Monitoring and therapeutics of joint pain in dogs

Monitorizarea și terapeutica durerii în durerile articulare la câine

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Rezumat

Osteoartrita la câine este una dintre cele mai frecvente musculo-scheletice care în timp a devenit o problemă medicală şi financiară pentru proprietari. O mare problemă este tratarea câinilor care suferă de efectele secundare care apar ca o consecinţă a terapiei durerii în OA. În prezentul studiu au fost incluşi 19 câini de diferite rase cu diagnostice certe de osteoartrită (OA) şi / sau displazie de şold (DSC). În urmărirea stării de healing / ameliorare a stărilor de osteoartrită la câine s-au urmărit două obiective: a) Monitorizarea durerii cronice la câine în urma acupuncturii, prin intermediul unui chestionar consacrat, o scală descriptiv multifactorială; b) Influenţa acupuncturii asupra unor parametri sanguini în durerea cronică la câinii cu probleme. Lotizarea câinilor s-a făcut în funcţie de utilizarea anterioară a analgezicelor şi după formularul HCPI (Indicele de durere cronică din Helsinki (Helsinki Chronic Pain Index). Câinii au fost împărţiţi aleatoriu în două loturi: un lot care a primit tratamente cu acupunctură și antalgice şi un lot placebo (martor), care nu a primit tratament. Inițial animalele din studiu au fost supuse unor radiografii exploratorii, iar probe de sânge au fost recoltate pentru determinarea valorilor prolactinei şi serotoninei. Rezultatele au relevat că atât serotonina cât şi prolactina nu au suferit modificări cantitative semnificative pe durata studiului la câinii ce au primit acupunctură, cât şi la cei din grupul martor. Chestionarul folosit este un instrument valid de a fi folosit pentru evaluarea durerii cronice locomotorii la câini şi îl recomandăm pentru studiile clinice.

Abstract

Osteoarthritis in the dog is one of the most common musculoskeletal disorders that has become a medical and financial problem for the owners. A big problem is treating of the dogs suffering from the side effects that arise as a consequence of OA pain therapy. In the present study, 19 dogs of different races with definite diagnoses of osteoarthritis (OA) and / or hip dysplasia (DSC) were included. In pursuit of the condition of healing / amelioration of osteoarthritis conditions in dogs, two objectives were followed: a) the chronic pain monitoring of acupuncture dogs, via a well-established questionnaire, a multifactorial descriptive scale; b) Influence of acupuncture on blood parameters in chronic pain in the dogs. Dogs grouping was based on the previous use of analgesics and on the Helsinki Chronic Pain Index (HCPI). The dogs were randomly divided into two lots: a group that received acupuncture and antalgic treatments and a placebo (control) not treated. Initially the animals in the study were subjected to exploratory radiographs, and blood samples were collected to determine prolactin and serotonin values. The results revealed that both serotonin and prolactin did not undergo significant quantitative changes during the study in both dogs groups, receiving acupuncture and those in the control group. The questionnaire used is a valid tool for assessing chronic locomotor pain in dogs and we recommend it for the clinical trials.

Introduction

Since the emergence of the first methods of recognizing pain in animals, the acceptance of animals experiencing subjective pain has increased in recent years [Sanford et al., 1986; Robertson, 2002; Rutherford, 2002].

Painful canine behavior can be divided into three large categories [ACVA, 1998]:

A. The first category consists of common pain responses for all dogs and for most mammals: physiological responses (pupillary

change, cardiac rhythm, respiratory rate, etc.) and vocalizers in acute pain [ACVA, 1998].

However, there are significant differences between individuals and species within their behavior towards pain [Sanford,1992].

In the canine species, the genetic manipulation manifested by the crossbreeding of different breeds has resulted in the appearance of breeds of dogs with different behaviors, different characters and uses (e.g. guard dogs, fighting, sports, shepherd, work, hunting and dogs company).

As a result, there will also be typical pain responses, strictly related to the difference between breeds, already knowing that certain dog breeds are more resistant to pain, adding to their individual behavior [Sanford,1992].

B. The second category contains responses to socially acquired pain.

Like humans, dogs seem to learn the pain behavior of their parents from an early age [Sinclair et al., 2003].

Dogs can learn the behavior of pain from their owners (some owners encourage dogs to show pain, others do not) as they would learn from their own packs in a wild environment [Dobromylskyj et al., 1999].

C. The third category of pain behaviors is the ability to differentiate pain. It seems that the canine species can establish their own behavioral patterns of pain that they can overcome autonomous genetic responses and normal muscle aches caused by pain [Wall, 1992].

These dog capacities can be used to show more pain or less. For example, all dogs can become "very lazy" if they learn that they will gain more attention from their family members.

On the other hand, a dog who, in his or her environment, has pain, may not show any visible sign of pain when, for example, he is taken out of the hunt [Dobromylskyj et al., 1999; Flecknell and Waterman-Pearson, 2000; Fox și Johnston, 1997].

Wall (1992), in a study highlights the need to understand the relationship of a particular animal with its environment at a given time [Wall, 1992].

The owner's absence, odd odors and sounds from other animals are factors that can influence the way or the way an animal shows signs of pain.

In this regard, due to changes in dog responses to pain, it was suggested that the owner's observations on changes in dog behavior should be taken into account in pain assessment and used to evaluate treatment outcomes in clinical research [Hardie, 2000, ACVA, 1998].

Objective

The main objective of the study was to bring knowledge into non-pharmacological treatments for canine osteoarthritis (OAC). In order to evaluate the outcome of the treatment, we had to use different means of assessing chronic pain in the dog.

The main objectives of the study were:

1. To monitor the evolution of chronic pain in dogs with osteoarthritis treated with acupuncture using the Multiple Factor Descriptive Scale - Helsinki Chronic Pain Index (HCPI).
2. To monitor the influence of acupuncture on blood parameters in dogs with osteoarthritis.

Materials and methods

Animals

In the study were included 19 dogs of different races with definite diagnosis of osteoarthritis (OAC) and / or hip dysplasia (DSC). The inclusion criterion for the dogs in the study was the radiographic diagnosis of:

- severe hip displasia (CHD),
- unilateral or bilateral (classified as D or E) and
- osteoarthritis (OA), without presenting problems to another part of the body (eg elbow or back).

The basic symptoms that were taken into account were:

• pelvic limb disability,
• the difficulty of jumping,
• the difficulty of getting up / down,
• pain on the extension of the pelvic limb
• the presence of symptoms (at least 3 months).

The dogs showed a certain level of pain to meet the conditions of the Pain Index (HCPI) that was part of the chosen questionnaire.

The dogs were not included in the study:

• have received acupuncture or implant with pre-wired gold,
• have undergone previous surgery,
• have been treated with caprofen in the last 3 months,
• have had neurological problems,
• have had joint, systemic or infectious diseases.
• Dogs who, although over 20 kg, had short legs and were unable to analyze walking (eg Staffordshire bull terrier) were also rejected.

Of the total of 79 dogs initially tracked for admission tests, 60 dogs were excluded because either they had complex problems with old-style joints in the joints or suffered from various infectious diseases.

Both females and males were included in the study, some were sterilized and others were not, the selected dogs being aged 2 to 13 years.

The main breeds of dogs introduced into the study were:

- Labrador Retriever,
- German Shepherd and
- Samoyed,
- but also dogs of other breeds and mixed breed dogs.

Dog owners initially signed a written agreement for the participation of their dogs in the present study.

In Tables 1-4. the main data regarding the dogs participating in the experiment are summarized.

Table 1.

Dog breeds accepted in the study and their distribution on batches

Breed	No. dogs	Group acupuncture	Group control
Labrador Retriever	7	4	3
Ciobănesc German	4	3	1
Samoyed	2	1	1
Flat-Coated retriever	1	0	1
Doberman Pinscher	1	1	0
Rottweiler	1	1	0
Rough Collie	1	0	1
Lagotto Romagnolo	1	0	1
Mongrell	1	0	1
Total	**19**	10	9

Table 2.

Sexul câinilor din studiu

Sex	Dogs
Males (among them castrated)	6 (3)
Females (among them sterilised)	13 (4)

Table 3.

Categories of dogs according to the use of analgesics and HCPI * / at the first visit

Distribution according to HCPI and pain	No.	Group acupuncture	Group control
HCPI low / Analgesia reduced	9	5	4
HCPI high / Analgesia increased	6	3	3
HCPI high / Analgesia reduced	4	2	2
HCPI low / Analgesia increased	0	0	0
Total	**19**	10	9

***HCPI** = *Helsinki Chronic Pain Index*) (multifactorial descriptive scale)

The experimental protocol

The study was organized as a randomized prospective study.

The dogs were randomly divided into two:

• a group that received treatment and
• the placebo (control) group who did not receive treatment.

The main factors for selecting dogs for the study were:

• use of pain relief products (less than once a week = low pain killer use, weekly or more often = high use of painkillers)

• Multifactorial descriptive questionnaire HCPI (≤ 16 HCPI = low and ≥ 17 = HCPI increased). These values were monitored in owner-filled questionnaires.

The *treatment group* received 3 acupuncture treatments, the duration of a session was about an hour.

The placebo group received no treatment, the dogs were kept in the room where the acupuncture treatment was given to the dogs in the treatment group for an hour in the presence of the same veterinarian.

From previous studies it is known that applying acupuncture needles to non-acupoints has the same effect as acupuncture on acupoints, but the effects are much lower [Hulea and Cristina, 2012].

In this way we have no response to treatment so that in order not to influence the results we decided not to administer any treatment to the placebo group.

Table 4.

Dogs individuals accepted in this study

No.	Breed	Age	Sex F/M	Weight (Kg)	BCS (1-5)	Afection	Group A / M
1.	Labrador Retriever	13	(M)	31,9	3	E/D+OA!	A
2.	Lagotto Romagnolo	8	M	18,8	4	E/D+OA!	M
3.	Dobermann Pinscher	4,5	F	34,6	3	D/D+OA	A
4.	Rough Collie	12	(F)	21,0	3	E/E+OA!	M
5.	Labrador Retriever	6	F	28,4	4	D/D	M
6.	Ciobănesc German	8	F	31,3	5	(D/D)	A
7.	Labrador Retriever	9	F	37,3	4	D/D	M
8.	Samoyed	4	F	25,1	5	E/E+OA!	A
9.	Labrador Retriever	7	F	26,8	4	A/E+OA!	A
10.	Labrador Retriever	12	(M)	36,9	3	A/D+OA!	M
11.	Labrador Retriever	13	(F)	33,6	3	E/E+OA!	A
12.	German Sheperd	3	M	35,7	4	D/D	A
13.	Mongrell	2,5	(M)	55,4	3	E/E+OA!	M
14.	Samoyed	8,5	F	22,5	4	E/D	A
15.	Rottweiler	5	F	36,4	3	E/E+OA!	A
16.	German Sheperd	4,5	M	40,3	4	E/E+OA!	A
17.	German Sheperd	8	F	34,0	3	D/D	M
18.	Flat-Coated retriever	8	F	28,7	4	D/D	M
19.	Labrador Retriever	2	F	30,9	4	E/E + OA	M

Legend: *BCS = Body Condition Score (1-5); OA= Canine osteoartritis; OA!= Severe asteoartritis ; (M) = Male castrated; (F) = Female sterilised*

Each dog individual included in the study visited the clinic 5 times, with a duration of approximately one week between each visit, the time between the first and second visit, after applying the first treatment for 5-7 days. Before the first visit, the owners were forced to stop any treatment applied to the dogs until the study was completed.

After the second treatment of the dogs, the owners received the medication they would administer to the dogs, if in their opinion they would have experienced major pains.

The use of painkillers was expected to be higher for dogs in the placebo group.

Meloxicam is 1,1-dioxide of 4-hidroxy-2-methyl-N-(5-methyl-2-tyazolyl)-2H-1,2-benzothiazin-3-carboxamide, a non-steroidal anti-inflammatory drug (NSAID) of the oxicam class. Meloxicam is commercially formulated as: oral suspension, a viscous yellowish honey odor.

Each milliliter of oral suspension contains meloxicam equivalent to 0.5 or 1.5 milligrams and sodium benzoate (1.5 milligrams) as a preservative; tablets (1.0 or 2.5 mg / tablet) or solution for injection (5 mg / ml) (Figure 1).

Figure 1. Metacam® dog formulations: **a.** oral, **b.** tablets, **c**. injectable

Source:https://www.google.ro/search?rlz=1C1GCEA_enRO776RO776&tbm=isch&q=metacam&chips=q:metacam,g_1:dog&sa=X&ved=0ahUKEwjblvXhksfbAhVFDZoKHStRCIcQ4lYIJygA&biw=1366&bih=647&dpr=1.

Table 5 lists all work performed on dogs.

The canine pain index used by us was developed at Helsinki University, Finland by Dr. Anna Hielm-Björkman [Hielm-Bjorkman, 2003].

Table 5.

Performed manoperas

Treatment	Activities performed / visit
Initial **visit**	• Evaluation of lame • Clinical examination • Catheter mounting • Blood samples collection • Radiography of coxal-femoral articulation • Completing the questionnaires
First **treatment**	• Evaluation of lame • Clinical examination • Blood samples collection • Acupuncture / placebo / tratament1h • Completing the questionnaires
Seccond **treatment**	• Evaluation of lame • Clinical examination • Acupuncture / placebo / treatment 1h • •Completing the questionnaires
Third **treatment**	• Evaluation of lame • Clinical examination • Catheter mounting • Rest for 15-30 minutes • Blood samples collection • Acupuncture / placebo / treatment 1h • Blood samples collection • Filling in forms
Last **visit**	• Evaluation of lame • Clinical examination • Blood samples collection • Completing the questionnaires

Evaluation of lame

For this procedure, the dogs were run on a triangle-shaped trace of 3 holes, the ground was asphalt. The dogs moved in step and trap, during which time the visual assessment of the grade of lame was assessed with grades from **0-4** (0 signifying the lack of limp and 4 the dog's refusal to move).

Each dog made 3 laps in one direction and another 3 turns in the opposite direction.

Clinical examination

This examination consisted of heart rate and orthopedic and neurological summary examinations.

The region of the back has throbbed, throbbed and flexed the thoracic limbs, pelvis and fingers to see if the individual is experiencing pain at these levels.

The dogs were also weighted at each presentation and received a Body Condition Score, with scores from 1-5, ie:

1 = severe underweight,
2 = medium subponderability,
3 = normal weight,
4 = medium overweight,
5 = severe overweight.

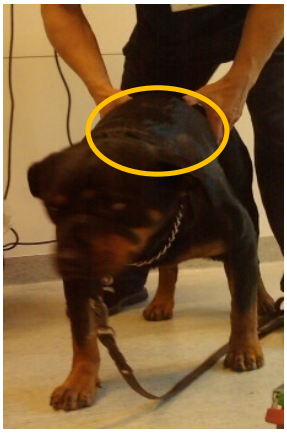

Figure 2. Performing of the clinical examination

Blood samples collection

To collect blood samples, we placed a 22G type catheter in the cephalic vein.

At the first visit, 6 ml of venous blood was collected to determine the baseline blood levels and observe eventual dysfunctions of the main organs.

At the second visit, 6 venous blood vaccinations were collected as follows:

- two 6 ml sera,
- one 9 ml with heparin,
- one 9 ml EDTA,
- one 3 ml with EDTA and
- a 2 ml vacutainer with heparin.

Samples were centrifuged, pipetted and then all stored at -80 ° C for further processing. At the first V1 presentation, blood samples were not taken, so they were not so stressed when they went to treatment.

At the second visit the dogs were cannulated and then left for 15-30 minutes at rest. Blood samples were taken before and after treatment to see changes in blood levels.

Radiografiile

Many dogs had recent x-rays and reports, and those who did not have been radiographized at the hips on their first visit (V1). Before being taken to the radiology room, the dogs were sedated i.m. butorphanol (Butordol 1mg/kgbw) and dexdomitor (Domitor 1mg/kgbw) and then, after the x-ray, their effect was reversed with atipamezole (Antisedan 1mg/kgbw). For the radiograph the dogs were placed in a ventro-dorsal position with caudally flexed pelvic limbs.

Factors assessed by ventro-dorsal radiographs of co-femoral joints in CHD dogs are shown in Table 6.

Table 6.

Factors assessed by ventro-dorsal radiographs of coxo-femoral joints in dogs with CHD

radiologic modification	Evaluated as
Norberg angle	>105° / 90°-105° / 75°-90° / 60°-75° / 45°-60°
Changes in femoral neck	
The length of the femoral neck	normal / short
Femoral neck	without exostosis / few / many
The metaphysical scar area	no changes / few / many
The shape of the femoral head	normal / slightly flattened / very flattened / extremely deformed
Changes in the acetabular margin	
The dorsal edge	without exostosis / few / many
The cranial edge	without exostosis / few / many
The caudal edge	without exostosis / few / many
The acetabular cavity	deep / shallow / unmodified
Incongruity of joint surfaces	without inconsistencies / few / total
Acetabular fossa	normal / slightly filled / full filled
Exodosis in the joint-anywhere	without exostosis / few / many
Fragments of bone in the joint	none / one / more

The survey

Owners have been asked about the pathological past of animals if they have recent X-ray hip joints, if dogs have problems with other parts of the body (elbow, back, knee) if they do not suffer from another illness, how long they suffer from osteoarthritis dysplasia, what are the symptoms and what treatments he received.

Table 7 presents the questionnaire used by us. Animal welfare questions have clarified whether dogs have had diarrhea, nausea, skin disorders, and whether their appetite has changed. These questions were included in the questionnaire to see if the treatment caused adverse reactions.

Owners also received questions about emergency medication if it was administered, how and when, in what dose.

Owners were also asked if they think they still need the medication for their dogs, the medication they used before this study, and now they have noticed differences in pain in dogs after the study.

The person who completed the questionnaire had to live with the dog to see the changes throughout the study.

Throughout the survey the questionnaires were completed by the same person for their own dog, it was not allowed to fill in the questionnaires by another family member.

The HCPI questionnaire, consisting of 11 questions, contained questions about owner's

personal data, dog's name, weight, age, race, sex, and was used to measure chronic pain.

On this basis, dogs scored **between 0-44.**

Table 7.

Used questionnaire after Dr. Anna Hielm-Björkman Helsinki University, Finland

QUESTIONNAIRE FOR PARTICIPANTS (OWNERS): HCPI 2

Date _____
Questionnaire no. _____
Dog's name _____
Diagnostic _____
Owner _____
Signature of the owner agreeing with the study: _____

Just tick **one answer** - one that **best** describes your dog's condition.

1. The condition of the dog is:				
Very alert	alert	nor alert, nor indiferent	indifferent	very indifferent
☐	☐	☐	☐	☐
2. The dog is playing:				
Very willing	Willing	Less willing	Wery low willing	It's not playing
☐	☐	☐	☐	☐
3. The rate at which the dog shows his pain (audible, icneli, screams, crying, etc.):				
Never	Rare	Sometimes	Often	Very often
☐	☐	☐	☐	☐
4. The dog runs / goes:				
Very easy	Easy	Nor easy, nor difficult	With difficulty	With great difficulty
☐	☐	☐	☐	☐
5. Symmetrical movements (legs movements in diagonal and at the same time 'jogging'):				
Very easy	Easy	With some difficulty	With great difficulty	At all
☐	☐	☐	☐	☐
6. Gallop (speed movement):				
Very easy	Easy	With some difficulty	With great difficulty	At all
☐	☐	☐	☐	☐
7. The jump (e.g. in the car, on the armchair, etc.)				
Very easy	Easy	With some difficulty	With great difficulty	At all
☐	☐	☐	☐	☐
8. The dog sits in the decubitus:				
Very easy	Easy	With some difficulty	With great difficulty	At all
☐	☐	☐	☐	☐
9. The dog rises from the decubitus:				
Very easy	Easy	With some difficulty	With great difficulty	At all
☐	☐	☐	☐	☐
10. The dog moves after a longer rest period:				
Very easy	Easy	With some difficulty	With great difficulty	At all
☐	☐	☐	☐	☐
11. The dog moves after a period of heavy exercise				
Very easy	Easy	With some difficulty	With great difficulty	At all
☐	☐	☐	☐	☐

Vet's notes::

_____
_____
_____

The **value 0** indicated the absence of chronic pain and the minimum limit set by us in the HCPI questionnaire in this study was **12** because we wanted chronic pain to be large enough to make visible treatments between treatments.

Acupuncture treatments

The technique consisted of dry needle acupuncture applied to the treatment group.

Once those were placed in the acupuncture points, they were left in situ for 45 minutes. The dogs in the placebo group were monitored in the treatment room for one hour without receiving any treatment.

Results and discutions

Chronic pain monitoring in dogs following acupuncture via HCPI

Table 8 shows the dogs and experimental protocol in the study, and figure 3 shows the evolution of placebo and acupuncture after HCPI scores.

This study consisted of chronic pain monitoring by owners through pain questionnaires (HCPIs) in dogs suffering from dysplasia of the hip and osteoarthritis who were treated with acupuncture.

During the study, owners completed 6 times the questionnaire on their dogs.

The HCPI questionnaire contains **11 questions** about mood, behavior and dog locomotor, each with a descriptive ordinal scale from **0-4 where** 0 indicates the absence of chronic pain.

The results revealed that the minimum limit set in the HCPI questionnaire in this study was 12, with the peak of chronic pain **being 44.**

Table 8.

Dogs participating in the study and HCPI evaluation values for visits

No.	Breed	Group A/M	HCPI V-1	HCPI V0 T1	HCPI V1 T2	HCPI V2 T3	HCPI V3
1.	Labrador Retriever	A	38	36	32	28	27
2.	Lagotto Romagnolo	M	34	33	32	32	31
3.	Dobermann Pinscher	A	34	33	32	30	28
4.	Collie	M	44	44	44	44	44
5.	Labrador Retriever	M	18	17	16	17	17
6.	German Sheperd	A	20	18	16	14	14
7.	Labrador Retriever	M	19	18	17	18	18
8.	Samoyed	A	41	39	39	37	35
9.	Labrador Retriever	A	32	32	30	27	25
10.	Labrador Retriever	M	41	41	40	40	39
11.	Labrador Retriever	A	44	42	40	40	38
12.	German Sheperd	A	18	18	16	15	13
13.	Mongrell	M	42	41	40	41	41
14.	Samoyed	A	21	20	18	16	14
15.	Rottweiler	A	42	42	41	39	37
16.	German Sheperd	A	16	15	12	9	8
17.	German Sheperd	M	12	11	10	10	10
18.	Flat-Coated retriever	M	13	13	13	13	13
19.	Labrador Retriever	M	22	21	20	19	19

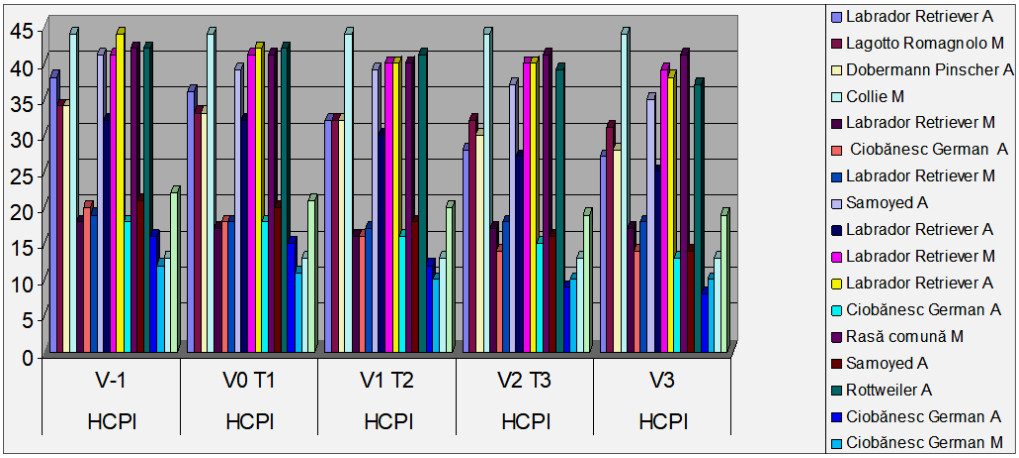

Figure 3. Evolution of treatment in the placebo group and the acupuncture group after HCPI scores

In a recent study conducted on a Beagle dog showing involuntary defecation and osteoarthritis in the co-femoral joints. Radiological examination revealed mild hip dysplasia and osteoarthritis. The dog was treated by acupuncture, three sessions per week for 3 weeks with the master's agreement.

After the dog's treatment, the clinical examination showed less severe pain and was willing to move, while involuntary defecation became voluntary by the recurrence of anal sphincter reflex. This result suggests that acupuncture can be used as an alternative therapy to reduce the effects of co-femoral osteoarthritis and stimulate anal sphincter reflex in the dog [Abrudean et al., 2016; Aragon et al., 2007; Brown et al., 2008; Hulea and Cristina 2012; Hulea et al., 2013].

Managing the dog's body weight is a particularly important factor in dogs with hip dysplasia and osteoarthritis placed under acupuncture treatment. There are currently a multitude of medicines available for pain, including non-steroidal anti-inflammatory drugs, gabapentin, amantadine and tramadol.

There are also methods of preventing the occurrence of chronic pain consisting in providing the appropriate mineral intake and controlling the body weight of the dogs. In a study using visual scales for dog owners, the authors postulated that the owners were able to observe and record the beneficial changes that occurred during the treatment [Bennett et al., 1996; Johnston, 1998].

Influence of acupuncture on blood parameters in chronic pain in dogs with locomotory problems

This study included the same 19 dogs of different breeds and suffering from locomotor disorders: osteoarthritis and / or hip dysplasia.

The dogs were divided into two groups, a batch of 10, who received acupuncture treatment and a group of 9, who received no treatment. Those in the treatment group were treated with dry acupuncture at three of the five visits performed throughout the study.

Prolactin and **serotonin** were dosed before acupuncture therapy throughout the study during visits (V-1, V0, V2, V3).

In one of the dogs accepted in the study, we only collected one sample of blood on the first visit (V-1). Analysis of samples for the quantitative detection of prolactin and serotonin was done by the ELISA method.

Prolactin

It is a peptide hormone produced by anterior pituitary (lactotrophic cells), myometrium and breast tissue. Structural prolactin consists of 199 amino acid residues, weighing approximately 23 kDa. In mammals, prolactin has a mammotropic effect (causes the growth of mammary glands), lactotropic (stimulates breast milk synthesis) and libido. Prolactin acts in a cytokine-like manner, being an important modulator of the immune system.

It can also influence angiogenesis, hematopoiesis and is involved in the regulation of blood clotting.

Prolactin exhibits significant variations in the amino acid sequence from one species to another. In terms of residues, the prolactin canine differs from human prolactin by about. 60%.

Secretion of prolactin by the pituitary gland is inhibited by the hypothalamus by the release of a prolactin-inhibitory factor (PIF). Although dopamine has long been considered to be the PIF molecule, there appears to be another particular peptide with prolactin inhibitory activity.

The release of prolactin is certainly stimulated by different peptides as release hormones, especially thyrotropin (TRH) and vasoactive intestinal peptide (VIP).

Estrogens and progesterone also appear to play a role in the secretion of prolactin and neurogenic factors that influence its release.

The most important role of prolactin is to stimulate the growth of the mammary gland and stimulate lactation.

Milking and suckling are immediately followed by increased serum prolactin levels.

During pregnancy, the level of prolactin in the blood increases slightly, but increases significantly during breast-feeding.

Prolactin has a wide variety of other physiological actions:

- affect the hydro-electrolytic balance;
- metabolism;
- gonadal function;
- is an important hormone of stress;
- and appears to play a role in maintaining the interethane interval between the bitch.

Studies have shown that in dogs with pituitary-dependent hyperadrenocorticism blood levels of prolactin were higher than in healthy animals. During false gestation, the level of prolactin is also increased [Ley et al., 1991].

Alkaloid therapy, such as e.g. bromocriptine decreases prolactin levels, lowers lactation and diminishes maternal behavior.

Serotonin

It is an indole-derived amine, also called benzopyrrole, a heterocyclic organic compound composed of two nuclei: an aromatic condensed with a pyrolytic nucleus; is found in the fraction of distilled tar at temperatures of 220-2600 ℃.

It has also been identified in volatile oils extracted from orange or jasmine flowers. Its name is given by the combination of indigo and oleum due to the fact that it was isolated by treating the indigo with oily mixture (SO3 and H2SO4). Serotonin is found in plant and animal tissues.

Serotonin acts as a neurotransmitter or as a chemical mediator.

Chemical mediators are chemical subchannels found in the nervous system (in the brain), through which the transmission, modulation and amplification of nerve impulses in the synapses occurs, interferes with the production of sleep in mental and emotional processes (depression and anxiety, obsessive-compulsive disorder), in motor functions, in thermoregulation, in regulation of blood pressure, in the act of vomiting and in hormonal functions.

În figura 4. este redată evoluția prolactinei la câinii din studiu și statistica descriptivă aferentă.

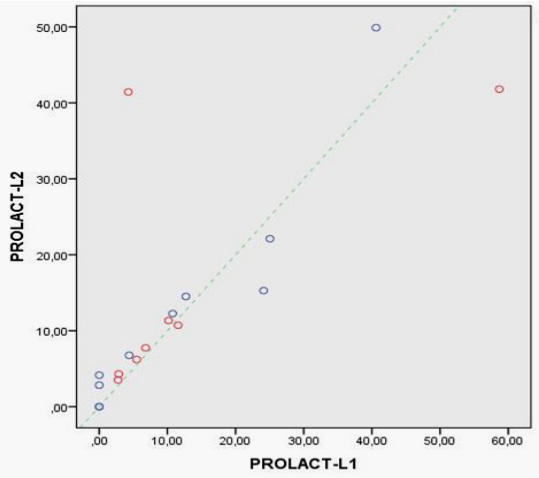

Figure 5. Evolution of prolactin in dogs in the study (**blue** = acupuncture / **red** = control group)

NPar test

Descriptive statistics									
Grupul	Hormon	N	Media	D.S.	Minim	Maxim	Procentual		
							25%	50% (Median)	75%
Acupunctură	Prolactină-L1	10	11,7650	14,02711	,00	40,61	,0000	7,5850	24,3450
	Prolactină -L2	10	12,7800	14,94744	,00	49,90	2,1225	9,5100	16,9875
Martor	Prolactină -L1	8	12,8275	18,80294	2,77	58,68	3,2175	6,1500	11,2250
	Prolactină -L2	8	15,8838	16,12030	3,51	41,80	4,7850	9,2300	33,9150

NPar test

Descriptive statistics								
Grup	N	Media	D.S.	Minim	Maxim	Procentual		
						25%	50% (Median)	75%
Prolactină-L1	18	12,2372	15,81271	,00	58,68	2,0775	6,1500	15,5600
Prolactină -L2	18	14,1594	15,09323	,00	49,90	3,9900	9,2300	16,9875
Treatment group	19	1,47	,513	1	2	1,00	1,00	2,00

Wilcoxon Signed Ranks Test

Ranks					
Grup	Hormon	Categorie	N	Medie	Sumă
Acupunctură	Prolactina-L2 - Prolactina-L1	Negative Ranks	2[a]	6,00	12,00
		Positive Ranks	6[b]	4,00	24,00
		Ties / legături	2[c]		
		Total	10		
Martor	Prolactina-L2 - Prolactina-L1	Negative Ranks	2[a]	5,00	10,00
		Positive Ranks	6[b]	4,33	26,00
		Ties / legături	0[c]		
		Total	8		
a. Prolactina-L2 < Prolactina-L1					
b. Prolactina-L2 > Prolactina-L1					
c. Prolactina-L2 = Prolactina-L1					

Mann-Whitney Test

Ranks				
Hormon	Grup	N	Medie	Sumă
Prolactina-L1	Acupunctură	10	9,20	92,00
	Martor	8	9,88	79,00
	Total	18		
Prolactina-L2	Acupunctură	10	9,10	91,00
	Martor	8	10,00	80,00
	Total	18		

Test Statistics

Grupul	Categoria	Prolactină-L2 - Prolactină-L1
Acupunctură	Z	-,840[b]
	Asymp. Sig. (2-tailed)	,401
	Exact Sig. (2-tailed)	,461
	Exact Sig. (1-tailed)	,230
	Point Probability	,039
Martor	Z	-1,120[b]
	Asymp. Sig. (2-tailed)	,263
	Exact Sig. (2-tailed)	,313
	Exact Sig. (1-tailed)	,156
	Point Probability	,031
a. Wilcoxon Signed Ranks Test		
b. Based on negative ranks.		

Test Statistics[a]		
Categoria	Prolactină-L1	Prolactină-L2
Mann-Whitney U	37,000	36,000
Wilcoxon W	92,000	91,000
Z	-,268	-,356
Asymp. Sig. (2-tailed)	,789	,722
Exact Sig. [2*(1-tailed Sig.)]	,829[b]	,762[b]
Exact Sig. (2-tailed)	,813	,742
Exact Sig. (1-tailed)	,406	,371
Point Probability	,018	,014
a. Grouping Variable: Treatment group		
b. Not corrected for ties.		

În figura 5 este redată evoluția serotoninei la câinii luați în studiu și statistica descriptivă aferentă.

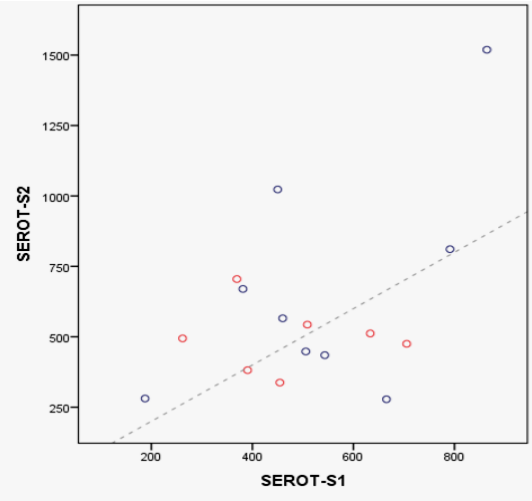

Figura 5. Evoluția serotoninei la câinii din studiu (albastru = lot acupunctură / roșu = lot martor)

5. Concluzii

- Chestionarul folosit este un instrument valid folosit pentru evaluarea durerii cronice locomotorii la câini şi poate fi folosit în studiile clinice, în care proprietarii sunt utilizaţi.
- Acest chestionar poate fi considerat ca o metodă de anamneză. Clinicienii ştiu că anamneza reprezintă 50% din datele care conduc la punerea diagnosticului
- Atât serotonina cât şi prolactina nu au suferit modificări cantitative semnificative pe durata studiului la câinii ce au primit acupunctură, cât şi la cei din grupul martor

NPar test

Descriptive statistics									
Group	Hormone	N	Average	D.S.	Min.	Max.	Procentual		
							25%	50% (Median)	75%
Acupuncture	Serotonin-S1	9	538,70	208,848	187	864	415,66	505,70	728,24
	Serotonin-S2	9	670,02	400,906	278	1519	357,82	565,67	916,99
Control	Serotonin-S1	7	474,78	154,648	262	705	369,31	454,37	633,35
	Serotonin-S2	7	492,62	118,821	338	705	381,45	494,34	543,17

NPar test

Descriptive statistics								
Hormone	N	Average	D.S.	Min.	Max.	Procentual		
						25%	50% (Median)	75%
Serotonin-S1	16	510,74	184,124	187	864	383,57	482,84	657,34
Serotonin-S2	16	592,41	315,641	278	1519	394,78	503,13	696,11
Total groups	19	1,47	,513	1	2	1,00	1,00	2,00

Wilcoxon Signed Ranks Test

Ranks					
Treatment	Hormone	Category	N	Media	Sumă
Acupunture	Serotonin-S2 - Serotonin-S1	Negative Ranks	3[a]	4,67	14,00
		Positive Ranks	6[b]	5,17	31,00
		Ties	0[c]		
		Total	9		
Control	Serotonin-S2 - Serotonin-S1	Negative Ranks	4[a]	3,25	13,00
		Positive Ranks	3[b]	5,00	15,00
		Ties	0[c]		
		Total	7		
a. Serotonin-S2 < Serotonin -S1					
b. Serotonin -S2 > Serotonin -S1					
c. Serotonin -S2 = Serotonin S1					

Mann-Whitney Test

Ranks				
Hormone	Group	No.	Average	Sum
Serotonin -S1	Acupuncture	9	9,22	83,00
	Control	7	7,57	53,00
	Total	16		
Serotonin -S2	Acupuncture	9	9,11	82,00
	Control	7	7,71	54,00
	Total	16		

Test Statistics[a]		
Category	Serotonin-S1	Serotonin S2
Mann-Whitney U	25,000	26,000
Wilcoxon W	53,000	54,000
Z	-,688	-,582
Asymp. Sig. (2-tailed)	,491	,560
Exact Sig. [2*(1-tailed Sig.)]	,536[b]	,606[b]
Exact Sig. (2-tailed)	,536	,606
Exact Sig. (1-tailed)	,268	,303
Point Probability	,033	,035
a. Grouping Variable: grup		
b. Not corrected for ties.		

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