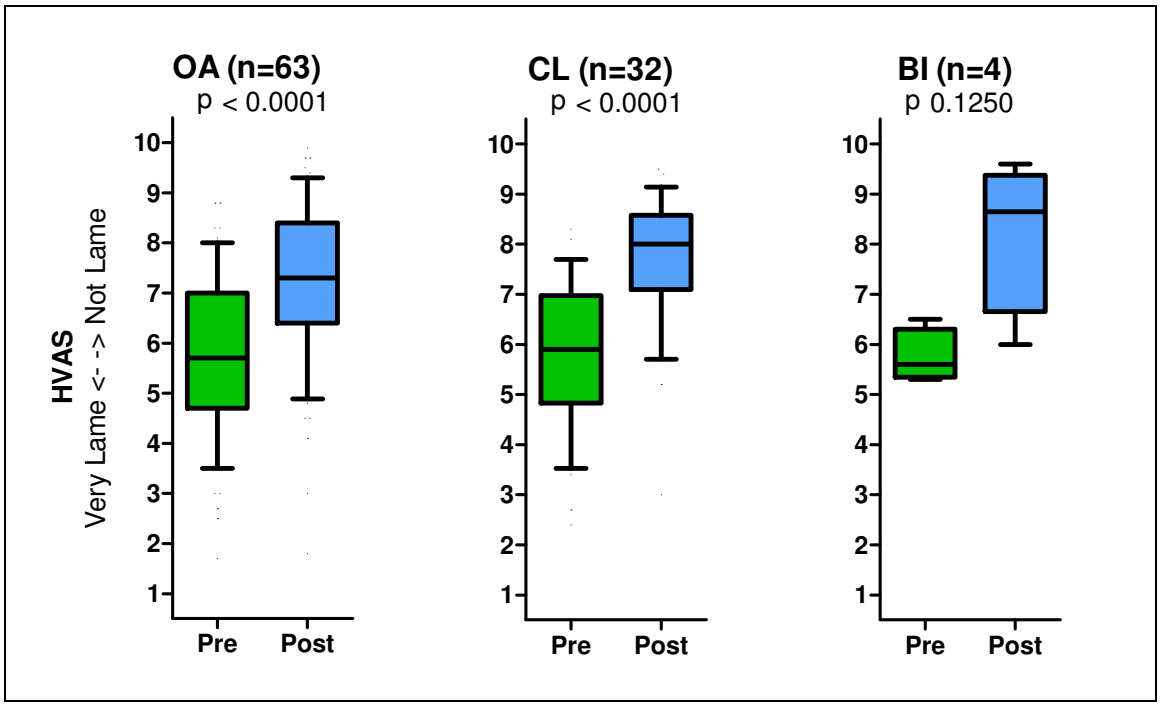

Figure 2: Average pre-treatment and 3 months post-treatment HVAS scores by indication. Wilcoxon sign ranked test p-values. Box plots of median, 25th and 75th percentiles, and whiskers 10th and 90th percentiles.

For patients with CL injuries, most veterinarians simply noted CCL (or ACL) as the indication being treated. However, enough veterinarians provided additional information that we are able to tease apart three small subpopulations. The first two

are patients whose records explicitly state that they suffered partial tears (N=6), and patients whose records explicitly state that they suffered full ruptures (N=4). Neither of these subpopulations reached statistical level of significance, however the observation that partial tears had better (less lame) pre-treatment HVAS scores than full ruptures provides a nice check on the measurement tool using a difference we would expect to see apriori. The third subpopulation is patients who explicitly received surgical intervention in addition to C-PET treatment. Perhaps not surprisingly, this group saw a significant improvement 3 months post treatment.

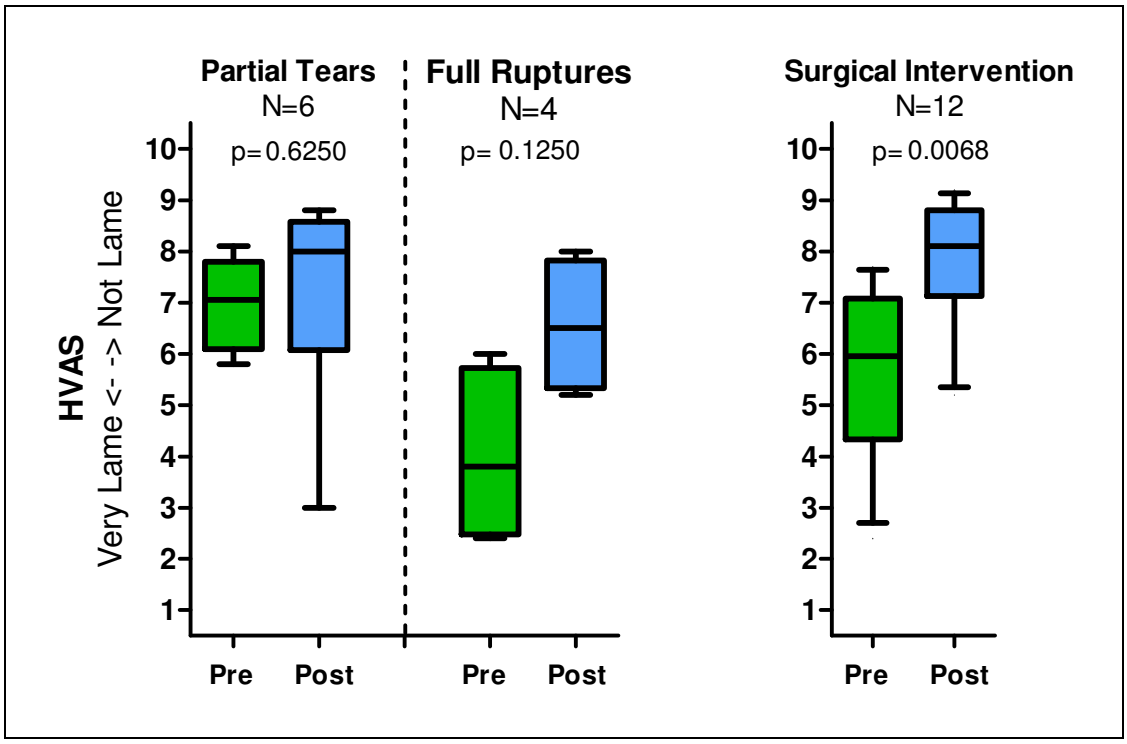

Figure 3: Average pre-treatment and 3 months post-treatment HVAS scores for patients with partial cruciate tears (Partial Tears), full cruciate ruptures (Full Ruptures), and patients who also received surgical intervention (Surgical Intervention). Wilcoxon sign ranked test p-values. Box plots of median, 25th and 75th percentiles, and whiskers of 10th and 90th percentiles.

Effect over Time: patients appear to still be doing well after 6 mo

A large number of veterinarians were generous enough to provide additional HVAS assessments beyond the 3 month time point. This data provides a rough picture of C-PET improvement for time intervals between 1 and 6 months (note: we are still collecting 6-12 month data).

It is impossible to run a proper time series analysis on this data since there are gaps in the patient records (e.g. there are only 5 patient records at month 4), but there are still several important observations that we can make. First is that the proportion of patients that saw improvement did not significantly change from month to month and are not significantly

different than the 84% observed at month 3 (Figure 4). Second is that the average change in HVAS was not different from month to month as determined by a Kruskal-Wallis test (Figure 5).

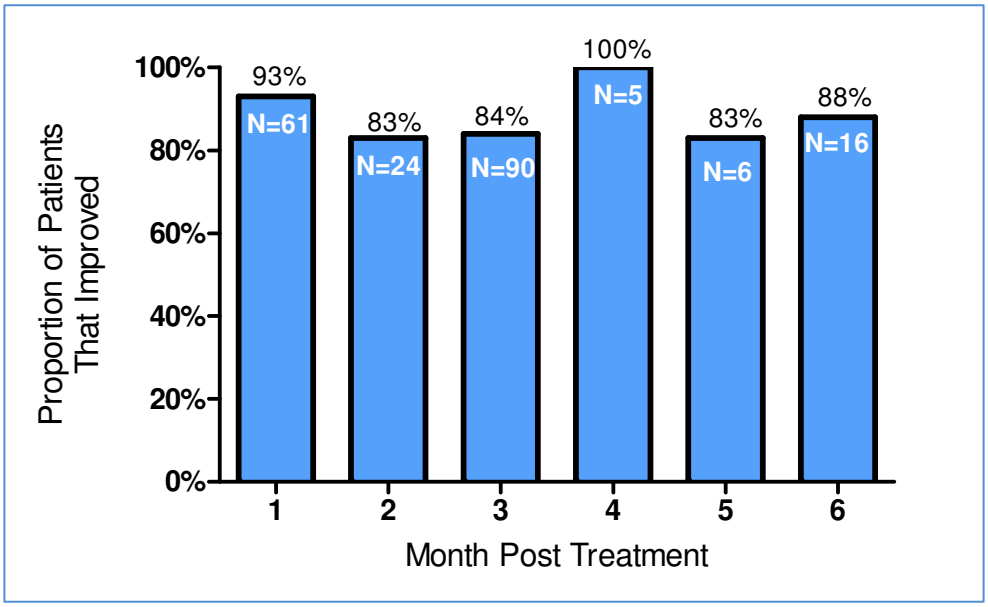

Figure 4: Proportion of patients that improved relative to their pre-treatment HVAS score by month.

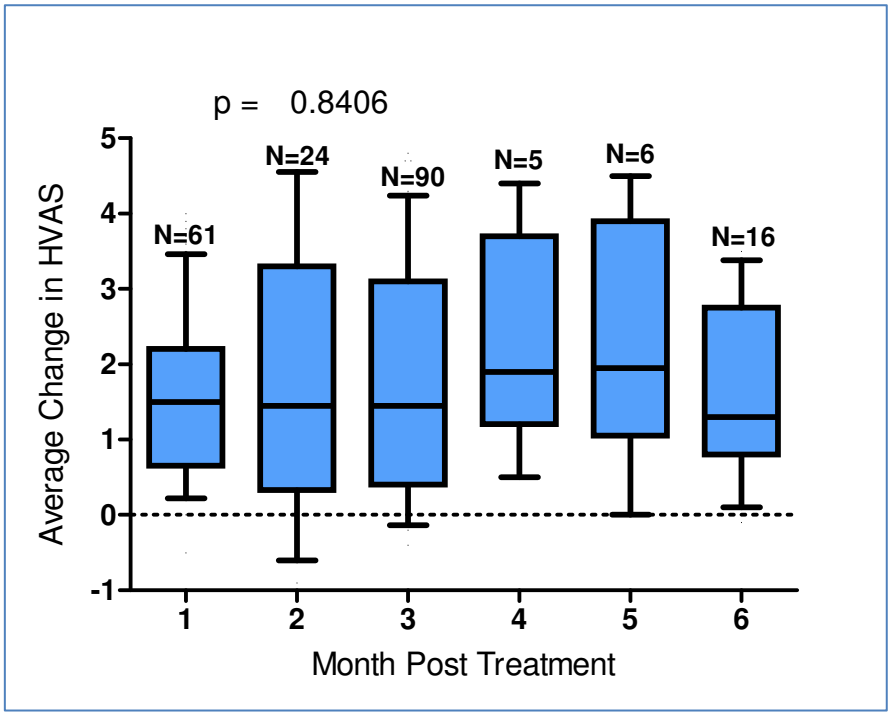

Figure 5: Average change in HVAS by month. Kruskal-Wallis p-value. Box plots represent median, 25th and 75th percentiles, and whiskers represent 10th and 90th percentiles.

In combination, these observations suggest that patients who see benefit from treatment with C-PET will likely see it within the first month, and that the results will likely be sustained for at least 6 months.

Age: No differences between age groups, but 11+ trending down

There were no differences between age groups when analyzing the average change in HVAS at 3 months using a Kruskal-Wallis test (nonparametric ANOVA, α=0.05). It is worth noting that the 11+ year old age group, while not statistically different, appears to improve less than the other age groups. It is possible that this trend is due to random variation, but it could also reflect a patient population that is simply more difficult to treat.

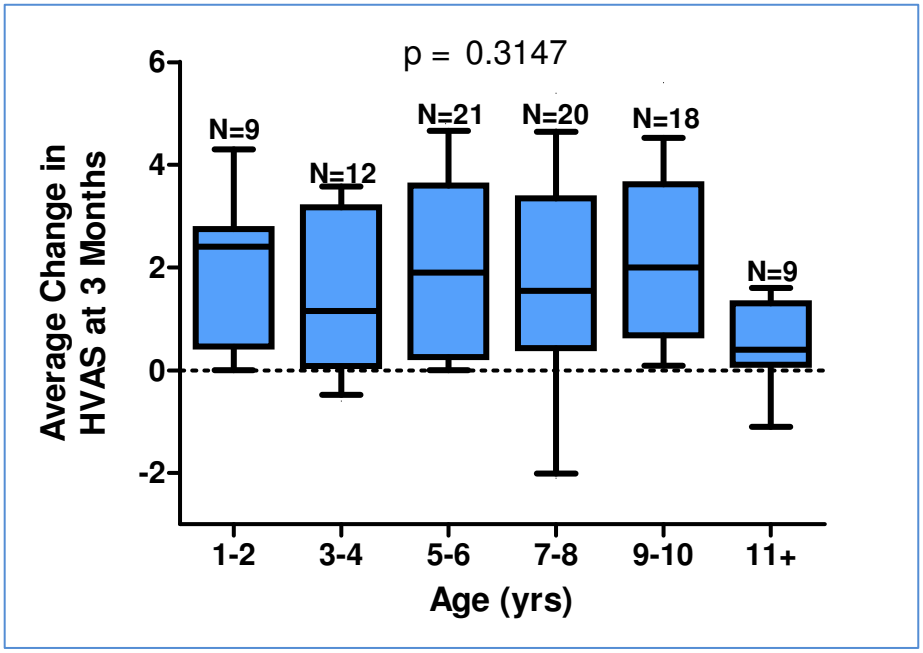

Figure 6: Average change in HVAS score at 3 months post-treatment. Kruskal-Wallis p-value. Box plots represent median, 25th and 75th percentiles, and whiskers represent 10th and 90th percentiles.

Summary

This observational dataset suggests that most patients saw improvement 3 months after a single treatment of SECUROS C-PET platelet therapy. These results are particularly compelling for patients treated for osteoarthritis since we would not expect this patient group to spontaneously improve without treatment. This observation was confirmed in our prospective, randomized control, double-blinded C-PET clinical trial, where patients treated with C-PET improved and no placebo effect for was observed in the saline treated controls. Our limited time-series analysis suggests that patients could see benefit as early as 1 month and that the patients are still doing well at least 6 months later. This observation matches anecdotal reports from owners who typically tell us they see benefit within the first 2 weeks of treatment, and reports from 3 out of the 4 owners whose dogs have reached 12 months (1 year) post treatment and have noted that their dogs are still doing well.

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