UNIVERSITY OF MEDICINE AND PHARMACY "CAROL DAVILA", BUCHAREST

DOCTORAL SCHOOL

FIELD OF MEDICINE

PhD Thesis Summary

Scientific Coordinator:
University Professor Doctor: CÎRSTOIU FLORIN CĂTĂLIN

PhD Candidate:
STANCIU ION-LUCIAN

2025

UNIVERSITY OF MEDICINE AND

PHARMACY "CAROL DAVILA", BUCHAREST

DOCTORAL SCHOOL

FIELD OF MEDICINE

PREDICTIVE EVALUATION OF THE DETERMINANT FACTORS OF ASEPTIC BONE NECROSIS

PhD Thesis Summary

Scientific Coordinator:
University Professor Doctor: CÎRSTOIU FLORIN CĂTĂLIN

PhD Candidate:
STANCIU ION-LUCIAN

2025

Table of contents

Introduction

Aseptic bone necrosis (avascular osteonecrosis) is a severely disabling condition characterized by bone cell death due to the interruption of microcirculation, typically diagnosed at advanced stages when structural damage becomes irreversible. This thesis starts from the premise that the disease results from a multifactorial interaction between thrombotic dysfunctions, behavioral factors (chronic alcohol use and smoking), and iatrogenic factors (glucocorticoid therapy), which act synergistically on bone microcirculation. Early identification and control of these elements can significantly reduce the need for arthroplasty and improve functional prognosis.

Globally (especially in Europe and the USA), numerous studies have reported an increasing incidence, correlated with the high prevalence of risk factors and longer life expectancy. In Romania, there are no national registries or coherent multicenter studies, and current data are fragmented. However, it is estimated that the incidence is comparable to the European average, considering the national profile of alcohol consumption, smoking, and corticosteroid use. This lack of systematic data justifies the need for a detailed regional investigation, aiming to outline an etiopathogenic picture that reflects the realities of the Romanian medical system.

The objectives of the thesis are:

1. To characterize the regional risk profile through comparative epidemiological analysis and identify the prevalence of hereditary (thrombophilias) and acquired thrombotic factors.
2. To develop a clinical predictive model capable of stratifying the population according to susceptibility to osteonecrosis, useful in screening programs.
3. To assess the impact on quality of life (QoL), both preoperatively and postoperatively, using a standardized questionnaire, in order to correlate functional, psychological, and socio-professional status with disease severity and type of surgical intervention.

The central hypothesis is that the coexistence of a prothrombotic status and harmful habits accelerates ischemic progression by disrupting bone homeostasis, and early intervention on these factors can prevent extensive structural resorption.

The study database was built in Microsoft Excel (version 2501) and statistically analyzed using IBM SPSS Statistics (version 30.0.0). The following were performed:

- descriptive analyses (means, medians, standard deviations);
- T-tests and ANOVA for intergroup comparisons;

- customized algorithms for additional calculations.

For the imaging component, high-resolution MRI protocols were applied, complemented by clinical correlation and paraclinical analyses (inflammatory and coagulation biomarkers). All data were collected in accordance with ethical standards, with approval from the Ethics Committee of Ilfov County Emergency Hospital, and patient information was fully anonymized.

The general section synthesizes the pathophysiology of aseptic bone necrosis, emphasizing the role of microthrombosis, recent international classifications (Ficat-Arlet, ARCO), the relationship between behavioral factors and microvascular dysfunction, the coagulation cascade, and the genetic mechanisms involved.

The personal contributions section details the data collection and analysis methodology, the development of the predictive model based on weighted risk scores, the evaluation of quality of life using a validated questionnaire correlated with radiological stage, and the proposal of a management guide adapted to Romania's healthcare resources, including screening recommendations for chronic alcohol consumers and smokers.

The research aims to validate a minimal set of clinical and paraclinical markers capable of indicating an increased risk of osteonecrosis before the appearance of classic radiological signs, to create personalized prevention protocols focused on lifestyle modification and selective thromboprophylaxis, and to demonstrate significant quality of life improvement through chronic pain reduction, increased mobility, and socio-professional reintegration.

This work addresses an urgent need to clarify the determinants of osteonecrosis and to standardize clinical practice at the national level. By integrating epidemiological research, genetic analyses, and behavioral profiling into the proposed predictive model, it allows for early interventions that reduce morbidity, healthcare costs, and socioeconomic impact. Through its integrative approach, the thesis provides a solid framework for implementing a personalized medicine program for aseptic bone necrosis and serves as a practical guide for Romanian clinicians dealing with this complex condition.

Chapter 1. Anatomy and Physiology of Bone Tissue

Bone is the most resilient natural biomaterial, capable of rapidly adjusting its shape and density to the mechanical and metabolic demands of the body [1]. The skeletal system serves multiple roles: structural support for musculature, protection of vital organs, maintenance of body

shape, reservoir for ions and growth factors, acid-base buffer, hematopoietic organ, and—according to recent discoveries—an endocrine gland [2,3].

Functionally, the skeleton is divided into the axial skeleton (skull, spine, thorax) and the appendicular skeleton (limbs, shoulder, and pelvic girdles). In adults, excluding sesamoid bones, it comprises 213 bones: 74 axial, 126 appendicular, and 6 ossicles of the middle ear [3].

Morphological classification distinguishes four groups:

- long bones (e.g., clavicle, humerus, femur),
- short bones (e.g., carpals, tarsals, patella),
- flat bones (e.g., skull, sternum, scapula, pelvis),
- irregular bones (e.g., vertebrae, sacrum, coccyx).

Internal structure varies based on mechanical demand: vertebrae consist of approximately 25% compact bone and 75% spongy bone, whereas the radial diaphysis has the opposite ratio (95% compact / 5% spongy).

Compact bone forms the outer layer of bones, containing Haversian osteons with concentric lamellae around Haversian canals, interconnected by Volkmann canals, providing high impact resistance. Spongy bone, located centrally (epiphyses, vertebrae), forms a trabecular network aligned along force directions, offering strength with minimal material.

Long bones feature a thick cortical diaphysis with a medullary canal (red marrow in children, yellow in adults), spongy epiphyses covered by articular cartilage, and intermediate metaphyses where the growth plate is located in youth. The external surface is covered by periosteum, a site of muscle attachment and source of osteoblasts for appositional growth. The internal surface is lined by endosteum, a metabolically active layer where turnover is negative, leading to canal widening with age.

The skeleton contains five major cell types, accounting for approximately 10% of total bone volume [4]:

1. **Osteoprogenitor cells** – Mesenchymal stem cells located in the periosteum, endosteum, and vascular canals; can differentiate into osteoblasts or, through fusion, into osteoclasts.
2. **Osteoblasts** – Responsible for forming the organic matrix (osteoid), secrete alkaline phosphatase and FGF-23 hormone; key transcription markers include RUNX2 and osterix. Once their role is complete, they can: (a) undergo apoptosis, (b) become bone-lining cells, (c) be incorporated as osteocytes.

3. **Osteocytes** – Mature osteoblasts embedded in lacunae, interconnected by canaliculi; they comprise 90–95% of all bone cells. They act as mechanosensors and regulate remodeling via signaling molecules (sclerostin, nitric oxide, IL-6).
4. **Osteoclasts** – Multinucleated cells derived from the monocyte-macrophage lineage, specialized in bone resorption; their differentiation is regulated by the RANKL–RANK interaction and M-CSF.
5. **Bone-lining cells** – Derived from osteoblasts, they remain inactive on the bone surface but can reactivate when stimulated.

Bones receive about 10–20% of the cardiac output [5]. The nutrient artery enters the diaphysis, bifurcates within the medullary canal, and supplies the inner two-thirds of the cortex. The periosteal network nourishes the outer layer and is dependent on adjacent muscles. Secondary branches enter the epiphysis and metaphysis, while fenestrated capillaries within Haversian/Volkmann canals enable nutrient diffusion—no viable cell lies more than 300 μm from a blood vessel.

From a morphological perspective, type H capillaries (CD31^high / endomucin^high) are rich in angiogenic factors and surrounded by osteoprogenitor cells. Their density decreases with age, correlating with reduced bone formation. Blood flow may be centrifugal (medullary → periosteal) or, after nutrient artery injury, reversed (centripetal), due to periosteal collaterals—an adaptive arteriogenesis phenomenon. Perfusion declines with age, and vascular remodeling (hypertrophy, hypotrophy, or eutrophy) adapts to metabolic demands and mechanical or endothelial stimuli (e.g., nitric oxide, endostatin).

Chapter 2. Avascular Bone Necrosis

Osteonecrosis, also known as aseptic or avascular bone necrosis, is a pathological process in which bone cells die due to a reduction in local blood flow. This ischemia gradually compromises the trabecular architecture of the bone, eventually leading to collapse of the articular surface. The condition represents a significant public health issue, with notable economic and social impact - particularly because it predominantly affects young adults under the age of 50 [6–8].

The classification of osteonecrosis depends on its anatomical location, and multiple classification systems exist, not all of which have clear clinical relevance. For aseptic necrosis of

the femoral head alone, we can mention several: the Ficat-Arlet classification (by far the most widely used in clinical practice), the Steinberg classification, ARCO 2014, and the revised ARCO 2019 classification [6].

The mechanisms leading to aseptic bone necrosis are still not fully understood, although several hypotheses are gaining increasing support. The most widely accepted theory describes a multifactorial process involving genetic predisposition, metabolic dysfunctions, and local factors that impair circulation—such as mechanical stress, increased intraosseous pressure, or vascular injury—that converge to reduce bone perfusion. Another theory attributes the disease to vasoconstriction caused by corticosteroid excess, along with dyslipidemia induced by these drugs, promoting the formation of fat emboli. In all these scenarios, the common denominator remains the disruption of blood flow and, consequently, impaired microperfusion in the affected bone [7,9,10].

Diagnosis of osteonecrosis is based on patient history, clinical examination, and imaging investigations. While history and clinical signs may raise suspicion, definitive confirmation is achieved through imaging. In advanced stages, simple radiographs may be sufficient, but in early stages, magnetic resonance imaging (MRI) is the gold standard. Bone scintigraphy, though less sensitive than MRI, remains useful as it can detect multiple concurrent foci of osteonecrosis in a single investigation [11,12].

Given the variety of possible locations for bone necrosis, treatment options can be summarized as follows:

- in advanced stages (Ficat-Arlet III–IV): the only option is total joint arthroplasty.
- in stage II (pre-collapse): core decompression drilling, with or without adjuncts (e.g., mesenchymal stem cells, vascularized bone grafts, osteoinductive factors), or unloading osteotomy.
- in early stages: conservative measures such as reduced weight-bearing, hyperbaric oxygen therapy, shockwave therapy, pulsed electromagnetic fields, and targeted drug treatment (e.g., anticoagulants/antiplatelets for associated thrombophilia, statins for dyslipidemia) [13–15].

Chapter 3. Thrombophilias

The coagulation cascade is a sequence of proteolytic reactions that primarily occur on the surface of platelets and is triggered when they adhere to damaged vascular endothelium. It includes two main pathways — intrinsic and extrinsic — which originate differently but converge and interact during the hemostatic process. The ultimate goal is to stop bleeding by stabilizing the platelet plug within a fibrin mesh [16].

Thrombophilia is a state of hypercoagulability that can lead to thrombotic events such as deep vein thrombosis, pulmonary embolism, pregnancy loss, coronary events, or aseptic bone necrosis [17].

II. PERSONAL CONTRIBUTIONS

Chapter 4. Working Hypothesis and General Objectives

I chose this doctoral topic because aseptic bone necrosis, often underdiagnosed and of multifactorial etiology, requires a comprehensive multidisciplinary approach. The research aims to thoroughly analyze the biological and clinical profile of the patient, focusing on predictive factors within a framework adapted to the realities of the Romanian healthcare system.

The central hypothesis is based on the idea that aseptic osteonecrosis occurs through the synergistic action of a prothrombotic state and harmful habits such as chronic alcohol consumption and smoking. The interaction of these factors promotes microthrombi formation in bone microcirculation, subsequently leading to ischemia and tissue necrosis [18].

Early identification of risk factors and their integration into a clinical-paraclinical predictive model may slow disease progression, reduce complications, and limit the need for major orthopedic interventions.

This study provides a detailed analysis of the etiopathogenic factors of aseptic osteonecrosis, emphasizing prothrombotic status and the impact of chronic alcohol and tobacco use. It also evaluates the disease's effect on quality of life across three dimensions: functional, emotional, and social.

Chapter 5. General Research Methodology

To support the working hypothesis, the research was structured along three main directions:

1. The first research direction involved an analytical observational case-control study: the hemostatic profile of patients with aseptic osteonecrosis was compared to that of a control group without any history of bone necrosis. The aim was to detect potential prothrombotic changes that could represent a biological susceptibility factor for non-traumatic bone ischemia.
2. The second research direction employed a descriptive-analytical cross-sectional study to outline the demographic and behavioral profile of patients with aseptic osteonecrosis. Variables such as age, sex, living environment, and biological parameters were correlated with risk behaviors (alcohol and/or tobacco use) to assess their influence on disease severity and to identify subgroups with increased vulnerability.
3. The third research direction consisted of a descriptive observational cross-sectional study aimed at evaluating the quality of life in patients with aseptic osteonecrosis.

The research was conducted between January 2023 and March 2025 at Ilfov County Emergency Hospital, a multidisciplinary medical facility. Patients included in the study were assessed clinically, biologically, and functionally according to approved protocols and methodologies.

The database was organized in Microsoft Excel (Microsoft 365), where clinical, biological, and demographic information was compiled. Using integrated descriptive functions (AVERAGE, MEDIAN, STDEV), central and dispersion parameters were calculated, and the generated graphs facilitated visual analysis of data distributions. For inferential statistics, IBM SPSS Statistics 30.0 was used, while custom calculations requiring tailored algorithms were performed in JavaScript, allowing the analysis to be adapted to the dataset's specifics and the methodological requirements of the study.

All patients included in the studies signed an informed consent agreement, including consent for data processing, which was used exclusively for scientific research purposes.

Chapter 6. Study I - Procoagulant Status as a Risk Factor in Aseptic Osteonecrosis: An Observational Analytical Case-Control Study

Introduction

The aim of the first research direction was to compare the hemostatic profile of a group of patients diagnosed with aseptic osteonecrosis with that of a control group without any history of

necrotic bone pathology. The working hypothesis was that a procoagulant status may play a significant role in biological susceptibility to osteonecrosis [19,20].

Material and Methods

The study focused on the genetic and biochemical analysis of hemostasis, comparing 28 patients with osteonecrosis to 38 individuals without the condition. The evaluation concentrated on the frequency of procoagulant mutations, serum levels of anticoagulant proteins, and homocysteine concentrations, aiming to identify a potentially altered prothrombotic status.

The research question formulated for this study was "Are there significant differences in the hemostatic profile between patients with aseptic osteonecrosis and those without this pathology?". This question rigorously adheres to the FINER criteria (Feasibility, Interest, Novelty, Ethics, Relevance), thus representing a valid and pertinent direction for scientific investigation.

The following variables were collected:

- Demographic data: age, sex, place of residence (urban/rural);
- Genetic and biochemical parameters:
  - Factor V Leiden mutation (G1691A);
  - Prothrombin gene mutation G20210A (Factor II);
  - MTHFR gene mutations (C677T and A1298C);
  - PAI-1 675 4G/5G polymorphism;
  - Antithrombin III (normal range: 83–128%);
  - Protein C (70–140%);
  - Protein S: males (74–146.1%), females (54.7–123.7%);
  - Homocysteine (4.3–11.1 μmol/L);
- Procoagulant status: classified as normal, moderate, or severe.

Results

The case group included 17 male patients and 11 female patients, while the control group consisted of 20 male and 18 female patients.

When analyzing the age group distribution between the two groups, it was observed that the 41–60 age group was slightly more prevalent in the case group (16 patients) compared to the control group (14 patients), and this may represent a higher-risk group, possibly due to the accumulation of vascular and metabolic risk factors.

Based on place of residence, a higher frequency of patients from urban areas was noted, which may reflect greater access to medical services, and consequently, a higher diagnostic rate.

Statistical analysis showed that almost all women in the case group (with osteonecrosis) presented with a procoagulant status (10 out of 11 patients). Among men, the distribution was more balanced, though there was a higher proportion of severe procoagulant status in the case group (7 out of 17 patients in the case group, compared to 1 out of 15 in the control group).

The following factors influencing procoagulant status were analyzed and compared across the two groups: Factor V Leiden mutation, Prothrombin 20210 mutation (Factor II), MTHFR mutations C677T and A1298C, PAI-1 gene polymorphism (675 4G/5G), Antithrombin III, Protein C, Protein S, Homocysteine levels,

Discussions

Our results highlight a statistically significant correlation between Factor V Leiden mutation and osteonecrosis, reinforcing the hypothesis that a procoagulant status plays a major role in the pathogenesis of this bone condition. This conclusion aligns with data previously reported in the literature [21,22]. As a result, incorporating Factor V Leiden testing into the evaluation protocol for osteonecrosis patients gains both prognostic and diagnostic relevance.

Age group distribution analysis of the mutation showed a possible association between the presence of Factor V Leiden, younger age, and bone disease. The Chi-square test confirmed the statistical significance of this relationship. These findings suggest that, at least in younger patients, identifying heterozygous or homozygous genotypes of Factor V Leiden may have important implications in the etiopathogenesis of osteonecrosis.

In the study group, the heterozygous variant of the prothrombin G20210 mutation was identified only in patients with osteonecrosis and was completely absent in the control group. This distribution suggests a potential etiopathogenic link between the mutation and the development of osteonecrosis, considering that the mutation leads to a hypercoagulable state and predisposes to recurrent thrombotic events.

The heterozygous C677T mutation of the MTHFR gene appeared more frequently in patients with an increased procoagulant status—particularly in moderate cases—and seemed to be present in severe cases only among patients with osteonecrosis. Although it is neither exclusive nor as strongly predictive as Factor II or Factor V mutations, this mutation may act as a modulating thrombophilic factor, especially in individuals with genetic predisposition.

The distribution of the MTHFR A1298C mutation did not show a clear association with the severity of procoagulant status, either in the control or osteonecrosis groups. In its heterozygous form, the mutation has low predictive value for severe hypercoagulability or osteonecrosis risk. Therefore, its pathogenic role seems limited, likely dependent on the coexistence of other mutations that enhance the prothrombotic tendency.

The PAI-1 4G/4G genotype was identified only among osteonecrosis patients, predominantly in those with moderate or severe procoagulant status. This distribution suggests that the polymorphism may have predictive or pathogenic significance in bone ischemia.

Comparing antithrombin III levels showed that women in the osteonecrosis group had a lower average, indicating a weakened anticoagulant profile. In men, the variation was broader, and the presence of very low values in the osteonecrosis group suggests the possibility of severe antithrombin deficiency contributing to ischemic pathogenesis.

In the osteonecrosis group, Protein C levels as low as 47% were recorded—well below the reference lower limit (70%). Such a marked deficit may indicate severe congenital thrombophilia or an acquired deficiency in a proinflammatory or prothrombotic context, mechanisms that can promote intraosseous microthrombosis and tissue ischemia.

By contrast, the lowest value in the control group was 52%, indicating that Protein C deficiency can occur without ischemic manifestations, though with less severity.

Analysis of Protein S levels by sex in the two groups showed that although values remained within reference ranges, women tended to have lower concentrations. This feature may indicate a relatively higher risk of triggering the coagulation cascade, potentially contributing to the onset of osteonecrosis, especially when combined with other risk factors.

Homocysteine level measurement was the final test in the hemostasis profile applied to patients. The sex-specific distribution of results revealed that women with osteonecrosis had higher levels of this prothrombotic marker, indicating an additional biological risk. The elevation of homocysteine levels in osteonecrosis patients—more pronounced in women—supports the hypothesis of its involvement in the ischemic mechanisms of the disease.

Conclusions

The study revealed a significant link between severe procoagulant status and the occurrence of osteonecrosis, analyzing both genetic and functional factors involved in hemostasis.

Among these, the homozygous Factor V Leiden genotype, the prothrombin G20210A mutation, and the PAI-1 gene polymorphism emerged as the most important indicators for assessing procoagulant tendency and bone ischemia. Additionally, deficiencies in natural anticoagulant proteins (C, S, and antithrombin III) can further disrupt the balance of the coagulation cascade, while in elderly patients, hyperhomocysteinemia may intensify the procoagulant state.

For future research directions, it is necessary to expand the study sample to validate these correlations and to develop screening algorithms that integrate the patient's hemostatic profile with environmental and behavioral factors.

Chapter 7. Study II – Analysis of Demographic, Behavioral, and Biological Factors in Aseptic Osteonecrosis: A Cross-Sectional Observational Study

Introduction

A cross-sectional, observational study with both analytical and descriptive components was designed, with the primary objective of evaluating the procoagulant status in patients with aseptic osteonecrosis and correlating it with various demographic and behavioral factors. The aim is to identify potential patterns of biological and lifestyle-related risk that may influence the severity or progression of the bone condition.

Material and Methods

The study was based on the clinical hypothesis that certain coagulation abnormalities, whether genetic or acquired, may lead to a predisposition for thrombotic events in the bone microcirculation, with bone ischemia as the final consequence. A total of 28 patients diagnosed with aseptic osteonecrosis, both clinically and through imaging, were included in the study.

A database was created for the patients, integrating demographic, clinical, biological, and behavioral variables, and blood samples were collected to perform an extended hemostasis profile:

- Genetic markers:
  - Factor V Leiden mutation (G1691A)
  - Prothrombin gene mutation (Factor II, G20210A)
  - MTHFR gene polymorphisms: C677T and A1298C
  - PAI-1 gene polymorphism (675 4G/5G)
- Functional parameters:
  - Antithrombin III (reference range: 83–128%)

- Protein C activity (reference range: 70–140%)
- Protein S activity (Males: 74–146.1%, Females: 54.7–123.7%)
- Plasma homocysteine levels (reference range: 4.3–11.1 μmol/L)

Based on the results obtained, patients were classified by procoagulant status into three distinct categories:

- Normal
- Moderate hypercoagulability
- Severe hypercoagulability

A behavioral questionnaire was used, consisting of 10 questions about alcohol consumption and 2 questions regarding tobacco use. Based on the accumulated score, patients were categorized into the following risk levels:

- Score I (0–7 points): Low risk
- Score II (8–15 points): Moderate risk
- Score III (16–19 points): High risk
- Score IV (20–40 points): Probable dependence

Results

The osteonecrosis patient group includes a total of 28 patients, of which 11 are female and 17 are male. Of the 28 patients, 20 come from urban areas (U) and 8 from rural areas (R). The distribution of the Factor V Leiden mutation (G1691A) in the osteonecrosis group shows its presence in both heterozygous and homozygous forms, unequally distributed across age groups and by sex.

When analyzing the 28 patients for the prothrombin G20210A mutation, all patients aged 20–40 years (4 women and 4 men) were found to have no mutation. In the 41–60 age group, the mutation was identified in 2 women and 5 men (all heterozygous). In the over 60 age group, 2 women were found without the mutation.

The MTHFR mutations (C677T and A1298C) are associated with abnormal folate and homocysteine metabolism, contributing to hyperhomocysteinemia, a recognized risk factor for thrombosis and endothelial dysfunction, which can lead to disruption of bone microcirculation secondary to elevated homocysteine levels.

Analysis of the PAI-1 gene distribution (Plasminogen Activator Inhibitor-1, polymorphism 675 4G/5G) revealed a predominance of the 5G/5G genotype, generally considered protective, but

there was also a subset of patients carrying genotypes with increased procoagulant potential (4G/4G and 4G/5G), especially among the younger and middle-aged groups.

The distributions of Protein S, Protein C, Antithrombin III, and homocysteine levels were also analyzed.

Based on the coagulation test results, the distribution of patients by procoagulant status was as follows - among female patients, 7 had a severe procoagulant status, 3 had moderate status, and 1 had a normal status; among male patients, 7 had a severe procoagulant status, 5 had moderate status, and 5 had a normal procoagulant status; additionally, the study examined whether there was an association between behavioral risk factors (i.e., dependence risk) and procoagulant status, but the analysis did not reveal a clear link between the two.

Discussions

A total of 28 patients diagnosed with aseptic osteonecrosis were analyzed in the study, the majority of whom were male (17 men and 11 women). This suggests a higher susceptibility among men, possibly influenced by behavioral or occupational factors.

The most frequently affected age group was 41–60 years, with 16 patients, a segment considered vulnerable due to the accumulation of risk factors such as alcohol use, smoking, and corticosteroid therapy. Young adults aged 20–40 (8 patients) may present with forms that have a strong genetic component, while older adults (>60 years) were underrepresented, possibly due to underdiagnosis or limited access to investigations.

Most patients were from urban areas (20 out of 28), likely due to the hospital's location and greater access to advanced medical services. Women from rural areas were almost absent, while men had a more uniform distribution.

Genetic analysis revealed a high prevalence of the Factor V Leiden mutation in women (8 out of 11), including 3 homozygous cases, which is associated with increased thrombotic risk. In men, the mutation was rare and only found in heterozygous form. The G20210A prothrombin mutation was present exclusively in the 41–60 age group, only in heterozygous form, and was more frequent in men.

MTHFR mutations (C677T and A1298C) were identified only in women. C677T was also found in women over 60, while A1298C was found only in women under 60, suggesting different genetic vulnerabilities. In such cases, vitamin B complex supplementation may be justified.

The PAI-1 (4G/5G) polymorphism was evenly distributed between sexes, but the 4G/4G genotype (associated with increased risk) was commonly found in individuals aged 20–60 years. Its absence in the elderly may suggest compensatory mechanisms or natural selection.

Functional analysis of hemostatic parameters revealed variable levels of Antithrombin III and Proteins C and S, with greater fluctuations in men and predominantly functional deficiencies in women. Hyperhomocysteinemia, observed mainly in women, correlated with MTHFR mutations.

Chronic smoking did not significantly influence the levels of these biomarkers, and the severity of the prothrombotic status appears to be more strongly linked to genetic factors than behavioral ones.

The conclusion emphasizes the need for a personalized approach, based on age, sex, and genetic profile, in the evaluation and treatment of patients with aseptic osteonecrosis.

Conclusions

The multidimensional evaluation of the 28 patients with aseptic osteonecrosis reveals a complex profile in which genetic, age, and sex-related factors play a key role. The majority of patients were men aged 41–60, suggesting an association between male sex, active age, and increased disease risk, also supported by behaviors such as smoking and alcohol consumption.

The prevalence of the Factor V Leiden mutation in women, especially in homozygous forms, and the exclusive presence of MTHFR mutations (C677T, A1298C) in women indicate an elevated genetic risk for this group. The PAI-1 4G/5G polymorphism was common among patients of working age, with no significant sex differences observed.

Functional deficiencies in Antithrombin III and Protein S were predominant in the 41–60 age group, suggesting an active hypercoagulable state. Hyperhomocysteinemia, correlated with MTHFR mutations, was observed mainly in women, indicating the need for B-complex vitamin supplementation.

No significant correlations were found between smoking and functional markers, but an inverse relationship between Antithrombin III and Protein C was identified. Genetic screening and sex- and age-specific functional evaluation are recommended for a personalized approach to diagnosis and management.

Chapter 8. Study III – Quality of Life Assessment in Patients with Aseptic Osteonecrosis: A Cross-Sectional Observational Study

Introduction

Aseptic osteonecrosis has a profound impact on quality of life, affecting physical functionality, emotional balance, and social integration. Chronic pain and the progressive loss of mobility reduce patients' autonomy and confidence, negatively influencing their perception of their own health status.

This work proposes a research direction focused on the multidimensional assessment of quality of life, using an integrative perspective—functional, social, and emotional. The objectives include: measuring the overall quality of life score, analyzing the psychosocial impact, correlating outcomes with demographic variables, and identifying a predictive threshold score for personalized interventions and multidisciplinary support.

Material and Methods

To assess the quality of life in patients with aseptic osteonecrosis, a custom questionnaire was developed, integrating concepts from internationally validated tools such as WHOQOL-BREF, Oxford Hip Score (OHS), SF-36, and EQ-5D. The goal was to capture not only the functional status but also the psychological and social impact of the disease, with questions tailored to the clinical realities of Romania.

Elements from WHOQOL-BREF were used to formulate questions on social support, integration, and emotional well-being. For the functional component, the questionnaire was inspired by the OHS, including items on mobility and level of dependence. The global score structure reflects the methodology of SF-36, focusing on physical functioning, emotional balance, and perception of an active life. Additionally, principles from EQ-5D were incorporated for the quick evaluation of essential domains such as self-care, pain, and anxiety.

Data from 60 patients were compiled into an Excel database, including demographic variables and individual scores. Statistical analysis, performed with IBM SPSS Statistics, included means, standard deviations, comparative tests, Pearson correlations, and logistic regression models, providing a robust framework for hypothesis validation and clinical interpretation of the results.

Results

The results showed that patients who underwent surgical treatment had a lower average quality of life score, indicating a better overall condition. In contrast, those treated conservatively (with anti-inflammatory medication, injections, or rehabilitation) had higher average scores, suggesting more pronounced impairment in physical function and social integration.

This difference aligns with findings from the literature, which emphasize that surgical interventions, when correctly applied and at appropriate stages of the disease, facilitate faster recovery of joint function, pain relief, restored mobility, and socio-professional reintegration. The results support the study's hypothesis that treatment type is a significant predictive factor for quality of life in aseptic osteonecrosis.

This finding highlights a crucial aspect of managing aseptic osteonecrosis: although surgical treatment can effectively restore joint biomechanics, it does not automatically ensure a high postoperative quality of life. Approximately half of the operated patients continue to experience moderate dysfunction, and over one-third report severely impaired general condition.

The results indicate that the success of surgical intervention is influenced by individual factors such as:

- the stage of the disease at the time of surgery
- the presence of comorbidities (e.g., obesity, diabetes, cardiovascular diseases)
- the quality of postoperative psychosocial support
- access to functional rehabilitation programs (e.g., physical therapy)

A comparative analysis of quality of life scores between patients treated surgically and those treated conservatively revealed statistically significant and clinically relevant differences.

The psycho-emotional component significantly influences the subjective perception of quality of life. Patients with anxiety, mood disorders, or lack of family support often perceive their recovery as unsatisfactory, even when the surgical outcome is objectively favorable. This underscores the need for a multidisciplinary approach that combines surgery with psychological support, physical rehabilitation, and monitoring of lifestyle and environmental factors.

The analysis of quality of life scores shows clear differences between surgically and conservatively treated patients. Among those who underwent surgery 16.7% reported a very good quality of life, 47.2% reported a moderately affected quality of life and 36.1% reported a severely affected quality of life.

This may reflect late interventions, lack of postoperative support, or presence of comorbidities.

Among conservatively treated patients only 6.7% reported a very good quality of life, 53.3% reported a moderately affected quality of life and 40% reported it as severely affected This suggests limited effectiveness in advanced cases and persistent impairment in functional and psycho-social domains.

It was observed that men tend to report a "very good" quality of life, reflecting an optimistic perception of health and recovery. Conversely, women more often report a negative perception, focusing on mobility limitations and psychological and emotional impact, with their responses mainly falling into the "severely affected" category. In the "moderately affected" category, scores were evenly distributed between sexes, with no significant differences.

Another finding highlights a direct link between negative perception of quality of life and lack of family support, whether physical or emotional, emphasizing the essential role of family involvement in the recovery process of patients with aseptic osteonecrosis.

Perception of mobility is closely tied to the general perception of quality of life, a relationship well-documented in the literature. Studies show that patients facing significant difficulties in daily activities—such as climbing stairs or walking on flat ground—tend to have a negative outlook on their health status and social integration potential. Thus, the results indicate that functional dependency level is a key determinant of quality of life.

Additionally, the findings revealed a strong link between sleep quality and overall perception of quality of life, clearly showing that sleep disturbances negatively impact emotional well-being, physical rehabilitation capacity, social integration, and daily autonomy.

Discussions

The application of the Spearman and Kruskal-Wallis tests identified the main predictors of the total quality of life score. The response to Question 5 (dependence on assistance) highlighted the importance of functional autonomy (Spearman 0.918, $p<0.00001$). Questions related to the psycho-emotional dimension, specifically sleep quality (Question 6) and active lifestyle (Question 7), showed very strong correlations (0.922 and 0.827, respectively). Additionally, scores related to daily mobility (Question 4) and pain (Question 1) further supported the functional assessment. These factors are also supported by findings in the medical literature.

The results suggest that functional and psycho-emotional aspects can serve as relevant predictors of perceived quality of life and validate the usefulness of the questionnaire employed. Regardless of the type of treatment, the success of functional recovery in aseptic osteonecrosis depends on the psychological integration of the disease, family support, degree of autonomy, and maintenance of an active lifestyle.

These findings highlight the need for multidisciplinary protocols that include personalized psychological, social, and functional interventions.

Conclusions

The study shows that the main factors negatively affecting quality of life in patients with aseptic osteonecrosis are: functional dependence, sleep disturbances, mobility difficulties, and the perceived ability to lead an active life. Family support significantly influences emotional well-being and adaptation to the disease.

Although surgical treatment improves functionality, it does not guarantee a better perception of quality of life, emphasizing the importance of the subjective dimension. Therefore, a patient-centered approach is necessary—one that includes the subjective assessment of quality of life in medical decision-making.

Future directions aim to expand the questionnaire's use to larger samples and integrate it into digital platforms for ongoing monitoring. Prospective studies can examine the correlation between score evolution and the therapeutic interventions applied. This research proposes a modern evaluation model in which the patient's voice becomes an active part of the decision-making process, reflecting the necessary shift toward a more comprehensive and humanized medical practice.

Chapter 9. Conclusions and Personal Contributions

This thesis explores an emerging area of interest at the intersection of molecular biology and musculoskeletal pathology. Aseptic osteonecrosis is approached as a condition with multifactorial mechanisms, whose socioeconomic impact justifies a rethinking of treatment strategies beyond a purely surgical approach. The study proposes a multidisciplinary vision focused on prevention, personalized diagnosis, and quality-of-life assessment.

The research demonstrates that osteonecrosis is caused by a combination of genetic, functional, and behavioral factors. A consistent association was identified between procoagulant

status and the disease, regardless of age or sex. Factor V Leiden, G20210A, and the PAI-1 4G/4G genotype define a genetic profile with high predictive value, suggesting the need for targeted screening. Anticoagulant protein deficiencies and hyperhomocysteinemia complete the pathogenic profile, especially in patients aged 41–60 years.

Another novel element is the adaptation of a specific quality-of-life questionnaire, which highlighted the negative impact of functional dependence, sleep disturbances, and lack of family support on the perception of health status. While surgery improves functionality, it does not automatically ensure a better quality of life, emphasizing the need for an integrated, patient-centered evaluation.

The limitations of the study include the small sample size (28 patients), recruitment challenges, costs of genetic testing, the need for interdisciplinary collaboration, and the lack of a nationally validated tool for assessing quality of life. Confounding factors such as smoking and alcohol consumption, which are difficult to quantify, were also taken into account.

The author's contributions include designing a multimodal research framework, validating an adapted QoL questionnaire, identifying a high-risk genetic triad for thrombosis, proposing clinical-genetic screening algorithms, applying advanced statistical methods, and initiating a pioneering integrated research direction in Romania.

This work provides solid evidence in favor of a personalized and multidimensional approach to osteonecrosis - one that includes genetic analysis, functional risk factors, and the subjective dimension of quality of life. This model may serve as a foundation for developing modern clinical protocols, expanding research on a national scale, and digitally integrating patient monitoring. Thus, the thesis contributes to the advancement of precision medicine in musculoskeletal pathology.

BIBLIOGRAPHY

[1] Hart NH, Newton RU, Tan J, Rantalainen T, Chivers P, Siafarikas A, Nimphius S. Biological basis of bone strength: anatomy, physiology and measurement.

[2] Clarke B. Normal bone anatomy and physiology., 2008.

[3] Moreira CA, Dempster DW, Baron R. *Anatomy and Ultrastructure of Bone – Histogenesis, Growth and Remodeling*. (2000).

[4] Buck DW, Dumanian GA. Bone biology and physiology: Part I. the fundamentals, Lippincott Williams and Wilkins, 2012.

[5] Tomlinson RE, Silva MJ. Skeletal Blood Flow in Bone Repair and Maintenance. *Bone Res*, 1, 311–22 , 2013.

[6] Yoon BH, Mont MA, Koo KH, Chen CH, Cheng EY, Cui Q, Drescher W, Gangji V, Goodman SB, Ha YC, Hernigou P, Hungerford MW, Iorio R, Jo WL, Jones LC, Khanduja V, Kim HKW, Kim SY, Kim TY, Lee HY, Lee MS, Lee YK, Lee YJ, Nakamura J, Parvizi J, Sakai T, Sugano N, Takao M, Yamamoto T, Zhao DW. The 2019 Revised Version of Association Research Circulation Osseous Staging System of Osteonecrosis of the Femoral Head. *Journal of Arthroplasty*, 35, 933–40 , 2020.

[7] Shah KN, Racine J, Jones LC, Aaron RK. Pathophysiology and risk factors for osteonecrosis. *Curr Rev Musculoskelet Med*, 8, 201–9 , 2015.

[8] Hernigou P, Trousselier M, Roubineau F, Bouthors C, Chevallier N, Rouard H, Flouzat-Lachaniette CH. Stem cell therapy for the treatment of hip osteonecrosis: A 30-year review of progress. *CiOS Clinics in Orthopedic Surgery*, 8, 1–8 , 2016.

[9] Lespasio MJ, Sodhi N, Mont MA. Osteonecrosis of the Hip: A Primer. *Perm J*, 23, 18–100 , 2019.

[10] Horii T, Matsumoto T, Nishino M, Tomita K. Effects of steroids on femoral diaphyseal intramedullary circulation in rabbits. *Arch Orthop Trauma Surg*, 122, 506–9 , 2002.

[11] Pijnenburg L, Felten R, Javier RM. A review of avascular necrosis, of the hip and beyond. *Revue de Medecine Interne*, 41, 27–36 , 2020.

[12] Marker DR, Seyler TM, Ulrich SD, Srivastava S, Mont MA. Do modern techniques improve core decompression outcomes for hip osteonecrosis? *Clin Orthop Relat Res*, 466, 1093–103 , 2008.

[13] Albers A, Carli A, Routy B, Harvey EJ, Séguin C. Treatment with acetylsalicylic acid prevents short to mid-term radiographic progression of nontraumatic osteonecrosis of the femoral head: a pilot study. *Canadian Journal of Surgery*, 58, 198–205 , 2015.

[14] Glueck CJ, Freiberg RA, Sieve L, Wang P. Enoxaparin prevents progression of Stages I and II osteonecrosis of the hip. *Clin Orthop Relat Res*, 435, 164–70 , 2005.

[15] Wang CJ, Huang CC, Wang JW, Wong T, Yang YJ. Long-term results of extracorporeal shockwave therapy and core decompression in osteonecrosis of the femoral head with eight- to nine-year follow-up. *Biomed J*, 35, 481–5 , 2012.

[16] Habib A, Petrucci G, Rocca B. Pathophysiology of Thrombosis in Peripheral Artery Disease. *Curr Vasc Pharmacol*, 18, 204–14 , 2020.

[17] Montagnana M, Lippi G, Danese E. An overview of thrombophilia and associated laboratory testing. *Methods in Molecular Biology*, 1646, 113–35 , 2017.

[18] Jones LC, Mont MA, Le TB, Petri M, Hungerford DS, Wang P, Glueck CJ. Procoagulants and osteonecrosis. *Journal of Rheumatology*, 30, 783–91 , 2003.

[19] Naik AA, Sivaramakrishnan V. Femoral Head Osteonecrosis is associated with thrombosis, fatty acid and cholesterol biosynthesis: A potential role for anti-thrombotics and statins as disease modifying agents. *Med Hypotheses*, 161, 110808 , 2022.

[20] Cheng EY, Mirzaei A. Differential risk of autoimmune disorders in non-traumatic osteonecrosis: clue to pathogenesis. *Expert Rev Clin Immunol*, 21, , 2025.

[21] Kechli AM, Wilimas JA, Pui CH, Park VM, Tonkel S, Deitcher SR. Factor V Leiden and other hypercoagulable state mutations are not associated with osteonecrosis during or after treatment for pediatric malignancy. *Journal of Pediatrics*, 134, 310–4 , 1999.

[22] Glueck CJ, Freiberg RA, Boriel G, Khan Z, Brar A, Padda J, Wang P. The role of the factor v Leiden mutation in osteonecrosis of the hip. *Clinical and Applied Thrombosis/Hemostasis*, 19, 499–503 , 2013.

LIST OF PUBLISHED SCIENTIFIC PAPERS

1. **Lucian I. Stanciu**, Dana G. Minca, Adrian Cursaru, Catalin Cîrstoiu; Avascular Bone Necrosis in Systemic Lupus Erythematosus: An European Review Uncovering Hidden Risks, Early Diagnostic Gaps and Clinical Case Insight. *Romanian Journal of Military Medicine.* 128(4), 2025.
   DOI: https://doi.org/10.55453/rjmm.2025.128.4.7
   Capitolul 2, pag. 21-24
2. **Lucian I. Stanciu**, Dana G. Minca, Adrian Cursaru, Catalin Cîrstoiu;
   Unraveling Risk Determinants in Avascular Necrosis: A Cross-Sectional Analysis of Demographic, Behavioral, And Hemostatic Profiles. *Romanian Journal of Military Medicine.* 128(5), 2025.
   Capitolul 6, pag. 31-74
3. **Lucian I. Stanciu**, Dan C. Blendea, Doroteea Teoibaș-Șerban, Cătălin Cîrstoiu; Where Endocrinology Meets Orthopedics: A Comprehensive Review of Aseptic Bone Necrosis in Cushing's Syndrome. *Balneo and PRM Research Journal.* 16(2), 2025
   DOI: https://doi.org/10.12680/balneo.2025.823
   Capitolul 2, pag. 21-24
4. **Lucian I. Stanciu,** Dan C. Blendea, Adrian Cursaru, Bogdan Crețu, Doroteea Teoibaș-Șerban, Cătălin Cîrstoiu; Quality of Life in Patients Diagnosed with Avascular Osteonecrosis. *Balneo and PRM Research Journal.* 16(3), 2025
   Capitolul 8, pag 117-138