

**CONTRIBUTION OF THE DOPPLER ECOGRAPHIC EFFECT TO EMPHASIZE THE  
EMERGENCE AND DEVELOPMENT STAGES OF VASCULARIZATION IN THE BONE  
CALLUS DURING FRACTURE HEALING AT DIFFERENT ANIMALS SPECIES**

Ph.D. Stud. Corina Stan<sup>1</sup>, Prof. Ph.D. Constantin Vlăgioiu<sup>1</sup>, Ph.D. Liviu Harbuz<sup>2</sup>,  
Assoc. Prof. Ph.D. Laur Manea<sup>3</sup>

<sup>1</sup>Faculty of Veterinary Medicine Bucharest,

<sup>2</sup>Veterinary College Romania,

<sup>3</sup>Valahia University of Târgoviște

E-mail: corina\_stan78@yahoo.com, vlagiouconstantin@fmvb.ro, office@cmv.ro, laur\_manea@yahoo.com

**Abstract**

*Feedback noninvasive, without adverse effects on patient health and healing in the early stages, are of great use to monitor the evolution of a fracture. The purpose of this study is to highlight the fitness and usefulness of normal echography technique (B-mode) and Doppler's, in terms of their use for determining and monitoring the dynamics of bone healing, as a result of osteosynthesis at some animals, like dogs and cats. The revascularization of bone callus can be visualized using Doppler ecography, and we can establish four periods during the fracture healing, when the Dopplers signals are present until these are null. As imaging method, however, Doppler ultrasound finds application only in certain stages of healing, depending on the weight of the neoformation of vascular tissue prebone. Using ecografic and radiologic methods, we can monitoring the process of callus formation and development and its weaknesses.*

Key words: doppler, callus, fracture healing, ecography, radiology

**1. INTRODUCTION**

Currently, animal disease problems can be investigated and monitored, like human medicine, in new ways, non-invasive diagnosis to establish a more secure, more detail, came to help the clinician, treatment, and not least the subject of the investigation, animal.

The main objective of these studies to expand the opportunities for imaging, based on radiological examination and radiographic images reflected by them in pursuit of healing a fracture, compared to the reported ultrasonographic technique.

Feedback noninvasive, without adverse effects on patient health and healing in the early stages, are of great use to monitor the evolution of a fracture. There are situations where valuable animals are subjected to intricate surgery that occurred in accidents and assessing the normal course of bone fracture is a priority for both the doctor and for the owner.

Monitoring of the development of bone callus is important to monitor the recovery of bone complications, caused by improper application of a technique of fixation, a septic infection or any other change that occurred during the period in which healing takes place, which may finally compromise the functionality of the affected limb. Doppler ultrasound, as a relatively new method of investigation because of high costs of specialized equipment, offers real-time investigation, the

dynamic process of vascularization in the callus bone and adjacent tissues.

The advantage of the method is the fact that the patient does not require anesthesia, a method is not painful, does not create discomfort, can be repeated whenever necessary and provide valuable information about the formation and normal development.

As imaging method, however, Doppler ultrasound finds application only in certain stages of healing, depending on the weight of the neoformation of vascular tissue prebone.

Since the healing of bone fractures during time consuming, and sometimes up to six months is necessary to note that during this period may occur a number of diseases that can disrupt the patient's health status. The current protocol includes a priority for clinical investigations hematological and biochemical widely used by most clinicians, but a real-time interpretation of the results by time since surgery is more comprehensive.

Thus, variations of different parameters, aspects of normal bone during the recovery process, which should be interpreted as such, properly reported to the animal's physiological state at the time.

**2. MATERIAL AND METHOD**

We have been studied a number of 21 cases, 11 dogs and 10 cats. The animals under study were

between 6 months and 10 years. The fractures were located in the limbs, healed by conventional surgical methods (plate with screws, centromedular with nails or rods). The ultrasound is an veterinary power Doppler for small animals, and the transducer is a linear one, with 7.5, 10.0 and 12.5 Mhz frequency.

Diagnostic sonography (ultrasonography) is an ultrasound-based diagnostic imaging technique used to visualize subcutaneous body structures including bonds, tendons, muscles, joints, vessels and internal organs for possible pathology or lesions. In physics, the term "ultrasound" applies to all acoustic energy (longitudinal, mechanical wave) with a frequency above the audible range of human hearing. The audible range of sound is 20 hertz-20 kilohertz. Ultrasound is frequency greater than 20 kilohertz. Sonography can be enhanced with Doppler measurements, which employ the Doppler effect to assess whether structures (usually blood) are moving towards or away from the probe, and its relative velocity. By calculating the frequency shift of a particular sample volume, for example flow in an artery or a jet of blood flow over a heart valve, its speed and direction can be determined and visualized. The Doppler information is displayed graphically using spectral Doppler, or as an image using color Doppler (directional Doppler). The Doppler effect (or Doppler shift), named after Austrian physicist Christian Doppler who proposed it in 1842, is the change in frequency of a wave for an observer moving relative to the source of the wave. An echocardiogram can, within certain limits, produce accurate assessment of the direction of blood flow and the velocity of blood and cardiac tissue at any arbitrary point using the Doppler effect. One of the limitations is that the ultrasound beam should be as parallel to the blood flow as possible.

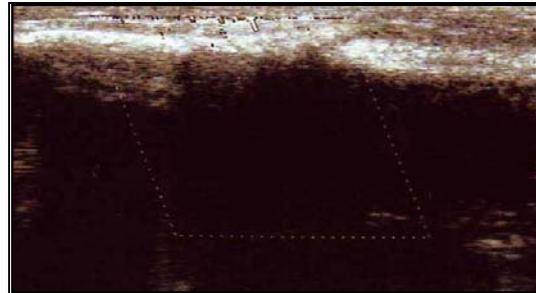
The radiographer is a Philips, Practix 33 plus.

### **3. RESULTS AND DISCUSSIONS**

The methods employed studied for all 21 cases were the normal echography (B-mode), Doppler echography and radiology. These tests were repeated from 10 to 14 days, until the fractures were healed, and the zero day was the first day after surgery. Thus, there were settled four major periods in which the Doppler signals had considerable variations. Time postoperatively was divided into periods of 12 days.

The first period, from zero day up to day 12 (a week and a half), was established as a period of

minimum activity even no activity of the neovascular system at the level of the fracture's focus and adjacent tissues. We are able to see this process in the image figure 1.



**Figure 1. Day 10 after surgery**

In this imaging is illustrated a detail of Doppler activity at the level of fracture in the first ten days after surgery.

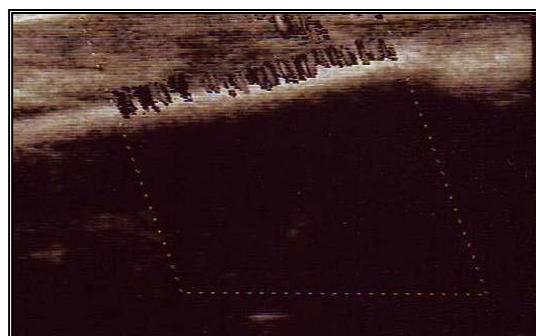
We have also an X-ray image of the process, at the same time, in figure 2.



**Figure 2. Radiological image after surgery, first 10 days**

The second period, from day 13 to day 24 (three weeks almost), was a period of maximum activity of the vascular processes in the fracture's focus.

This intense process is visible in figure 3, a detail of Doppler window.



**Figure 3. Doppler image in the day 16 after surgery**

During the 3<sup>rd</sup> period, from day 25 to day 36 (3 weeks to 5 weeks), Doppler signals decrease in intensity and number, until the last period, the period of total healing of bone. So that in the 4<sup>th</sup>

period that we established, namely from day 37 to end of healing process, until days 72-84, the signals become very weak and finally void zero. We see in figure 4 a decrease of vascular signals in the day 41 after surgery and a detail also of the repairing process.



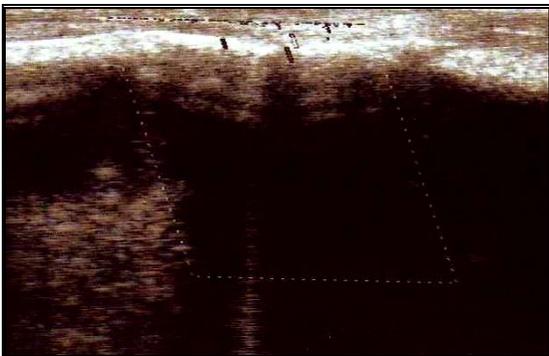
**Figure 4. Day 41 of healing process**

Figure 5 illustrated a image with callus formation, at 6 weeks after reconstruction of the bone.



**Figure 5. Radiographic image of the healing process, at 6 weeks after surgery**

In the image figure 6 we can see a very poor image of the vascularisation at the end of healing process, in the day 67 after surgery, and in the last picture, fig. 10 a detail of this image.



**Figure 6. Doppler image at 67 days of callus developing process**



**Figure 7. Radiograph image at 65 days after surgery**

The X-ray is the correspondent of the ultrasonographic image, at 2 and half months after surgery in figure 7.

#### **4. CONCLUSIONS**

- The normal process of bone healing at the level of the fracture, can be traced some stages using Doppler technique. One can establish four periods during the normal process, when the variation of blood at the fracture healing place are major and obvious.
- The neovascularization in the callus is a normal process during the period of healing, and variation of number and intensity of vessels from the process, can be visualized and monitoring using Doppler technique.
- Using of ultrasonographic and radiologic methods, we can monitoring the process of callus formation and development.

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**COMPARATIVE ASPECTS OF ECOTOXICITY BETWEEN DIFFERENT ENVIRONMENTAL COMPARTMENTS AS EVIDENCED BY SPECIFIC BIOMARKERS**

Assoc. Prof. Ph.D. Laur Manea<sup>1</sup>, Assoc. Prof. Ph.D. Gabriel Dima<sup>1</sup>, Lect. Ph.D. Daniela Cristiana Alexandrescu<sup>1</sup>, Lect. Ph.D. Iulia Manea<sup>1</sup>, Lect. Ph.D. Loredana Beatrice Neagu Frăsin<sup>1</sup>, Assist. Prof. Ph.D. Aurelia Corina Cosac<sup>1</sup>, Ph.D. Stud. Corina Stan<sup>2</sup>

<sup>1</sup>Valahia University of Târgoviște

<sup>2</sup>University of Agronomical Sciences and Veterinary Medicine, Bucharest

E-mail: laur\_manea@yahoo.com, gabidima@valahia.ro, danaalex@yahoo.com, yulia1081967@yahoo.com, loredana\_beatrice2003@yahoo.com, aureliacorinacosac@yahoo.com, corina\_stan78@yahoo.com

**Abstract**

*Biomarkers can be defined in ecotoxicological meanings as the body's biological response to the action or the impact of a toxic chemical that leads to a deviation from normal. Under this definition are analyzed only at an individual level those considered relevant and specific biological responses in ecotoxicological point of view. In this study, trace the development of ALAD (delta-aminolevulinic acid dehydratase), specific biomarker of lead in fish and ruminant mammals to be correlated with the concentrations of lead in the environment. State environmental health of the biotic and abiotic thus indirectly can thus be monitored and may show clear histotoxicologic and citotoxicologic alterations type generated by the chemicals. Comparison of data from monitoring biomarkers and potentially toxic chemicals from abiotic environmental compartments (water, air, soil) can make a major contribution to the establishment of tolerable levels of concentrations of these toxic substances. On the other hand, emphasizing the functional levels of biomarkers help to clarify the etiopathogenic mechanisms of many ecotoxicological processes, risk to human and the environment health, thereby establishing the relationship between cause and effect.*

Keywords: biomarker, metals, lead, enzymes, ecotoxicological, spectrometry.

**1. INTRODUCTION**

The main reason is the knowledge of biomarkers of human health problems and also the biotic environment caused by metals, usually with high atomic weight such as lead (Pb), mercury (Hg), cadmium (Cd) and arsenic (As), with negative implications, but also from low atomic mass, the best known example is aluminum (Al). Decontamination for toxic metals is different from organic toxins: because toxic metals are elements, they cannot be destroyed. Toxic metals may be made insoluble or collected, possibly by the aid of chelating agents. Toxic metals can bioaccumulate in the body and in the food chain. Therefore, a common characteristic of toxic metals is the chronic nature of their toxicity. This is particularly notable with radioactive heavy metals such as thorium, which imitates calcium to the point of being incorporated into human bone, although similar health implications are found in lead or mercury poisoning. The exceptions to this are barium and aluminum, which can be removed efficiently by the kidneys.

Ecotoxicological chemicals risk create problems as a result of relatively recent highly accelerated industrial and economic development, being less severe in developed countries where more stringent measures have been introduced in recent decades.

These chemicals persist in the environment because they have natural origin and are biodegradable.

Exposure to metals is the occurrence of adverse consequence of chronic (Akesson et al.2006, Chen et al.2007).

Target populations of these studies were changed to bioindicator species monitoring. It is necessary however, biological studies to be correlated with the presence of these substances in environmental compartments (water, air, soil).

In the case of human fetuses are studied as they are at high risk from exposure to metals, can be observed combined effects of these metals. Important to investigate is the degree of bioaccumulation of chemical elements in various bodies mentioned above, placental transfer of these metals and their relationships with maternal and fetal circulation.

The specificity of the biomarkers is an important criterion in ecotoxicological investigations. And non-specific biomarkers in the division are necessary because the effects of bioindicator organisms by specific biomarkers (biological responses) are safe and effective means of specifying the toxic chemicals involved in metabolism.

Identifying specific biomarkers is possible in kingdoms, plant and animal.

Thus, seleno-or fluorosensible plants, plant selenoproteins. They are excellent biomarkers for