Investigative Study

ANORGANIC BOVINE BONE MATRIX TREATED WITH P15 EFFECTS IN OSTEOBLASTIC STEM CELL DIFFERENTIATION

P. Daliu[1], B. Zeza[2] and M. Martinelli[3]

[1]Dept Pharmacology, Albanian University, Tirana, Albania;
[2]Dental School, Albanian University, Tirana, Albania;
[3]Dept of Medical and Surgical Sciences, University of Bologna, Bologna, Italy

Correspondence to
Marcella Martinelli, PhD
Department of Medical and Surgical Sciences,
University of Bologna,
40138 Bologna, Italy
e-mail: marcella.martinelli@unibo.it

ABSTRACT

Molecular interaction between stem cells and grafting biomaterial is a key factor for the success of regenerative surgery. The covalent attachment of P15, a synthetic peptide mimicking the cell-binding site of collagen type I, to biomaterials could favor integrins binding and activation of multiple cell signaling pathways. In this investigation, we studied the effect of P15-coated anorganic bone matrix on bone marrow stem cells cultured *in vitro*. The expression level of a panel of differentiation markers was monitored to verify cell differentiation towards the osteoblast lineage. We observed some signs of osteoblast differentiation, such as the over-expression of osterix and under-expression of endoglin. However, most typical markers, osteonectin, osteopontin, osteocalcin, and alkaline phosphatase, were strongly under-expressed.

KEYWORDS: *p15, bone marrow stem cells, bone regeneration, gene expression*

INTRODUCTION

Bone replacement graft materials are used in orthopedic and oral surgery to encourage new bone formation. Experimental data demonstrated that different extracellular matrix proteins may improve cell attachment to synthetic and anorganic graft materials (1). Cells can bind extracellular matrix proteins with a family of transmembrane receptors named integrins. (2). Integrins can activate and modulate multiple signaling pathways. As a result, cell-substrate interaction may influence cell behavior throughout cell cycle regulation, directing cells to live or die, to proliferate, or to exit the cell cycle and differentiate (3). Specifically, integrins can play an important role in osteoblast differentiation as well as in bone remodeling (4).

Cells bind collagen type I, the main bone matrix protein, using the integrin α2β1 receptor. The specific binding site on type I collagen is the short aminoacidic sequence 766-GTPGPQGIAGQRGVV-780, (5, 6). A synthetic peptide mimicking the cell-binding site of collagen type I, named P15, has been frequently employed in attempting to enhance the regenerative potential of biomaterials. In osteoregenerative surgery, P15 has been conjugated with different materials to improve osteoblast and mesenchymal cell attachment and to promote cell activities essential for bone formation and regeneration (5, 7). The covalent attachment of P15 to titanium surfaces has been shown to improve cell adhesion, proliferation, and maturation, highlighting its efficacy in promoting tissue regeneration in orthopedic procedures (8).

A common source of bone allograft material is bovine or porcine bone. The bone is treated to obtain a scaffold for new bone growth that is essentially composed of its anorganic part, the hydroxyapatite, a crystalline form of calcium

Received: 12 August 2023
Accepted: 10 September 2023

phosphate. This porous material is commonly used in periodontal surgery, dental implantology, and maxillofacial surgery to fill bone defects, support dental implants, and reconstruct jaw bones. To improve the osteoconductivity of anorganic bone and promote the natural healing process, this material was coated with the peptide P15. Some investigations were aimed to compare P15 treated and untreated hydroxyapatite with alternate results (9).

Bone marrow stem cells (BMSCs) are multipotent stromal cells able to differentiate into various cell lineages, including osteoblasts, adipocytes, chondrocytes, and myocytes (10). They contribute together with periosteal cells to bone fracture healing and play a central role in orthopedic and oral regenerative applications (10, 11). BMSCs can be isolated from bone marrow aspirates obtained from the iliac crest or other long bones through minimally invasive procedures or from peripheral blood (12, 13). The BMSCs have been extensively studied and represent one of the main sources of cells for several regenerative medicine applications (14).

In this investigation, we studied the effect of P15-coated anorganic bone matrix on BMSCs cultured *in vitro*. The expression level of different differentiation markers was monitored to verify cell differentiation towards the osteoblast lineage.

MATERIALS AND METHODS

Isolation of Bone Marrow Stem Cells (BMSCs)

Bone marrow was extracted from the iliac crest of subjects (operated for grafting rehabilitation after trauma) and digested for 1 h at 37 °C in a solution containing 1 mg/ml collagenase type I and 1 mg/ml dispase, dissolved in phosphate-buffered saline supplemented with 100 U/ml penicillin, 100 µg/ml streptomycin, and 500 µg/ml clarithromycin.

The solution was then filtered using 70 µm Falcon strainers (Sigma-Aldrich, St Louis, MO, U.S.A.) to remove debris and large cell aggregates. Isolated cells were cultivated in α-MEM culture medium (Sigma-Aldrich, St Louis, MO, U.S.A.) supplemented with 20% Fetal Bovine Serum, 100 µM 2P-ascorbic acid, 2 mM L-glutamine, 100 U/ml penicillin, and 100 µg/ml streptomycin (Sigma-Aldrich, St Louis, MO, U.S.A). The flasks were incubated at 37 °C and 5% CO_2, and the medium was changed twice weekly. Adherent cells cultured in this medium were identified as BMSCs.

Immunofluorescence staining revealed the presence of mesenchymal stem cell markers, such as CD90 and CD73, while the hemopoietic marker CD34 was not detected.

Cell treatment

BMSCs were maintained in a humidified atmosphere containing 5% CO_2 at 37 °C. The cells were seeded at a concentration of 1.0×10^5 cells/ml with P15-coated anorganic bovine bone (Dentsply Friadent Ceramed, Lakewood CO) at the concentration of 10 mg/ml in 9 cm^2 (3 ml) wells containing DMEM supplemented with 10% serum and antibiotics.

The treatment was carried out at two time points: 24 h and 4 days. The treatments were triplicated in different wells. Another set of wells containing untreated cells was used as a control. At the end of the treatment period, the cells were lysed and processed for total RNA extraction.

RNA isolation, reverse transcription, and quantitative real-time PCR

Total RNA was isolated from the cells using RNeasy Mini Kit (Qiagen, Hilden, Germany) according to the manufacturer's instructions. The pure RNA was quantified using a NanoDrop 2000 spectrophotometer (Thermo Fisher Scientific, Wilmington, DE, USA).

cDNA synthesis was performed starting from 500 ng of total RNA using the PrimeScript RT Master Mix (Takara Bio Inc., Kusatsu, Japan). The reaction mixture was incubated at 37 °C for 15 min and inactivated by heating at 70 °C for 10 s. cDNA was amplified by real-time quantitative PCR using an ABI PRISM 7500 (Applied Biosystems, Foster City, CA, USA).

All PCR reactions were performed in a 20 µl volume. Each reaction contained 10 µl of 2X qPCRBIO SYGreen Mix Lo-ROX (PCR Biosystems, Ltd., London, UK), 400 nM of each primer, and cDNA.

Custom primers belonging to the "extracellular matrix, adhesion molecule" pathway, "osteoblast differentiation" and "inflammation" pathway were purchased from Sigma Aldrich. The selected genes grouped by functional pathways are listed in Table I.

All experiments were performed using non-template controls to exclude reagent contamination. PCR was performed using two analytical replicates.

The amplification profile was initiated by incubation for 10 min at 95 °C, followed by a two-step amplification for 15 s at 95 °C and 60 s at 60 °C for 40 cycles. In the final step, melt curve dissociation analysis was performed.

Table I. *Selected genes used in real-time PCR grouped by functional pathway.*

Pathway	Gene
Osteoblast differentiation	SPP1 (Osteopontin) SPARC (Osteonectin) RUNX2 (Runt-related transcription factor 2) ALP (Alkaline phosphatase) BGLAP (Osteocalcin) FOSL1 (FOS-like antigen 1) SP7 (Osterix) ENG (Endoglin)
Extracellular matrix, adhesion molecule	COL1A1 (Collagen type I alpha1) COL3A1 (Collagen type III alpha 1) MMP7 (Matrix Metallopeptidase 7) MMP12 (Matrix Metallopeptidase 12)
Inflammation	IL1A (Interleukin 1 alpha) IL1R1 (Interleukin 1 Receptor type 1)
Reference gene	RPL13 (Ribosomal protein L13)

Statistical analysis

Quantification was performed using the delta-delta Ct method. The gene expression levels were normalized to compare the expression of the reference gene RPL13 and expressed as fold-changes relative to the expression in untreated cells.

RESULTS

The BMSCs were phenotypically characterized using immunofluorescence. Fig. 1a shows cytoskeletal filaments stained with vimentin. The cell surfaces were positive for mesenchymal stem cell markers CD90 (Fig. 1b) and CD73 (Fig. 1c) and negative for markers of hematopoietic origin CD34 (Fig. 1d).

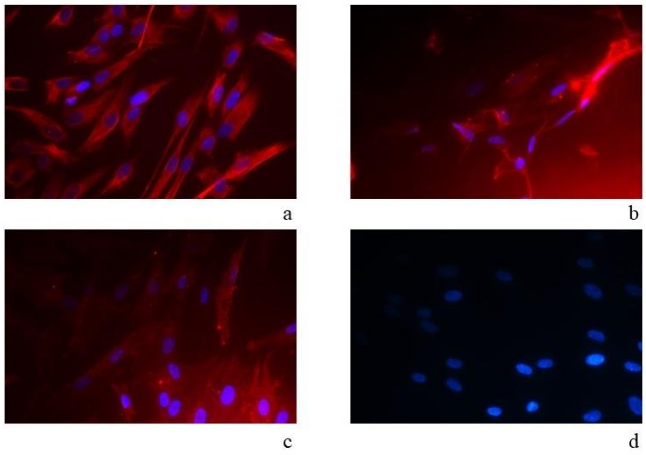


Fig. 1. *BMSCs by indirect immunofluorescence (Rhodamine). Immunofluorescence staining of vimentin (a), mesenchymal stem cell marker CD90 (b), CD73 (c), and hematopoietic markers CD34 (d). Nuclei were stained with DAPI. Original magnification x40.*

The effect of P15-coated anorganic bone matrix treatment in BMSCs was analyzed by quantitative real-time PCR after 24 h and 4 days of treatment, and the expression levels of osteoblast-related genes, extracellular matrix, and inflammation pathways were measured. Table II reports expression fold change obtained after 24 h and 4 days. An expression level change equal to or greater than 2 or equal to or less than 0.5 was considered biologically relevant.

An up-regulation of the IL1A gene was observed after 24 h of treatment. After 4 days, SP7 was the only osteogenic marker upregulated, while all the other appeared under-expressed.

Table II. *Gene expression in DPSCs after 24 h and 4 days of treatment. Numbers express the fold changes of the de-regulated genes in treated cells vs. untreated cells. ND – not determined. In bold significant gene expression level.*

	24 h	**4 days**
SPP1	ND	ND
SPARC	0.7	**0.3**
RUNX2	0.8	**0.4**
ALP	0.6	**0.1**
BGLAP	ND	ND
FOSL1	0.9	**0.4**
SP7	1.5	**7.3**
ENG	1.3	**0.3**
COL1A1	0.9	**0.4**
COL3A1	0.7	**0.4**
MMP7	1.5	1.7
MMP12	1.7	1.4
IL1A	**2.6**	1
IL1R1	1.2	**0.2**

DISCUSSION

The peptide P15 coating has emerged as a common practice to enhance the performance of biomaterials used in bone regeneration processes. P15, a synthetic peptide that mimics the integrins binding site on collagen type I, should favor cell adhesion to biomaterials, thus activating signaling pathways for cell proliferation and differentiation. In this investigation, we tested the ability of bovine bone to stimulate BMSCs through osteoblastic differentiation. The expression profiles of several differentiation markers were monitored at 24 hours and 4 days of treatment. At 24 hours, the expression profile resulted unchanged, apart from a 2.6-fold increase of the IL1A gene. This is not considered a sign of cell differentiation but may represent a response to cell stress or damage.

The IL1A gene codes for interleukin-1 alpha (IL-1α), a pro-inflammatory cytokine belonging to the interleukin-1 family, which plays a crucial role in the regulation of immune responses, inflammation, and tissue homeostasis (15). IL1A is constitutively expressed in many cell types in healthy tissues at a steady state, and its expression can be increased in response to growth factors and proinflammatory or stress-associated stimuli, such as canonical proinflammatory mediators that activate toll-like receptors (16).

IL-1α plays a pivotal role in the early stages of inflammation by promoting the recruitment of immune cells to the site of injury or infection and stimulating the production of other inflammatory mediators like IL6 and TNFα (17). Dysregulation of IL-1α is associated with various inflammatory diseases, including autoimmunity, cancer, and infectious diseases. It is one of the key cytokines involved in the pathogenesis of graft-*versus*-host disease, a major limiting factor in transplant success (18).

At four days of treatment, the IL1A expression appears normalized, while other differentiation markers appear dysregulated. Indeed, a marked over-expression of osterix, coded by the SP7 gene was observed.

Osterix is a transcription factor that plays a critical role in osteoblast differentiation and bone formation (19, 20). Indeed, osterix is essential for the activation of osteoblast-specific genes, such as osteonectin, osteopontin, osteocalcin, and alkaline phosphatase, which are fundamental components of bone extracellular matrix and its mineralization (21, 22). Identified as a key regulator of the genetic network controlling osteogenesis, SP7 functions downstream of RUNX2, another fundamental transcription factor in bone development (22, 23). Other findings suggest that osterix is regulated via both RUNX2-dependent and -independent mechanisms and that osterix controls osteoblast differentiation, at least in part, by regulating the expression of genes not controlled by RUNX2 (24). Given its role in bone formation, mutations in or dysregulation of SP7 are associated with various bone disorders. Common SP7 polymorphisms are associated with bone mineral density and fracture risk, while rare SP7 mutations cause skeletal dysplasia, and SP7 may contribute to bone metastasis (25). Osteogenesis imperfecta, a genetic disorder characterized by brittle bones, has been linked to mutations

in SP7 (26). Patients with these mutations exhibit symptoms such as frequent fractures, bone deformities, and growth deficiencies, reflecting impaired osteoblast function and bone matrix production (27).

Osteoporosis is a condition characterized by reduced bone mass and increased fracture risk. Dysregulation of SP7 expression or activity can disrupt the balance between bone formation and resorption, contributing to the development of osteoporosis (25). In addition to its role in osteoblasts, SP7 also influences the differentiation of chondrocytes, which are responsible for cartilage formation. Although primarily known for its osteogenic functions, emerging evidence suggests that SP7 may play a role in regulating the balance between osteogenesis and chondrogenesis, which is crucial for endochondral ossification, a process by which long bones are formed (28).

At 4 days after treatment with P15-hydroxyapatite, osterix appeared overexpressed, while its typical activator, RUNX2, or their downstream targets resulted in under-expressed. These data need to be confirmed and seem difficult to explain. Possibly, additional time points may help to explain this anomaly. On the other hand, we observed endoglin under-expression, a potential sign of osteoblast differentiation (29, 30). Indeed, endoglin is normally expressed in human mesenchymal stem cells, while its expression decreases as cells differentiate into an osteogenic lineage (31).

Osteonectin, osteopontin, osteocalcin, and alkaline phosphatase are all considered markers of osteoblast differentiation because they are abundantly synthesized by active osteoblasts and play essential roles in bone development, remodeling, and mineralization (32). Osteonectin is a matrix glycoprotein that interacts with hydroxyapatite, calcium, and type I collagen (33). Osteopontin plays a crucial role in bone mineralization and the attachment of osteoclasts to the mineral matrix. Osteopontin is involved in various physiological and pathological processes, including bone remodeling, immune response, and inflammation (34). Osteocalcin is specifically expressed in osteoblasts and is the most abundant non-collagenous protein in bone (35). Osteonectin is not involved in the regulation of bone formation and bone quantity, but osteonectin regulates bone quality by aligning biological apatite (BAp) parallel to the collagen fibrils (36). Some evidence indicates that osteocalcin functions as a hormone that regulates insulin secretion in the pancreas, testosterone synthesis in the testis, and muscle mass based; however, this is still a matter of debate (37).

Alkaline phosphatase (ALP) is an enzyme crucial for bone mineralization by dephosphorylating organic phosphate groups, essential for the formation of hydroxyapatite crystals in bone tissue (38, 39). Alkaline phosphatase is considered a marker of early osteoblast differentiation, even if it is also expressed in other tissues (40).

CONCLUSIONS

P15 is considered a promising tool for increasing the osteointegration of biomaterials used in bone regeneration surgery. BMSCs represent a valuable resource in regenerative medicine and tissue engineering, offering versatile applications in musculoskeletal regeneration, hematopoietic transplantation, and immunomodulatory therapies. BMSCs are a good model for studying P15 because they have a strong role in bone repair. In our *in vitro* experiments, we observed some elements of osteoblast differentiation, such as overexpression of SP7 and underexpression of ENG. However, most typical markers, osteonectin, osteopontin, osteocalcin, and alkaline phosphatase, were strongly underexpressed.

REFERENCES

1. Lallier TE, Yukna R, Moses RL. Extracellular matrix molecules improve periodontal ligament cell adhesion to anorganic bone matrix. *J Dent Res.* 2001;80(8):1748-1752. doi:https://doi.org/10.1177/00220345010800081301
2. Ruoslahti E, Pierschbacher MD. New perspectives in cell adhesion: RGD and integrins. *Science.* 1987;238(4826):491-497. doi:https://doi.org/10.1126/science.2821619
3. Giancotti FG, Ruoslahti E. Integrin signaling. *Science.* 1999;285(5430):1028-1032. doi:https://doi.org/10.1126/science.285.5430.1028
4. Globus RK, Moursi A, Zimmerman D, Lull J, Damsky C. Integrin-extracellular matrix interactions in connective tissue remodeling and osteoblast differentiation. *ASGSB Bull.* 1995;8(2):19-28.
5. Hanks T, Atkinson BL. Comparison of cell viability on anorganic bone matrix with or without P-15 cell binding peptide. *Biomaterials.* 2004;25(19):4831-4836. doi:https://doi.org/10.1016/j.biomaterials.2003.12.007
6. Bhatnagar RS, Qian JJ, Wedrychowska A, Sadeghi M, Wu YM, Smith N. Design of biomimetic habitats for tissue engineering with P-15, a synthetic peptide analogue of collagen. *Tissue Eng.* 1999;5(1):53-65. doi:https://doi.org/10.1089/ten.1999.5.53
7. Nguyen H, Qian JJ, Bhatnagar RS, Li S. Enhanced cell attachment and osteoblastic activity by P-15 peptide-coated matrix in hydrogels. *Biochem Biophys Res Commun.* 2003;311(1):179-186. doi:https://doi.org/10.1016/j.bbrc.2003.09.192
8. Liu Q, Limthongkul W, Sidhu G, et al. Covalent attachment of P15 peptide to titanium surfaces enhances cell attachment, spreading, and osteogenic gene expression. *J Orthop Res.* 2012;30(10):1626-1633. doi:https://doi.org/10.1002/jor.22116

9. Gillman CE, Jayasuriya AC. FDA-approved bone grafts and bone graft substitute devices in bone regeneration. Mater Sci Eng C Mater Biol Appl. 2021;130:112466. doi:10.1016/j.msec.2021.112466
10. Salgado AJ, Oliveira JM, Martins A, et al. Tissue engineering and regenerative medicine: past, present, and future. *Int Rev Neurobiol.* 2013;108(1-33. doi:https://doi.org/10.1016/B978-0-12-410499-0.00001-0
11. Han Y, Li X, Zhang Y, Han Y, Chang F, Ding J. Mesenchymal Stem Cells for Regenerative Medicine. *Cells.* 2019;8(8):doi:https://doi.org/10.3390/cells8080886
12. Zhao Y, Glesne D, Huberman E. A human peripheral blood monocyte-derived subset acts as pluripotent stem cells. *Proc Natl Acad Sci U S A.* 2003;100(5):2426-2431. doi:https://doi.org/10.1073/pnas.0536882100
13. Guo CH, Han LX, Wan MR, Deng GJ, Gan JH. Immunomodulatory effect of bone marrow mesenchymal stem cells on T lymphocytes in patients with decompensated liver cirrhosis. *Genet Mol Res.* 2015;14(2):7039-7046. doi:https://doi.org/10.4238/2015.June.26.13
14. Costela-Ruiz VJ, Melguizo-Rodriguez L, Bellotti C, et al. Different Sources of Mesenchymal Stem Cells for Tissue Regeneration: A Guide to Identifying the Most Favorable One in Orthopedics and Dentistry Applications. *Int J Mol Sci.* 2022;23(11):doi:https://doi.org/10.3390/ijms23116356
15. Malik A, Kanneganti TD. Function and regulation of IL-1alpha in inflammatory diseases and cancer. *Immunol Rev.* 2018;281(1):124-137. doi:https://doi.org/10.1111/imr.12615
16. Weber A, Wasiliew P, Kracht M. Interleukin-1 (IL-1) pathway. *Sci Signal.* 2010;3(105):cm1. doi:https://doi.org/10.1126/scisignal.3105cm1
17. Di Paolo NC, Shayakhmetov DM. Interleukin 1alpha and the inflammatory process. *Nat Immunol.* 2016;17(8):906-913. doi:https://doi.org/10.1038/ni.3503
18. Cullup H, Stark G. Interleukin-1 polymorphisms and graft-vs-host disease. *Leuk Lymphoma.* 2005;46(4):517-523. doi:https://doi.org/10.1080/10428190400029833
19. Hojo H, Ohba S. Sp7 Action in the Skeleton: Its Mode of Action, Functions, and Relevance to Skeletal Diseases. *Int J Mol Sci.* 2022;23(10):doi:https://doi.org/10.3390/ijms23105647
20. Nakashima K, Zhou X, Kunkel G, et al. The novel zinc finger-containing transcription factor osterix is required for osteoblast differentiation and bone formation. *Cell.* 2002;108(1):17-29. doi:https://doi.org/10.1016/s0092-8674(01)00622-5
21. He S, Choi YH, Choi JK, Yeo CY, Chun C, Lee KY. Protein kinase A regulates the osteogenic activity of Osterix. *J Cell Biochem.* 2014;115(10):1808-1815. doi:https://doi.org/10.1002/jcb.24851
22. Yang D, Okamura H, Nakashima Y, Haneji T. Histone demethylase Jmjd3 regulates osteoblast differentiation via transcription factors Runx2 and osterix. *J Biol Chem.* 2013;288(47):33530-33541. doi:https://doi.org/10.1074/jbc.M113.497040
23. Yang D, Okamura H, Teramachi J, Haneji T. Histone demethylase Utx regulates differentiation and mineralization in osteoblasts. *J Cell Biochem.* 2015;116(11):2628-2636. doi:https://doi.org/10.1002/jcb.25210
24. Matsubara T, Kida K, Yamaguchi A, et al. BMP2 regulates Osterix through Msx2 and Runx2 during osteoblast differentiation. *J Biol Chem.* 2008;283(43):29119-29125. doi:https://doi.org/10.1074/jbc.M801774200
25. Wang JS, Tokavanich N, Wein MN. SP7: from Bone Development to Skeletal Disease. *Curr Osteoporos Rep.* 2023;21(2):241-252. doi:https://doi.org/10.1007/s11914-023-00778-7
26. Hamon F, Wirotius JM, Nadeau G. [The nurse in a rehabilitation service and the handicapped person]. *Soins.* 1985;451):7-10.
27. Wang JS, Kamath T, Mazur CM, et al. Control of osteocyte dendrite formation by Sp7 and its target gene osteocrin. *Nat Commun.* 2021;12(1):6271. doi:https://doi.org/10.1038/s41467-021-26571-7
28. Komori T. Runx2, an inducer of osteoblast and chondrocyte differentiation. *Histochem Cell Biol.* 2018;149(4):313-323. doi:https://doi.org/10.1007/s00418-018-1640-6
29. Ishibashi O, Ikegame M, Takizawa F, et al. Endoglin is involved in BMP-2-induced osteogenic differentiation of periodontal ligament cells through a pathway independent of Smad-1/5/8 phosphorylation. *J Cell Physiol.* 2010;222(2):465-473. doi:https://doi.org/10.1002/jcp.21968
30. Song H, Rosano JM, Wang Y, et al. Continuous-flow sorting of stem cells and differentiation products based on dielectrophoresis. *Lab Chip.* 2015;15(5):1320-1328. doi:https://doi.org/10.1039/c4lc01253d
31. Cameron K, Travers P, Chander C, Buckland T, Campion C, Noble B. Directed osteogenic differentiation of human mesenchymal stem/precursor cells on silicate substituted calcium phosphate. *J Biomed Mater Res A.* 2013;101(1):13-22. doi:https://doi.org/10.1002/jbm.a.34261
32. Sodek J, Chen J, Nagata T, et al. Regulation of osteopontin expression in osteoblasts. *Ann N Y Acad Sci.* 1995;760(223-241. doi:https://doi.org/10.1111/j.1749-6632.1995.tb44633.x
33. Kelm RJ, Jr., Swords NA, Orfeo T, Mann KG. Osteonectin in matrix remodeling. A plasminogen-osteonectin-collagen complex. *J Biol Chem.* 1994;269(48):30147-30153.
34. Icer MA, Gezmen-Karadag M. The multiple functions and mechanisms of osteopontin. *Clin Biochem.* 2018;59(17-24. doi:https://doi.org/10.1016/j.clinbiochem.2018.07.003
35. Ducy P, Desbois C, Boyce B, et al. Increased bone formation in osteocalcin-deficient mice. *Nature.* 1996;382(6590):448-452. doi:https://doi.org/10.1038/382448a0

36. Moriishi T, Ozasa R, Ishimoto T, et al. Osteocalcin is necessary for the alignment of apatite crystallites, but not glucose metabolism, testosterone synthesis, or muscle mass. *PLoS Genet.* 2020;16(5):e1008586. doi:https://doi.org/10.1371/journal.pgen.1008586
37. Komori T. Functions of Osteocalcin in Bone, Pancreas, Testis, and Muscle. *Int J Mol Sci.* 2020;21(20):doi:https://doi.org/10.3390/ijms21207513
38. Stein GS, Lian JB. Molecular mechanisms mediating proliferation/differentiation interrelationships during progressive development of the osteoblast phenotype. *Endocr Rev.* 1993;14(4):424-442. doi:https://doi.org/10.1210/edrv-14-4-424
39. Orimo H. The mechanism of mineralization and the role of alkaline phosphatase in health and disease. *J Nippon Med Sch.* 2010;77(1):4-12. doi:https://doi.org/10.1272/jnms.77.4
40. Vimalraj S. Alkaline phosphatase: Structure, expression and its function in bone mineralization. *Gene.* 2020;754(144855. doi:https://doi.org/10.1016/j.gene.2020.144855

Comparative Study

PROPHYLACTIC USE OF ANTIBIOTIC-LOADED BONE CEMENT IN PRIMARY HIP REPLACEMENT: A SYSTEMATIC REVIEW OF THE LITERATURE AND META-ANALYSIS

G. Bettinelli[1], M. Alessio-Mazzola[2], C. Santacroce[3], A. Belluati[4], V. Salini[3] and G. Placella[3]

[1]Azienda Ospedaliera Carlo Poma, Orthopaedic and Trauma Unit, Mantua, MN, Italy;
[2]IRCCS Ospedale San Raffaele, Milan, Italy;
[3]Università Vita-Salute San Raffaele, Milan, Italy;
[4]Orthopaedic and Trauma Department, Hospital Santa Maria delle Croci, Ravenna, Italy

Correspondence to:
Mattia Alessio-Mazzola, MD
IRCCS Ospedale San Raffaele,
Via Olgettina 60,
20132, Milan, Italy
e-mail: mattia.alessio@hotmail.com

ABSTRACT

Periprosthetic joint infection (PJI) is a challenging complication following joint replacement and represents a significant health expense. Literature comparing antibiotic-loaded bone cement (ALBC) and plain bone cement (PBC) for total hip replacement is lacking and inconclusive. This research aims to meta-analyze the available literature in order to state if ALBC is superior to PBC for the prevention of PJI and, therefore, justify its widespread use in primary hip replacement. A systematic review of the literature was carried out about survival, in terms of septic revision, of cemented primary hip replacement, comparing ALBC and PBC following PRISMA guidelines. Articles published before 2005 have been excluded because cementing techniques have significantly improved over the last few years. The indication for surgery was both for hip osteoarthritis and for femoral neck fracture. National registry studies, cohort studies, and case series were included in this systematic review. Five articles were eligible for the meta-analysis, with 502.702 hip replacements. The forest plot comparing ALBC and PBC, with a CI of 99%, was in favor of the usage of antibiotics [χ^2=5.88, Risk Ratio=1.55 (1.29, 1.85)]. The sub-group analysis of the effect of ALBC when compared to PBC was not possible, based on total and partial replacement, nor the surgical indication. This meta-analysis, critically analyzing the available literature, proved the superiority and rationality of ALBC usage vs PBC. The present article proved a statistically significant reduction in PJI rate in primary hip replacements cementing with ALBC compared to PBC, but strong recommendations cannot be made. Further prospective randomized trials are requested to confirm the efficacy of ALBC in preventing PJI.

KEYWORDS: *antibiotic bone cement, hip, infection, revision, prosthesis*

Received: 3 September 2023
Accepted: 6 October 2023

INTRODUCTION

Periprosthetic joint infection (PJI) is a challenging complication following joint replacement and represents a significant health expense. Incidence of revision for PJI accounts for 5 to 20% of total revisions, and it has been estimated that less than 1% of total hip replacements will undergo revision surgery for an infection in the ten years after surgery (1-3).

Cemented hip arthroplasty is indicated in displaced intra-capsular femoral neck fracture or primary osteoarthritis (OA) in patients with poor bone quality, age 70 or greater, and diagnosis of osteoporosis or osteopenia (4). Bone cement during hip replacement can be loaded with antibiotics: since its introduction in the '70, antibiotic-loaded bone cement (ALBC) is routinely used and globally accepted for septic revision surgery, but there are still doubts and inhomogeneity about its use in primary hip replacement with different treatment trends depending on the country (5).

The cost-efficacy of ALBC in preventing periprosthetic infection is still debated, and scientific evidence is lacking and contradictory. Only a few articles systematically compare the use of ALBC and plain bone cement (PBC) for primary hip replacement with contrasting findings.

The aim of this study is to analyze the effects of ALBC in primary hip replacement, both performed for femoral neck fracture or osteoarthritis, in terms of implant failure. The primary endpoint was to evaluate the number of cemented hip prostheses revised for infection by comparing ALBC and PBC. The secondary endpoint was to assess and compare the number of revisions for any cause.

METHODS

Systematic research has been performed in Google Scholar, Cochrane Library e PubMed, about the efficacy of ALBC in comparison to PBC implants, in terms of primary hip implant survival. The study was conducted in conformity with the guidelines of the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) (6). The primary outcome was the comparison of revision rate due to PJI between cemented primary hip replacements with plain PBC and ALBC.

As a temporal cutoff, we included only articles published from 2005 on, since the cementing techniques significantly improved in recent past years (7). The keywords inserted in the research engines, properly matched using the Boolean operators AND or OR, were: "antibiotic-loaded bone cement", "primary hip prosthesis", "laden cement", "plain bone cement", "total hip arthroplasty", "periprosthetic", "infection", "cost analysis", "septic", and "revision". The research was screened for randomized controlled trials (RCT), retrospective analyses, national registry studies and retrospective case series.

The final inclusion criteria were the following:

1) RCT, systematic revisions, observational cohort studies, national prosthesis registries, retrospective case series;
2) articles dealing with primary total or partial cemented hip replacement, both performed for fracture or osteoarthritis;
3) articles comparing the usage of ALBC vs PBC in hip replacement;
4) date of publication later than 2005.

The exclusion criteria were the following:

1) studies published before 2005;
2) hip resurfacing;
3) revision surgery, not primary implants.

Whenever more than one article with data obtained from a different year but the same national prosthesis registries were available, the latest paper was included. The search results were independently assessed, filtered and selected by two authors (BG, PG) for eligibility. Every disagreement was resolved by a consensus meeting with a third author (MAM). The risk of bias was classified using the Methodological Index for Non-Randomized Studies (MINORS) (8). Each item of the MINORS was scored 0 when absent, 1 when present but inadequate, and 2 when present and adequate. The ideal score for comparative studies was 24, and 16 for non-controlled studies.

Comparative studies were classified as at high risk of bias if the overall score was ≤ 20, and at low risk of bias when > 20. Non-controlled studies were considered at high risk of bias when the overall score was ≤ 12 and at low risk of bias when > 12.

RESULTS

The research identified 1042 articles; deleting duplicates and non-inherent studies based on title and abstract, 714 papers remained, of whom only 97 were available for full-text assessment. Those have been screened in full-text: only 5 papers respected the inclusion criteria and two among them were retrospective analyses of the national United Kingdom prosthesis register. Only the newest published articles were included (Fig. 1).

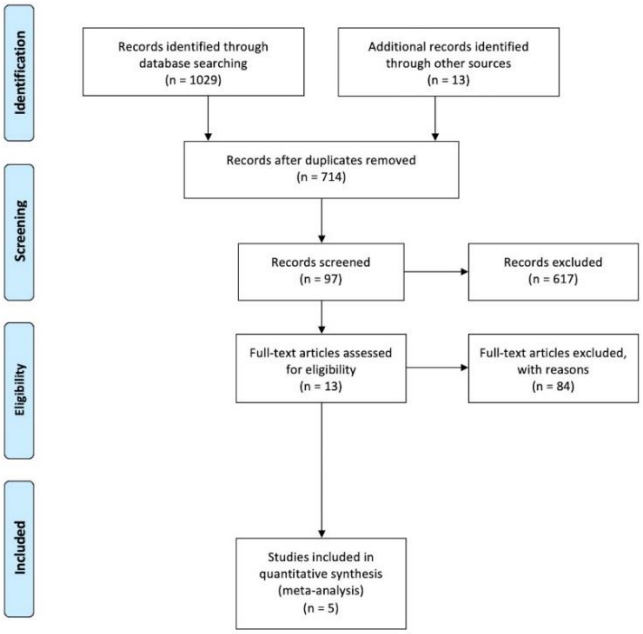


Fig. 1. *PRISMA flow diagram.*

Five papers have been included (9-13); all five were retrospective descriptive studies based on national prosthesis registries or case series; the publication date was between 2009 and 2020 (Table I, II).

Table I. *MINORS.*

	Clearly stated aim	Inclusion of consecutive patients	Prospective data collection	Endpoint appropriate to study aim	Unbiased assessment of study endpoint	Follow-up period appropriate to the study aim	<5% lost to follow-up	Prospective calculation of study size	Adequate control group	Contemporary groups	Baseline equivalence of	Adequate statistical analyses	total	Adequate number of patients	Risk of bias
HOSKINS	2	1	1	2	0	1	2	1	2	2	2	1	17/24	1	high
AEDO-MARTIN	2	1	1	2	0	2	0	1	2	2	2	2	17/24	1	high
SANZ-RUIZ	2	2	1	2	1	2	2	1	2	1	2	2	20/24	1	high
LEONG	2	1	1	2	0	2	0	1	1	2	0	2	14/24	2	low
DALE	2	1	1	2	0	2	0	1	1	2	0	2	14/24	2	low

Data have been analyzed with RevMan V.5.0.18.33, to create a forest plot. Possible publication bias has been assessed using a funnel plot.

Table II. *List of the studies included in the meta-analyses and their characteristics.*

	Year of publication	Study design	Nation	Study period	Diagnosis and implant type	PBC
Hoskins (9)	2020	retrospective case series	USA	2016-2018	total and partial hip replacement	50
Aedo-Martín (11)	2019	retrospective descriptive	Spain	2011-2017	partial hip replacement for fracture	147
Sanz-Ruiz (12)	2017	retrospective	Spain	2009-2012	total and partial hip replacement	262
Leong (10)	2020	retrospective from national prosthesis registry	UK	2005-2017	THA for OA	20961
Dale (13)	2009	retrospective from national prosthesis registry	Norway	1987-2007	THA	17991

The total THA collected was 502.702, 39.411 in the PBC group, and 463.291 in the ALBC group. Two studies included a population of elective total hip replacements (10, 13), one study (11) included hip replacements performed for femoral neck fracture, and other studies (9, 12) had a mixed population of elective and trauma cases.

It was impossible to provide the proportion of male and female patients because one of the studies (9) included hip and knee surgeries, not specifying the relative percentages for the two sub-groups of replacement.

Table III itemizes the number of cases of hip replacement for each article included, giving information about the type of cement used and the respective number of periprosthetic deep infections detected during the follow-up.

The primary outcome was to evaluate the number of events of "deep infection" encountered in the two categories, PBC vs ALBC.

Table III. *Cases of hip replacement for each article.*

	PBC	ALBC	PJI PBC	PJI ALBC	p-value	
Hoskins (9)	50	39	0	0	1.000	
Aedo-Martín (11)	147	94	28	8	**0.027**	*
Sanz-Ruiz (12)	262	257	22	6	**0.003**	*
Leong (10)	20961	397896	92	1288	**0.005**	*
Dale (13)	17991	65005	157	360	**<0.001**	*

***ALBC** = antibiotic-loaded bone cement, **PBC** = plain bone cement, **PJI** = periprosthetic joint infection. Asterisks highlight significant p-values (Fisher's exact test).*

Gathering the cases presented by these articles, a Forest plot assessed the odds ratio (CI 95%) of developing the event. Analyses demonstrated a significant prophylactic effect in preventing PJI with the usage of ALBC ($p<0.001$), $\chi^2=6.52$, $I^2=54\%$, OR 1.65 (1.27 to 2.14) (Fig. 2).

Precisely, the prevalence of PJI in patients who underwent THA with PCB was 0.8% versus 0.4% of patients treated with ALBC ($p<0.001$).

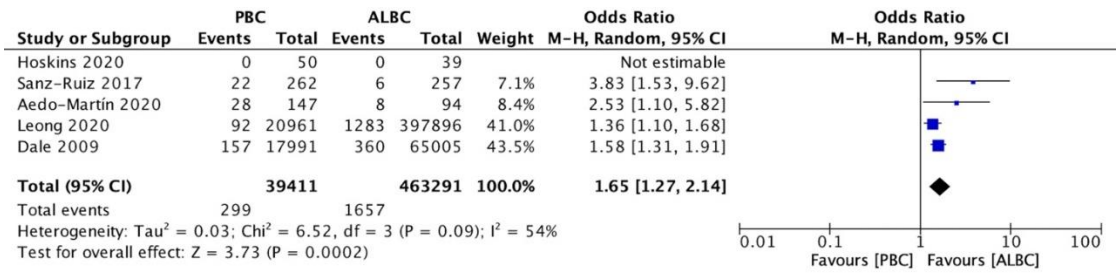


Fig. 2. *Forest plot.*

The Funnel plot (Fig. 3) referred to the included studies reveals a low data dispersion; a low risk of publication bias is expected.

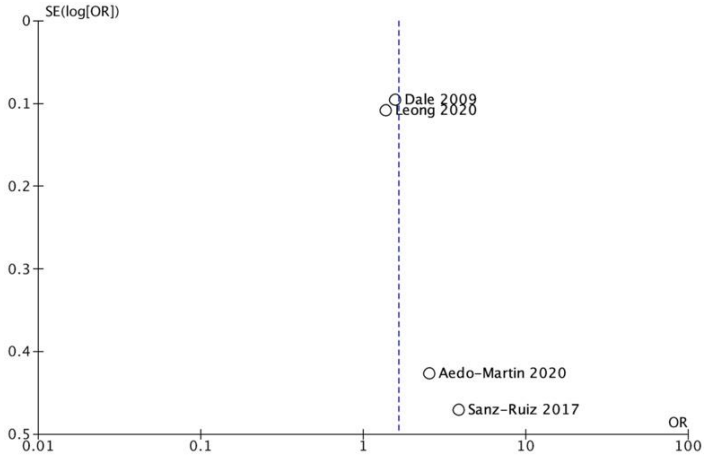


Fig. 3. *Funnel plot of the primary outcome.*

Compared to the analysis of Farhan-Alaine (3), this systematic review and meta-analysis provided updated data, replacing the article of Trela-Larsen (17) with the more recent analysis from the UK national register report performed by Leong et al. (10). However, further prospective randomized studies are necessary to provide a precise and high-level meta-analyze.

DISCUSSION

The main finding of this research is a statistically significant decrease in PJI rate in primary hip replacements when cementing with ALBC compared to PBC. However, considerable heterogeneity of reported data in the study leads to a careful interpretation of reported results.

The effectiveness of ALBC in PJI treatment is globally accepted. Its usage during septic revision received Federal Drug Administration (FDA) approval in 2003; on the other hand, its employment for primary joint arthroplasties is still debated (18). Literature about the benefit of ALBC usage in primary hip replacement is scarce and inconclusive, and no available guidelines provide strict recommendations.

Although there are only a few published studies in the literature with low quality, the number is even lower if only recent articles are selected. The difficulty in the design of a prospective study lies in the prolonged follow-up phase, the possibility of choosing different hip approaches, different cement brands, and other factors (i.e., operating room environment, operative field preparation method, and specific surgeon-related factors).

Regarding primary knee replacement, a meta-analysis published in 2022 (19) found no significant evidence to declare a decrease in peri-prosthetic infection rate using ALBC and authors recommended the routine use of PBC, to reduce surgeries cost.

On the contrary, other recent studies (20, 21) found a significant benefit from routine ALBC usage in total joint replacement, reducing the incidence of deep infection. This research concluded that the addition of antibiotics to cement is a safe and effective method for PJI prevention in primary total knee arthroplasties (TKA), although it is not effective in preventing superficial wound infections. The findings of this study can be justified by the difficulty encountered by the antibiotic mixed in the cement matrix to reach the superficial wound at a therapeutic concentration (20).

Similar conclusions in TKA and THA are given by Zhang et al. (21) as systemic administration of antibiotics had a role in preventing superficial surgical site infections, whereas the utility of ALBC is represented by the efficacy to prevent deep infections.

Data published in the literature support the findings of the present article: a previous meta-analysis by Parvizi (22), evaluated the use of ALBC in the prevention of PJI, however, the included articles have been published between 1987 and 1997 and several improvements in cement proprieties (6, 23) have been achieved over the years. Furthermore, this research showed a reduction in all causes of revision using antibiotic-soaked PMMA. These results potentially supported those who promote the routine use of ABLC, even in primary implants.

A systematic review and meta-analyses of RCT by Wang et al. (24) evaluated the prophylactic role of ALBC in primary THA and its effectiveness in preventing peri-prosthetic infection. The authors demonstrated a significant reduction of PJI without differences in superficial surgical site infections.

The present research study included a population of proximal femoral fractures and elective hip replacement that allowed to select only five studies. Furthermore, this systematic review and meta-analysis provided updated national United Kingdom registry data (10).

The rationale for the routine use of ALBC during primary hip replacement is still unconfirmed due to a lack of high-quality studies. Further prospective randomized trials are necessary to support the reduction of the incidence of PJI, decreasing the risk of re-intervention, with an advantage in terms of money-saving and patients' health.

Leong (10) found a protective role, in terms of revision for aseptic loosening, by using ALBC during THR and TKA. This result could highlight that some aseptic revisions may ultimately be caused by low-grade infections. Furthermore, these data supported that the mechanical properties of cement are not compromised by antibiotic addition at recommended doses (23).

The results found in the study by Farhan-Alaine et al. (3) showed a lower, but not significant, revision rate for any cause, in the group treated with ALBC.

To our knowledge, the present study is the most recent meta-analysis to compare the efficacy of ABLC and PBC for preventing PJI in primary hip replacement. Nevertheless, this study presents several limitations: the main limitation is the design of selected articles including only observational studies. Furthermore, due lack of high-level studies reporting comparative outcomes there is high heterogeneity in follow-up duration, type of cement used, type of antibiotic and implant design. Finally, the assessment of outcome "revision" could underestimate those patients with PJI treated with debridement, antibiotics and implant retention (DAIR), whenever not specified.

CONCLUSIONS

This meta-analysis critically analyzed the available literature and proved the superiority and rationality of ALBC usage vs PBC in primary THA.

The present article proves a statistically significant reduction in PJI rate in primary hip replacements cementing with ALBC compared to PBC, but strong recommendations cannot be made. Further prospective randomized trials are requested to confirm the efficacy of ALBC in preventing PJI.

Conflicts of interest statement

All authors certify that they have no affiliations with or involvement in any organization or entity with any financial interest or non-financial interest in the subject matter or materials discussed in this manuscript.

REFERENCES

1. Corbett KL, Losina E, Nti AA, Prokopetz JJ, Katz JN (2010) Population-based rates of revision of primary total hip arthroplasty: a systematic review. *PLoS One*. Oct 20;5(10):e13520. doi: 10.1371/journal.pone.0013520.
2. Nugent M, Young SW, Frampton CM, Hooper GJ (2021) The lifetime risk of revision following total hip arthroplasty. *Bone Joint J*. 103-B(3):479-485. doi: 10.1302/0301-620X.103B3.BJJ-2020-0562.R2.
3. Farhan-Alanie MM, Burnand HG, Whitehouse MR (2021) The effect of antibiotic-loaded bone cement on risk of revision following hip and knee arthroplasty. *Bone Joint J*. 103-B(1):7-15. doi: 10.1302/0301-620X.103B1.BJJ-2020-0391.R1.
4. Scanelli JA, Reiser GR, Sloboda JF, Moskal JT (2019) Cemented Femoral Component Use in Hip Arthroplasty. *J Am Acad Orthop Surg*. 15;27(4):119-127. doi: 10.5435/JAAOS-D-17-00245.
5. Yi Z, Bin S, Jing Y, Zongke Z, Pengde K, Fuxing P (2014) No decreased infection rate when using antibiotic-impregnated cement in primary total joint arthroplasty. *Orthopedics*. 37(12):839-45. doi: 10.3928/01477447-20141124-07.
6. Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffmann TC, Mulrow CD, et al. (2021) The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. *BMJ*. 372:n71
7. Magnan B, Bondi M, Maluta T, Samaila E, Schirru L, Dall'Oca C (2013) Acrylic bone cement: current concept review. *Musculoskelet Surg*. 97(2):93-100. doi: 10.1007/s12306-013-0293-9.
8. Slim K, Nini E, Forestier D, Kwiatkowski F, Panis Y, Chipponi J (2003) Methodological index for non-randomized studies (minors): development and validation of a new instrument. *ANZ J Surg* 73(9):712-6.

9. Hoskins T, Shah JK, Patel J, Mazzei C, Goyette D, Poletick E, Colella T 2nd, Wittig J (2020) The cost-effectiveness of antibiotic-loaded bone cement versus plain bone cement following total and partial knee and hip arthroplasty. *J Orthop*. 20:217-220. doi: 10.1016/j.jor.2020.01.029.
10. Leong JW, Cook MJ, O'Neill TW, Board TN (2020) Is the use of antibiotic-loaded bone cement associated with a lower risk of revision after primary total hip arthroplasty? *Bone Joint J*. Aug;102-B(8):997-1002. doi: 10.1302/0301-620X.102B8.BJJ-2020-0120.R1.
11. Aedo-Martín D, Crego-Vita D, García-Cañas R, Espigares-Correa A, Sánchez-Pérez C, Areta-Jiménez FJ (2020) Periprosthetic infection in elderly patients treated with hemiarthroplasty of the hip following intracapsular fracture. Should we use antibiotic-loaded bone cement? *Rev Esp Cir Ortop Traumatol (Engl Ed)*. Jan-Feb;64(1):28-34. English, Spanish. doi: 10.1016/j.recot.2019.07.005.
12. Sanz-Ruiz P, Matas-Diez JA, Sanchez-Somolinos M, Villanueva-Martinez M, Vaquero-Martín J (2017) Is the Commercial Antibiotic-Loaded Bone Cement Useful in Prophylaxis and Cost Saving After Knee and Hip Joint Arthroplasty? The Transatlantic Paradox. *J Arthroplasty*. Apr;32(4):1095-1099. doi: 10.1016/j.arth.2016.11.012.
13. Dale H, Hallan G, Hallan G, Espehaug B, Havelin LI, Engesaeter LB (2009) Increasing risk of revision due to deep infection after hip arthroplasty. Acta Orthop. Dec;80(6):639-45. doi: 10.3109/17453670903506658.
14. Australian Orthopaedic Association (2009) National joint replacement registry Committee hip and knee arthroplasty annual report 2009 Adelaide AOA.
15. Colas S, Collin C, Piriou P, Zureik M (2015) Association Between Total Hip Replacement Characteristics and 3-Year Prosthetic Survivorship: A Population-Based Study. *JAMA Surg*. Oct;150(10):979-88. doi: 10.1001/jamasurg.2015.1325.
16. Engesaeter LB, Espehaug B, Lie SA, Furnes O, Havelin LI (2006) Does cement increase the risk of infection in primary total hip arthroplasty? Revision rates in 56,275 cemented and uncemented primary THAs followed for 0-16 years in the Norwegian Arthroplasty Register. *Acta Orthop*. Jun;77(3):351-8. doi: 10.1080/17453670610046253.
17. Trela-Larsen L, Sayers A, Blom AW, Webb JCJ, Whitehouse MR (2018) The association between cement type and the subsequent risk of revision surgery in primary total hip replacement. *Acta Orthop*. Feb;89(1):40-46. doi: 10.1080/17453674.2017.1393224.
18. Martínez-Moreno J, Merino V, Nácher A, Rodrigo JL, Climente M, Merino-Sanjuán M (2017) Antibiotic-loaded Bone Cement as Prophylaxis in Total Joint Replacement. *Orthop Surg*. Nov;9(4):331-341. doi: 10.1111/os.12351.
19. Li HQ, Li PC, Wei XC, Shi JJ (2022) Effectiveness of antibiotics loaded bone cement in primary total knee arthroplasty: A systematic review and meta-analysis. *Orthop Traumatol Surg Res*. Sep;108(5):103295. doi: 10.1016/j.otsr.2022.103295.
20. Xu T, Wu KL, Jie K (2022) Comprehensive meta-analysis of antibiotic-impregnated bone cement versus plain bone cement in primary total knee arthroplasty for preventing periprosthetic joint infection. *Chin J Traumatol*. Nov;25(6):325-330. doi: 10.1016/j.cjtee.2022.06.001.
21. Zhang J, Zhang XY, Jiang FL, Wu YP, Yang BB, Liu ZY, Liu D (2019) Antibiotic-impregnated bone cement for preventing infection in patients receiving primary total hip and knee arthroplasty: A meta-analysis. *Medicine* (Baltimore). Dec;98(49):e18068. doi: 10.1097/MD.0000000000018068.
22. Parvizi J, Saleh KJ, Ragland PS, Pour AE, Mont MA (2008) Efficacy of antibiotic-impregnated cement in total hip replacement. *Acta Orthop*. Jun;79(3):335-41. doi: 10.1080/17453670710015229.
23. Wall V, Nguyen TH, Nguyen N, Tran PA (2021) Controlling Antibiotic Release from Polymethylmethacrylate Bone Cement. *Biomedicines*. Jan 1;9(1):26. doi: 10.3390/biomedicines9010026.
24. Wang J, Zhu C, Cheng T, Peng X, Zhang W, Qin H, Zhang X (2013) A systematic review and meta-analysis of antibiotic-impregnated bone cement use in primary total hip or knee arthroplasty. *PLoS One*. 12;8(12):e82745. doi: 10.1371/journal.pone.0082745.

Retrospective Study

RETROSPECTIVE CLINICAL AND RADIOGRAPHIC STUDY ON HETEROTOPIC OSSIFICATIONS AND HIP PROSTHESES FOR PRIMARY AND SECONDARY COXARTHROSIS: OUR EXPERIENCE

E.J. Samie[1], E. Andreoli[1], F. Belfiglio[1], S.R. Tari[2], A. Pantalone[3] and D. Bruni[3]

[1]Clinic of Orthopaedics and Traumatology, "SS. Annunziata" Hospital, Chieti, Italy;
[2]Department of Innovative Technologies in Medicine & Dentistry, University of Chieti-Pescara, Italy;
[3]Department of Medicine and Science of Aging, Clinic of Orthopaedics and Traumatology, University G. d 'Annunzio, Chieti-Pescara, Chieti, Italy

**Correspondence to:*
Danilo Bruni, MD
Department of Medicine and Science of Aging,
Clinic of Orthopaedics and Traumatology,
University G. d 'Annunzio Chieti-Pescara,
Chieti, Italy
e-mail: danilo.bruni@unich.it

ABSTRACT

Possible complications of hip prosthesis implants include the appearance of heterotopic ossifications (HO), i.e., the neoformation of bone tissue in the periprosthetic areas: it occurs in the months following the intervention due to a structural transformation of parts of the gluteal muscle. Materials and Methods: In the period between October 2014 and November 2023, at the same hospital facility by the same orthopedic team, 715 hip arthroplasty implantation surgeries were performed, of which 366 patients with coxarthrosis in the absence of recent fractures of the proximal epiphysis of the femur. Results The average age at the date of hip arthroplasty implantation surgery is 70.9 years. Radiographic checks performed after at least 3 months were taken into account to study the occurrence of heterotopic ossifications. In conclusion, in the event of the onset of HO, bloodless, physiotherapeutic, medical, and possibly radiant, timely, and close follow-up treatments are necessary to avoid the aggravation of local conditions.

KEYWORDS: *hip prosthesis implants, heterotopic ossifications, coxarthrosis*

INTRODUCTION

After the insertion of a hip prosthesis, the radiographic finding of small asymptomatic ossifications is common; it can rarely cause severe functional limitation up to complete blockage of the hip (1). The origin of this complication is almost unknown; individual constitutional factors are often called into question, although it is known that it occurs statistically more frequently in interventions following traumatic events.

Many authors have studied the possible prophylaxis of this complication, and the most effective practical suggestions have resulted:

- selection and care of the surgical technique;
- pharmacological prophylaxis (1);
- prophylaxis with radiation therapy (2);

Received: 24 September 2023
Accepted: 26 October 2023

Heterotopic calcifications tend to reduce joint motility with varying severity depending on the location and extent of ossifications. In sporadic cases it may be necessary to remove such ossifications in order to improve joint movement.

Risk factors

The precise pathogenesis is still debated. Some risk factors are recognized, some are preventable, and others are not, and can be schematically divided into two categories: patient-related risks and risks related to the surgical technique (3-6).

Patient-related risk factors are:

- over 65 years of age;
- male sex;
- high BMI;
- previous brain or spinal trauma;
- presence in other ossification sites;
- systemic disorders such as spondylitis, hypertrophic osteoarthritis, diffuse idiopathic skeletal hyperostosis, Paget's disease, post-traumatic osteoarthrosis, osteonecrosis, rheumatoid arthritis;
- previous hip surgery.

Surgery related to risk factors are:

- invasiveness of the technique on muscle tissues;
- persistence of bone debris;
- hematoma;
- detachment of the gluteal muscles;
- cementation of the plant;
- release of the psoas tendon;
- trochanteric or femoral osteotomy;
- procedure duration exceeding 90 minutes;
- prolonged drainage beyond 24 hours.

Epidemiology

It is one of the most frequently encountered complications after first intervention prosthesis, reported variably from 5 to 87%, with an average incidence of 30%. However, clinically significant forms have a much lower incidence, from 0.5 to 12%, as determined by countless studies in the literature (7). The prostheses most prone to ossifications are those that require the use of cement to attach to the bone; today, they are used very little in young patients and are reserved for the elderly (8).

Pathophysiology

Chalmers et al. (9) have proposed 3 necessary conditions for the onset and formation of heterotopic calcifications:

- osteogenic precursor cells;
- inducing agents;
- triggering event.

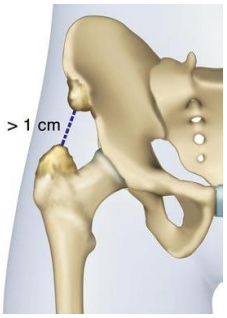
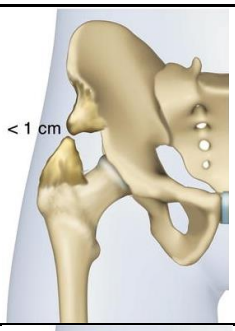
In fact, it has been seen that the main cause of heterotopic ossifications consists in an inappropriate differentiation of pluripotent mesenchymal cells into osteoblastic stem cells; however, the definitive trigger remains unclear, although Zaccalini highlighted that the demineralization of the bone matrix induces the formation of a morphogenetic protein (BMP) that could transfer from the traumatized bone to the surrounding tissues and thus stimulate the transformation of perivascular mesenchymal cells into osteoblasts (10). It has recently been proposed that prostaglandin E can also mediate the differentiation of progenitor cells.

A relevant figure is, finally, the observation of Tonna and e Cronckite (11), who noted the osteoblastic differentiation of primitive stem cells in the first 16 hours following an experimentally induced trauma on cat femurs with a peak of activity observed at 32 hours. However, although the causal factor remains unknown, there is no doubt that the formation of heterotopic calcifications depends on factors that, acting locally or systemically, influence the balance between osteogenesis and osteo-inhibition.

Classification

The Brooker Classification divides the extent of the formation of heterotopic calcifications after total arthroplasty into five grades: Grade 0, which is the absence of periprosthetic calcifications; Grade 1 is described as bone islands within soft tissues around the hip; Grade 2 includes bone spurs that originate from the pelvis or proximal end of the femur, leaving at least 1 cm between opposite bone surfaces; Grade 3 consists of bone spurs that originate from the pelvis or proximal end of the femur, reducing the space between opposite bone surfaces to less than 1 cm; Grade 4 shows apparent bone ankylosis of the hip (Table I) (12, 13).

Table I. *Brooker's radiographic Classification of the hip's heterotopic calcifications.*

Grade 0	Absence of calcification and ossification	
Grade 1	Bone islands in soft tissues around the hip	
Grade 2	Bone spurs starting from the pelvis and/or proximal end of the femur leaving the space of at least 1 cm between the opposite bone surfaces.	
Grade 3	Bone spurs starting from the pelvis or proximal extreme of the femur reduce the space between the opposite bone surfaces to less than 1 cm.	
Grade 4	Evident bone ankylosis of the hip	

Treatment

Asymptomatic patients have a quality of life comparable to patients without ossifications and do not need special attention. For patients with symptoms, especially those with severe movement deficit (degrees 3/4 sec. Brooker), ossification removal surgery is indicated (14).

Surgical indications for the excision of heterotopic ossification include:

- the improvement of the function;
- upright posture, sitting or walking;
- independence in dressing;
- the power supply;
- hygiene and repeated pressure sores from the underlying bone mass.

This surgery should usually be performed 12-18 months from the first to avoid the error of removing an ossification not yet fully formed, greatly increasing the risk of recurrence. For this reason, these patients are often asked to perform a bone scan. This examination gives us a general indication of the evolution of the pathology and its metabolic activity. The ideal candidate for surgical treatment before 18 months should not have joint pain or swelling, a normal level of alkaline phosphatase, and a 3-step bone scan indicating mature heterotopic ossification (15).

MATERIALS AND METHODS

In the period between October 2014 and November 2023, at the same hospital facility by the same orthopedic team, 715 hip arthroplasty implantation surgeries were performed, of which 366, in patients with coxarthrosis in the absence of recent fractures of the proximal epiphysis of the femur. The most widely used access route was the direct lateral route (Hardinge Bauer), carried out in 335 cases (92%) (Table II).

Table II. *Report of hip arthroplasty interventions on coxarthrosis and in the absence of recent fractures of the proximal epiphysis of the femur carried out in the period between October 1, 2014 and November 30, 2023 in our center.*

YEAR	N. HIP ARTHROPROSTHESIS IMPLANT SURGERY ON COXARTHROSIS	LATERAL ACCESS ROUTE (HARDINGE BAUER)	FRONT ACCESS ROUTE (SMITH - PETERSEN)	FRONT - LATERAL ACCESS ROUTE (WATSON - JONES)	POSTERO - LATERAL ACCESS ROUTE (GIBSON - MOORE)
2014	13	13	-	-	-
2015	41	41	-	-	-
2016	75	72	3	-	-
2017	70	55	15	-	-
2018	69	68	1	-	-
2019	28	28	-	-	-
2020	13	10	-	3	-
2021	17	15	-	1	1
2022	21	17	-	4	-
2023	19	16	1	2	-
TOTALE	366	335	20	10	1

The anterior access route to the hip (Smith - Petersen) was preferred in 20 interventions (5%). The anterior-lateral access route to the hip (Watson-Jones) was chosen in 10 interventions (3%). Only in 1 case has the posterior-lateral (Gibson-Moore) access route (Table III, Fig. 1).

Table III. *Stratification of the severity of heterotopic calcifications that appeared during the follow-up of hip arthroplasty implatation interventions on coxartrosis using the lateral access route.*

Brooker Classification	Number of cases	Number of cases (%)
,Grade 0	76	43.4%
Grade 1	48	27.4%
Grade 2	31	17.7%
Grade 3	13	7.4%
Grade 4	7	4%

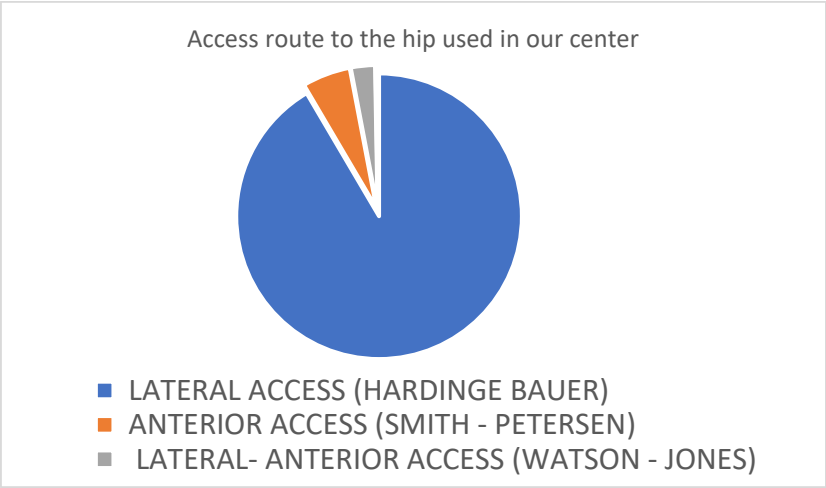


Fig. 1. *Access route to the chosen hip in hip arthroplasty implantation operations on coxarthrosis.*

The average age of hip arthroplasty implantation surgery is 70.9 years. Radiographic checks performed after at least 3 months were taken into account to study the occurrence of heterotopic ossifications. Patients who were unable to follow up for at least 3 months were therefore excluded from the study.

Initially the design provided for a follow-up of at least 6 months, but looking at the radiographic data shows an early appearance of the complication of ossifying myositis, which can become aggravating over time but, from our experience, there is no late onset (late-onset), in case of absence of early appearance of calcifications (first trimester post-intervention). This has therefore made it possible to enlist a greater number of patients.

The number of cases included in the study is therefore 196, of which 175 operated by preferring the direct lateral route, 14 the anterior route and 7 the anterolateral route. The heterotopic calcifications were classified according to the classification proposed by Brooker, which, as already mentioned, stratifies the severity of heterotopic ossifications into 4 classes.

According to this classification, the heterotopic calcifications that appeared at the periprosthetic site were then stratified within subgroups of patients operated with the direct lateral, anterior and anterolateral access routes.

RESULTS

Based on Brooker's classification:

- the heterotopic calcifications that appeared in cases of arthroprostheses implanted through direct lateral access were stratified into the following (Fig. 2):

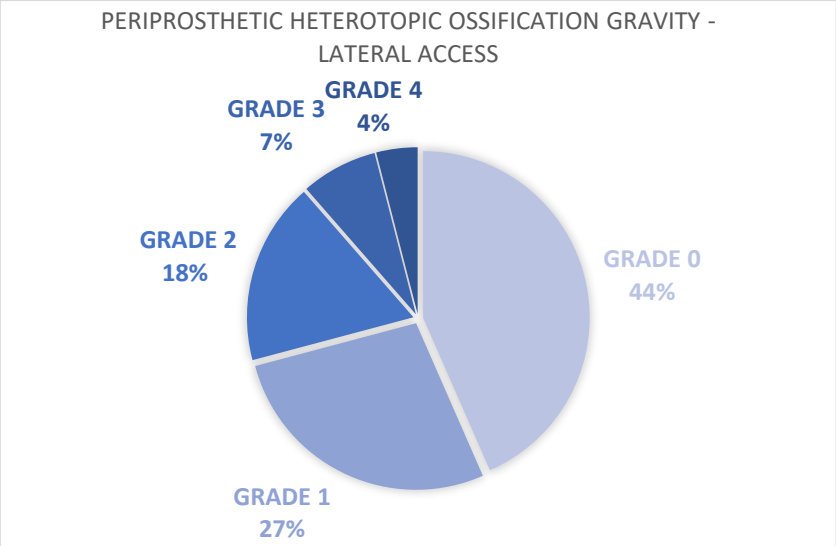


Fig. 2. *Stratification of the severity of heterotopic calcifications that appeared during the follow-up of hip arthroplasty implantation interventions on coxarthrosis using the lateral access route.*

- the heterotopic calcifications that occurred in the case of arthroproseses implanted through the anterior access were stratified into the following (Table V, Fig. 3):

Table V. *Stratification of the severity of heterotopic calcifications that appeared during the follow-up of hip arthroplasty implantation interventions on coxarthrosis using the anterior access route.*

Brooker Classification	Number of cases	Number of cases (%)
Grade 0	10	71.4%
Grade 1	2	14.3%
Grade 2	1	7.1%
Grade 3	1	7.1%
Grade 4	-	0%

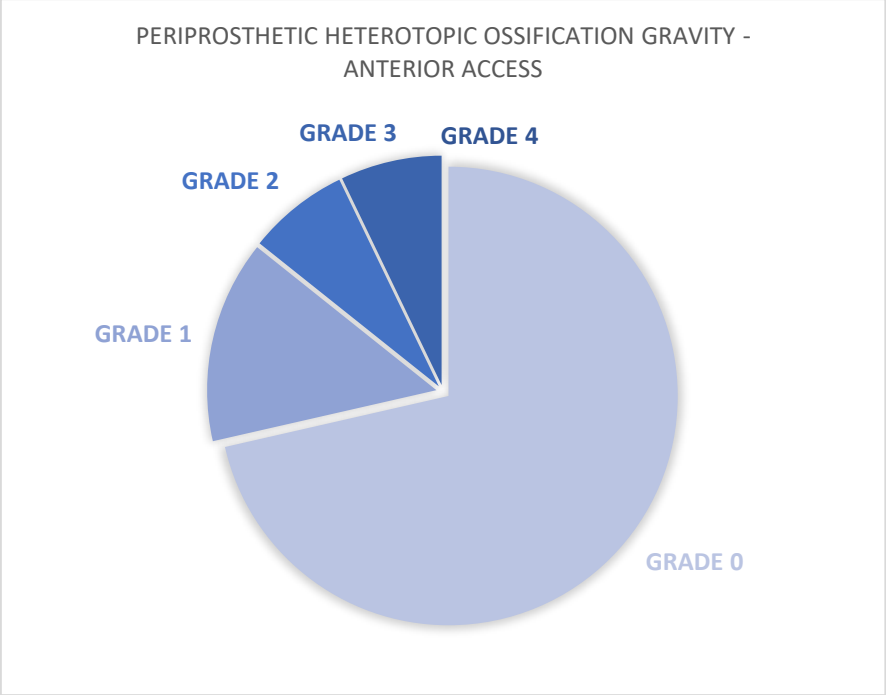


Fig. 3. *Stratification of the severity of heterotopic calcifications that appeared during the follow-up of hip arthroplasty implantation interventions on coxarthrosis using the anterior access route.*

- the heterotopic calcifications that appeared in the case of arthroprostheses implanted through the anterolateral access were stratified into the following (Table VI, Fig. 4):

Table VI. *Stratification of the severity of heterotopic calcifications that appeared during the follow-up of hip arthroplasty implantation interventions on coxarthrosis using the anterolateral access route.*

Brooker Classification	Number of cases	Number of cases (%)
Grade 0	5	71.4%
Grade 1	1	14.3%
Grade 2	1	14.3%
Grade 3	-	0%
Grade 4	-	0%

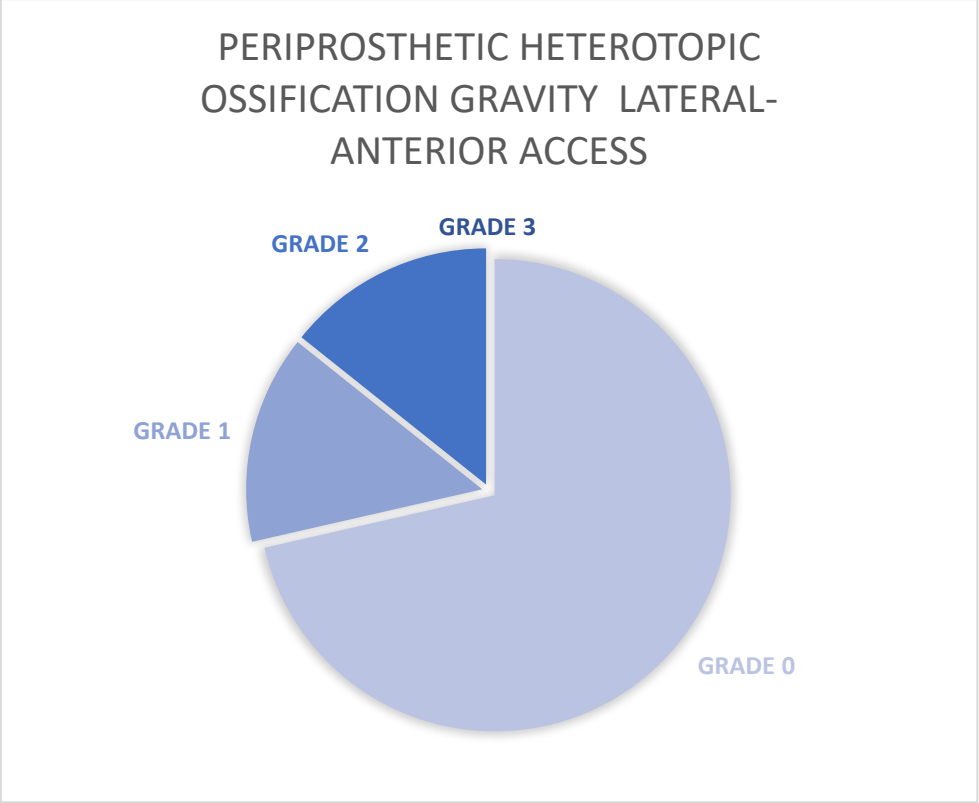


Fig. 4. *Stratification of the severity of heterotopic calcifications that appeared during the follow-up of hip arthroplasty implantation interventions on coxarthrosis using the anterolateral access route*

Considering the significant disproportion in absolute numbers regarding the use in our experience of the lateral access route compared to the others, the data that emerged were then reworked in percentage terms, demonstrating that the lateral access route direct to the hip does not increase the risk of developing symptomatic and severe periprosthetic heterotopic calcifications (Fig. 5).

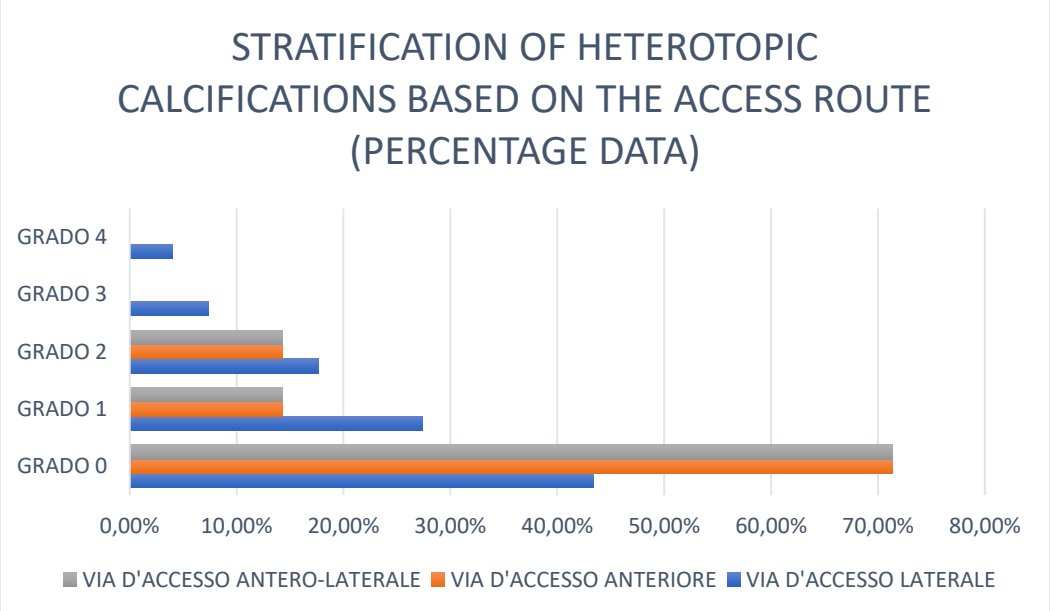


Fig. 5. *Stratification of heterotopic calcifications by access route (percentage data).*

In fact, these data show that, although the lateral access route direct to the hip is preferred in absolute terms, when the data are reported as a percentage, the three access routes studied do not show substantial differences in the appearance

of severe ossifying myositis. Using the reworked data in percentage terms, there is therefore no statistically detectable superiority in the choice of the access route to reduce the severity of the periprosthetic heterotopic calcifications that have appeared.

DISCUSSION

The data that emerged from this retrospective study, not free of bias and performed on a limited sample in accordance with what is reported in the literature for the postero-lateral route (16, 17), show that the choice of the lateral access route directed to the hip, preferred in our center, does not expose patients to a greater risk of developing severe and therefore symptomatic heterotopic ossifications. From the literature review, the same conclusion was reached for the comparison between the AMIS (Anterior Mini Invasive Surgery) approach and the direct lateral route: AMIS has been associated with better clinical and functional outcomes, but with regard to HO, the superiority of the AMIS approach in terms of incidence has not been demonstrated (18).

Contrary to what one might therefore think, it is not the extension of the access route itself that determines a risk factor for the onset of ossifications, nor, in our opinion, is the unique search for an intermuscular-only approach. Still, it is the muscular trauma, the tearing of tissues, the bleeding and the subsequent formation of periarticular haematomas, and the use of intraarticular drainage, especially in predisposing contexts (19).

The review by Cohn and colleagues on the appearance of HO after the implantation of PTA (20) reports the great variability of theses (sometimes in opposition to each other) relating to how the development of HO can be influenced by the surgical approach used in PTA. In a study of 507 consecutive patients with osteoarthritis or avascular necrosis, Morrey et al. (21) found that the incidence of severe HO was lower with a posterior approach (22%) than with an anterolateral (29%) or transtrocanteric approach (28%).

In a study of 1420 consecutive PTAs using the direct lateral approach, an overall incidence of HO of 27% was found, with HO occurring around the large trochanter in 15% of the hips. These results were confirmed by Eggli and Woo (5), who found that the incidence of HO was greater than 8.1% with an anterior or anterolateral approach compared to a posterior approach and that the incidence was greater than 15.1% when a trochanteric osteotomy was performed. Retrospectively, various lateral approaches were compared and found to have a 5x greater risk of HO with the Liverpool method than the Hardinge or transtrocanteric approach. Other authors (22) found no effect of the surgical approach on HO.

CLINICAL CASES

There are two clinical cases in which the appearance of periprosthetic heterotopic calcifications was rapid and severe (grades 4 and 3, respectively). In the first case, it was necessary to intervene surgically about 14 months after the implant by performing an arthrolysis (exal of the femoral-acetabular bone bridge and joint mobilization); in the second case, however, an adequate and regular physiotherapy treatment, at the same time as the pharmacological treatment, led to good results (see clinical images at six months) (Fig. 6-11).

Clinical case A

76 years old - male - PTA system sx - Direct lateral access route performed.

Fig. 6. *Preoperative Rx: III-degree sec. Kellgren-Lowrance classification.*

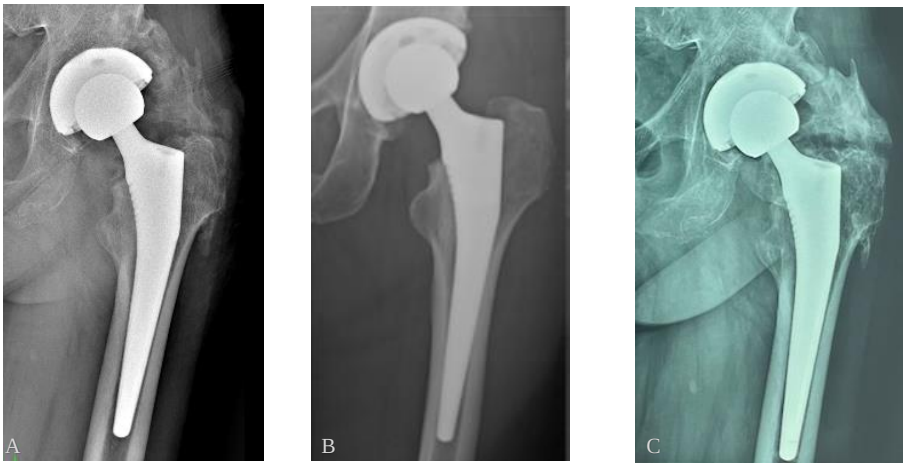


Fig. 7. *Postoperative X-ray checks at 0, 3 and 12 months. **A**): Post operative; **B**): At 3 months **C**): At 12 months.*

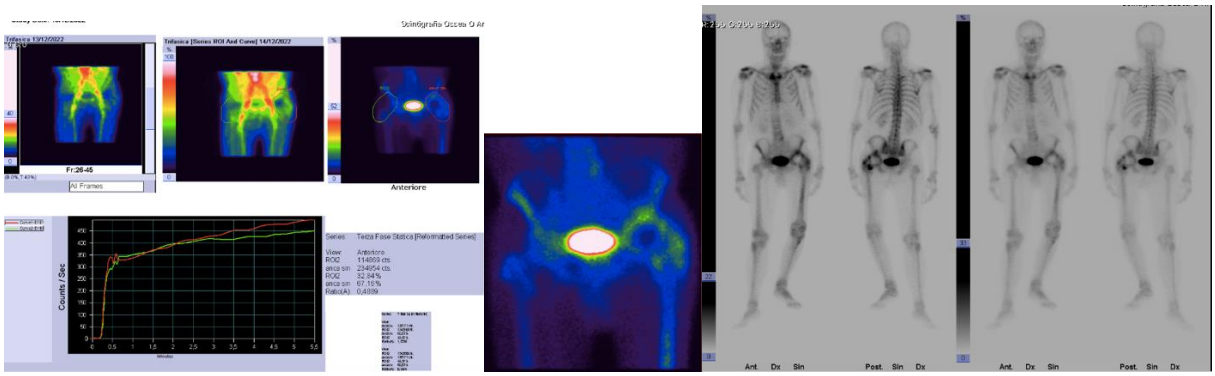


Fig. 8. *Scintigraphy performed at 4 months documenting significant hyperfixation in the late phase only as by osteoblastic type reaction.*

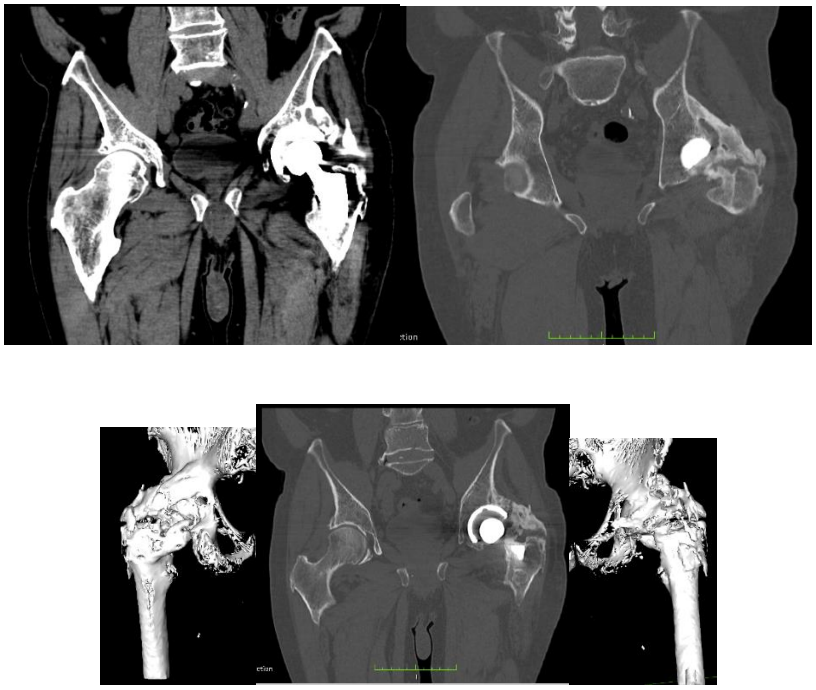


Fig. 9. *CT performed at 12 months post-implantation of PTA.*

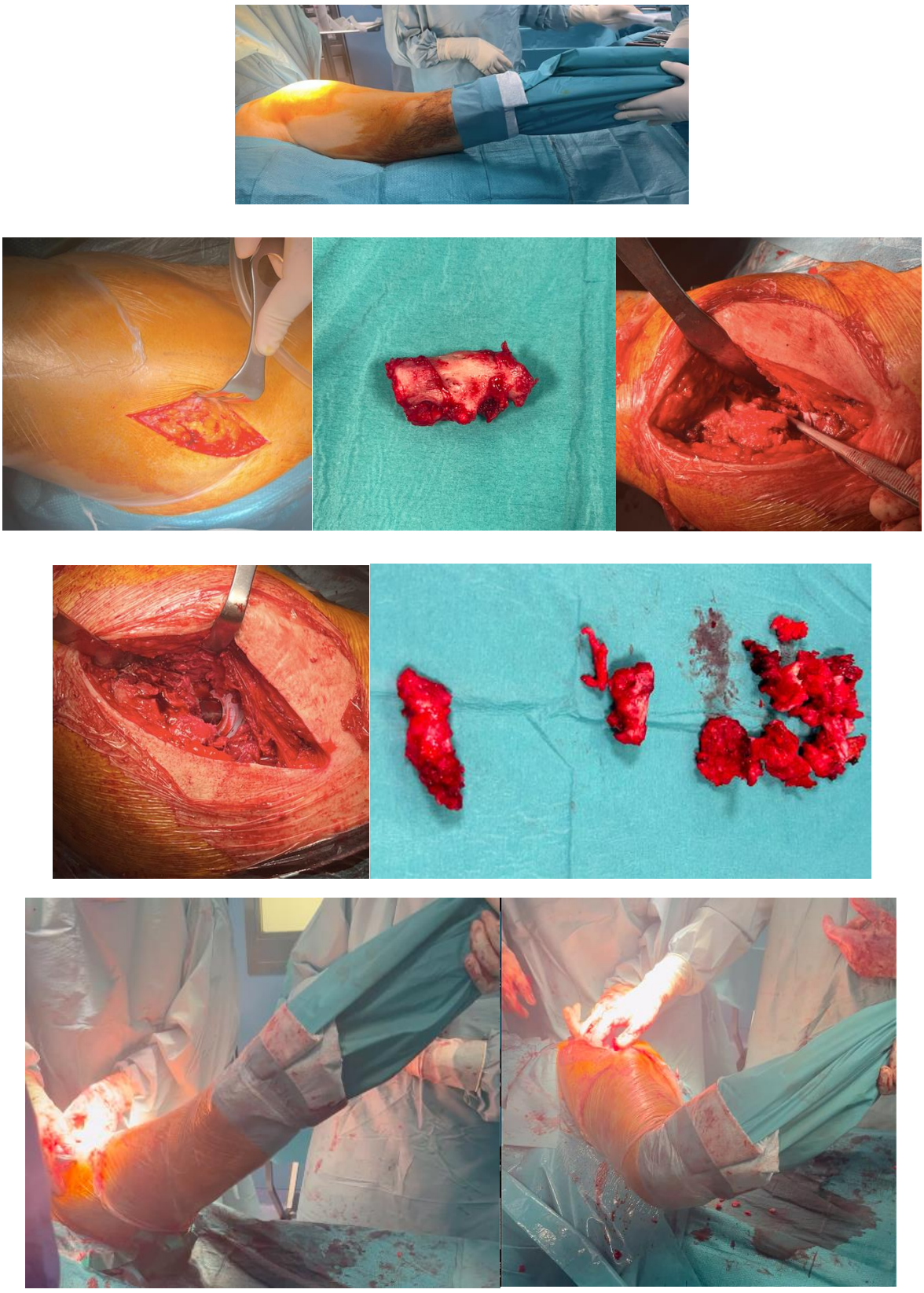


Fig. 10. *Intraoperative photographic sequence of arthrolysis surgery (bone bridge removal and joint mobilization), performed 14 months after PTA implantation.*

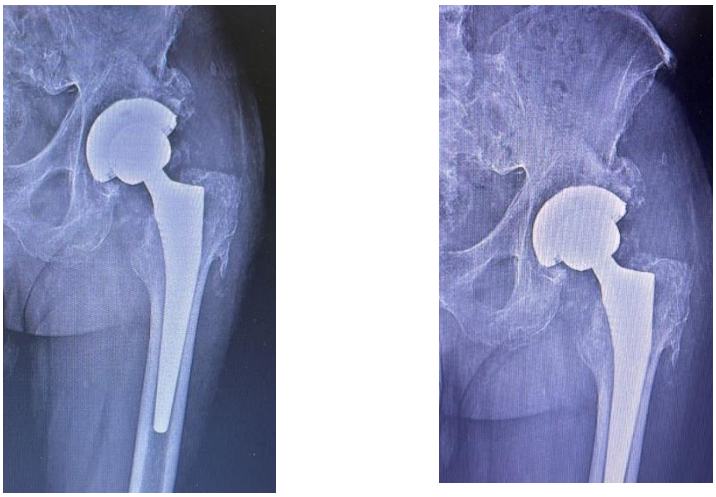


Fig. 11. *Radiographic control post-intervention of arthrolysis and clinical control at 1-month postarthrolysis.*

Clinical case B

65 years old - female - PTA system sx - Direct lateral access route.

Fig.12. *Preoperative: II-degree sec. the Kellgren-Lowrance classification, with severe limitation of pace and daily activities, more intense pain on the right.*

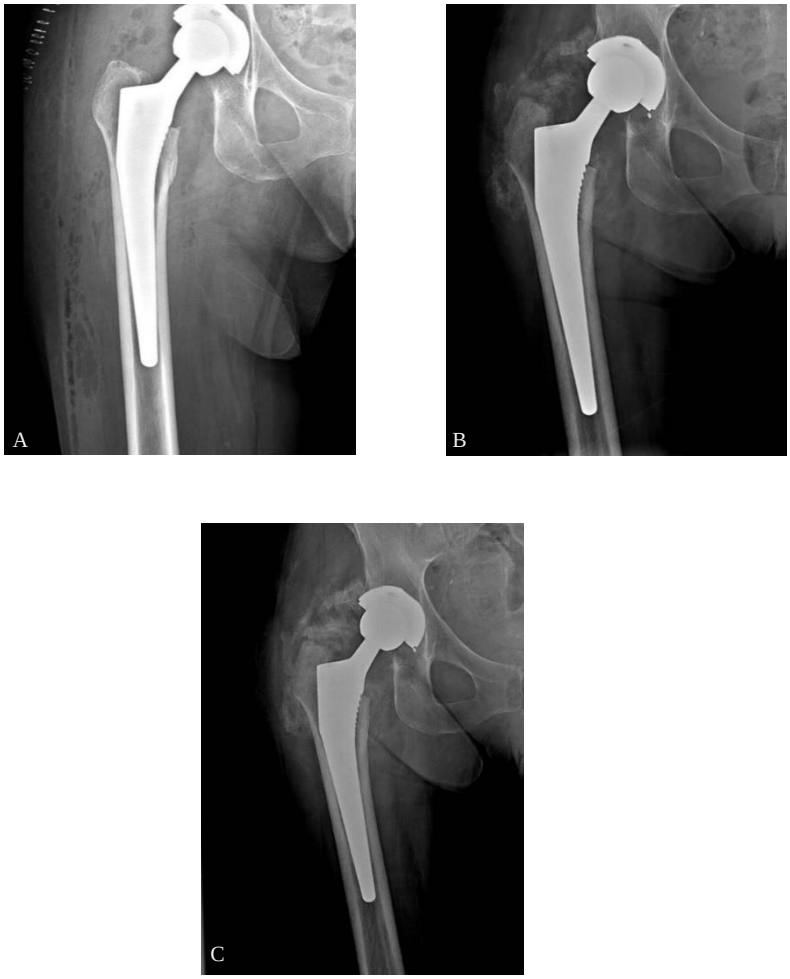


Fig. 13. *Postoperative X-ray checks at 0, 1, and 3 months. **A**): postoperative; **B**): at 1 month **C**): at 3 months.*

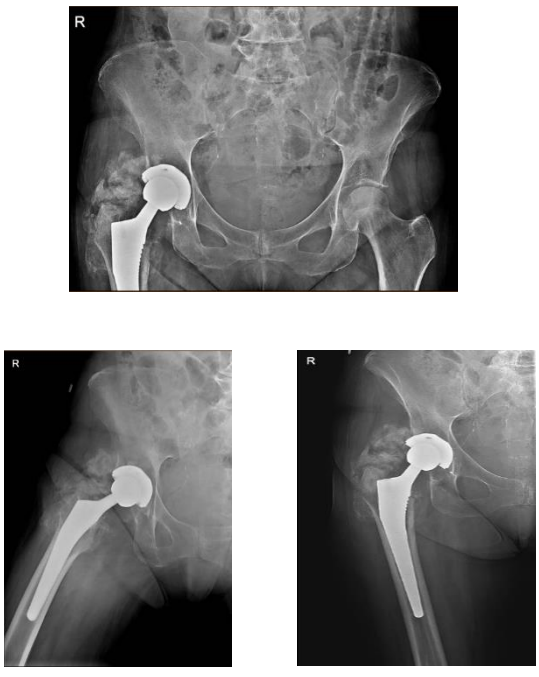


Fig. 14. *Radiographic and clinical control after 6 months (grade 3 Broker): Increase of ROM after a correct and regular cycle of FKT.*

CONCLUSIONS

Although heterotopic calcifications are a rarely disabling complication, they must be prevented with all the strategies known to date, especially because the weapons of prevention are clear and simple (23). Nevertheless, even with proper prevention, it is impossible to avoid it in all cases to substantially reduce its incidence, accurately evaluate patients with pre- and postoperative clinical and radiological controls, administer the right medical prophylaxis, and minimize surgical stress without the peremptory need to change the surgical approach radically, but managing the tissues in a less traumatic way possible, respecting muscles and tendons and limiting bleeding as much as possible with adequate intraoperative hemostasis are the best procedures.

Obviously, in the event of the onset of HO, a bloodless, physiotherapeutic, medical, and possibly radiant, timely, and close follow-up treatment is necessary to avoid the aggravation of local conditions. In cases where it was not possible to stem the appearance of calcifications with severe limitation of the ROM (grade 3-4 sec. Brooker), it will then be appropriate to evaluate a surgical treatment preceded by appropriate second and third level investigations and to be scheduled with appropriate timing.

REFERENCES

1. Amstutz HC, Fowble VA, Schmalzried TP, Dorcy FJ. Short-course indomethacin prevents heterotopic ossification in a high-risk population following total hip arthroplasty. *The Journal of Arthroplasty*. 1997;12(2):126-132. doi:https://doi.org/10.1016/s0883-5403(97)90058-9
2. Chao ST, Lee SY, Borden LS, Joyce MJ, Krebs VE, Suh JH. External Beam Radiation Helps Prevent Heterotopic Bone Formation in Patients With a History of Heterotopic Ossification. *The Journal of Arthroplasty*. 2006;21(5):731-736. doi:https://doi.org/10.1016/j.arth.2005.08.014
3. Willburger RE, Brinkhoff F, Nottenkämper J, Krapp J, Oberberg S. Heterotopic ossification after total hip arthroplasty: When is development completed? Journal of Orthopaedic Surgery and Research. 2022;17(1). doi:https://doi.org/10.1186/s13018-022-02959-z
4. Willburger RE, Brinkhoff F, Nottenkämper J, Krapp J, Oberberg S. Heterotopic ossification after total hip arthroplasty: When is development completed? *Journal of Orthopaedic Surgery and Research*. 2022;17(1). doi:https://doi.org/10.1186/s13018-022-02959-z
5. Eggli S, Woo A. Risk factors for heterotopic ossification in total hip arthroplasty. *Archives of Orthopaedic and Trauma Surgery*. 2001;121(9):531-535. doi:https://doi.org/10.1007/s004020100287
6. Ahrengart L, Lindgren U. Heterotopic Bone After Hip Arthroplasty Defining the Patient at Risk. *Clinical Orthopaedics and Related Research*. 1993;293(293):153-159. doi:https://doi.org/10.1097/00003086-199308000-00020
7. Eggli S, Rodriguez J, Ganz R. Heterotopic ossification in total hip arthroplasty: the significance for clinical outcome. *PubMed*. 2000;66(2):174-180.
8. Higo T, Mawatari M, Shigematsu M, Hotokebuchi T. The Incidence of Heterotopic Ossification After Cementless Total Hip Arthroplasty. *The Journal of Arthroplasty*. 2006;21(6):852-856. doi:https://doi.org/10.1016/j.arth.2005.10.016
9. Chalmers J, Gray DH, Rush J. Observations on the induction of bone in soft tissues. *The Journal of Bone and Joint*

Surgery British volume. 1975;57-B(1):36-45. doi:https://doi.org/10.1302/0301-620x.57b1.36

10. Zaccalini PS, Urist MR. Traumatic periosteal proliferations in rabbits. *The Journal of Trauma: Injury, Infection, and Critical Care*. 1964;4(3):344-357. doi:https://doi.org/10.1097/00005373-196405000-00008
11. Tonna EA, Cronkite EP. Autoradiographic Studies of Cell Proliferation in the Periosteum of Intact and Fractured Femora of Mice Utilizing DNA Labeling with H3-Thymidine. *Experimental Biology and Medicine*. 1961;107(4):719-721. doi:https://doi.org/10.3181/00379727-107-26733
12. Brooker AF, Bowerman JW, Robinson RA, Riley LH. Ectopic Ossification Following Total Hip Replacement. *The Journal of Bone & Joint Surgery*. 1973;55(8):1629-1632. doi:https://doi.org/10.2106/00004623-197355080-00006
13. Hug KT, Alton TB, Gee AO. In Brief: Classifications in Brief: Brooker Classification of Heterotopic Ossification After Total Hip Arthroplasty. *Clinical Orthopaedics & Related Research*. 2015;473(6):2154-2157. doi:https://doi.org/10.1007/s11999-014-4076-x
14. Edwards DS, Kuhn KM, Potter BK, Forsberg JA. Heterotopic Ossification. *Journal of Orthopaedic Trauma*. 2016;30(3):S27-S30. doi:https://doi.org/10.1097/bot.0000000000000666
15. Mavrogenis AF, Soucacos PN, Papagelopoulos PJ. Heterotopic Ossification Revisited. *Orthopedics*. 2011;34(3). doi:https://doi.org/10.3928/01477447-20110124-08
16. 12.Andreani L, Picece C, Castellini I, Ciapini G, Parchi PD, Lisanti M. Le calcificazioni eterotopiche nella protesica d'anca con accesso postero-laterale: incidenza, significato clinico e fattori di rischio. Giornale Italiano di Ortopedia e Traumatologia. 2016;51(4, Supplemento 1):384-385. doi:http://hdl.handle.net/11568/816491
17. 13.Bonicoli E. Via Anteriore Diretta vs Postero-Laterale. Revisione Clinica E Radiografica in Un Gruppo Omogeneo Di 60 Pazienti. Congresso Nazionale Siot Società Italiana di Ortopedia e Traumatologia; 2015.
18. Łęgosz P, Sarzyńska S, Pulik Ł, et al. Heterotopic ossification and clinical results after total hip arthroplasty using the anterior minimally invasive and anterolateral approaches. *Archives of Medical Science*. 2020;16(3):613-620. doi:https://doi.org/10.5114/aoms.2018.78653
19. Di Benedetto P, Zangari A, Magnanelli S, et al. Heterotopic Ossification in Primary Total Hip Arthroplasty: which is the role of drainage? *PubMed*. 2019;90(1-S):92-97. doi:https://doi.org/10.23750/abm.v90i1-s.8077
20. Cohn R, Schwarzkopf R, Jaffe F. Heterotopic ossification after total hip arthroplasty. *American journal of orthopedics (Belle Mead, NJ)*. 2024;40(11).
21. Morrey BF, Adams RA, Cabanela M. Comparison of Heterotopic Bone After Anterolateral, Transtrochanteric, and Posterior Approaches for Total Hip Arthroplasty. *Clinical Orthopaedics and Related Research*. 1984;&NA;(188):160???167. doi:https://doi.org/10.1097/00003086-198409000-00020
22. Schara K, Herman S. Heterotopic bone formation in total hip arthroplasty: predisposing factors, classification and the significance for clinical outcome. *PubMed*. 2001;68(2):105-108.
23. Baird EO, Kang QK. Prophylaxis of heterotopic ossification – an updated review. *Journal of Orthopaedic Surgery and Research*. 2009;4(1). doi:https://doi.org/10.1186/1749-799x-4-12

Case Report

PREDICTION OF LUMBAR DISC HERNIATION RESORPTION IN PATIENT WITH ACUTE LEFT SCIATICA: AN MR STUDY

A. Fontana[1], F. Bonetti[2], G. Sabetta[2], M. Frigerio[3] and M. Bonetti[3]

[1]Department of Radiology, Casa di Cura Villa Barbarano, Barbarano, Brescia, Italy;
[2]Vita-Salute San Raffaele University, Milan, Italy;
[3]Department of Neuroradiology, City of Brescia Clinical Institute , Brescia, Italy

Correspondence to:
Alessandro Fontana, MD
Casa di Cura Villa Barbarano,
Via Spiaggia d'Oro 21,
25087 Barbarano, (BS), Italy
e-mail: fontanagemini@gmail.com

ABSTRACT

Symptomatic lumbar disc herniations are very common. Hernia resorption can occur through a "self-healing" process, however, this phenomenon remains poorly understood. The Authors present a rare case of a patient suffering from acute left lumbosciatica, in which the results of the Magnetic Resonance Imaging (MRI) suggested a future natural healing of the herniated disc. In regard to the treatment, in particular, thanks to the MRI scans, it was possible to appreciate the concomitance of partial gaseous vacuolization of the hernia with colliquation of the peripheral portion, which are predictive signals of a future complete natural reabsorption of the herniated disc.

KEYWORDS: *disc, herniation, outcomes, prediction, prognosis, resorption*

INTRODUCTION

Almost 80% of sciatica due to herniated discs evolve favorably towards spontaneous healing after about a year; numerous studies have shown that herniated discs can reduce in size or even disappear, simultaneously with the reduction of clinical signs.

To date, it is difficult to formulate a precise prognosis of disc sciatica and, above all, to be able to determine its duration and evolution because various factors come into play, mainly mechanical and enzymatic-inflammatory phenomena. Mechanical factors are those linked to the anatomy of the hernia and the structures that surround it, such as nerve roots, ligaments, veins, meninges, and epidural fat.

In regards to enzymatic and inflammatory factors, it is necessary to underline that although the intervertebral disc is paradoxically the most important avascular tissue in the body, its posterior part is the site of degradation reactions. These reactions are caused by metalloproteases and cytokines, which are responsible for pain but also the degradation and reabsorption of the herniated material.

Thanks to the development of Computed Tomography (CT) and especially Magnetic Resonance Imaging (MRI), regression and even the disappearance of herniated discs, simultaneously with the improvement of clinical signs, could be highlighted in several studies. The clinical improvement has often preceded the radiographic modification of the hernia: the decrease in the volume of the hernia is more important and rapid if it is a voluminous and migrated hernia; small hernias with intact posterior vertebral ligaments are less likely to regress.

Received: 19 August 2023
Accepted: 27 September 2023

Two types of cells can be involved in the degradation of the intervertebral disc: the disc's own cells and macrophages; morphologically the cells of the annulus and nucleus are very similar to articular chondrocytes. These cells synthesize the extracellular matrix and produce proteoglycans; macrophages and other mononuclear cells come from the neo-vascularization and granulation tissue surrounding the fibro-cartilaginous fragment. This granulation tissue is present around the extra-ligamentous hernias. Furthermore, there are enzymatic factors, such as metalloproteases (collagenase, stromelysin, etc.), which play a major role in the disc degradation process.

Cytokines belong to the other family of enzymes, of which interleukin I is the main pro-inflammatory present in the degenerated disc, as well as TNF, which plays a direct role in the genesis of pain, as it causes lesional edema and demyelination of nerve fibers.

Therefore, as a direct consequence of the presence of herniated material, a true immunological reaction develops: neo-vascularization appears, and the active macrophages produce TNF, which induces the production of stromelysin, which leads to the reabsorption of the disc tissue due to progressive degradation of the extracellular matrix. This mechanism could thus explain the more rapid degradation of large migrated hernias; it is because of this mechanism that ED can regress over time until it disappears spontaneously (1-4).

CASE REPORT

G.A., a 67-year-old male, following the sudden onset of acute left lumboscitalgia, asks for advice from his doctor, who recommends thiocolchicoside 4mg/2ml IM associated with piroxicam 20mg/1ml. In the case of the persistence of the symptoms, the physician asks to carry out an MRI scan. In fact, the symptoms appear to be completely resistant to the established pharmacological therapy so the MRI scan is carried out 20 days after the onset of the symptomatology.

The Magnetic Resonance investigation shows the presence of a herniated extruded disc in the left paramedian area corresponding to L5-S1, with extruded material that migrates caudally, positioning itself behind the posterior wall of S1, imprinting the left S1 root, therefore with a situation of disc-radicular conflict, a finding consistent with the sciatica symptoms in the distribution area of the left S1 complained of by the patient.

The disc herniation presents a dual component of signal hypointensity in T1-dependent sequences with a more cranial area characterized by marked hypointensity, a finding compatible with a partial vacuolation of the hernia and a more cranial portion with disc-like signal hypointensity (Fig. 1). The T2-dependent scans document both in axial and sagittal projections, the presence of a more cranial area in the context of the hernia characterized by signal hypointensity compatible with the partial vacuolization of the hernia and a peripheral "target" portion around the vacuolized portion characterized by signal hyperintensity compatible with a pattern of colliquation of this tissue. In contrast, in the more caudal scans, there is an evident hernial component of disc-like intensity. (Fig 2 a-c).

Fig 1. *Sagittal T1 MRI: left paramedian extruded disc herniation. The area of more marked hypointensity* ***(arrow)*** *appears to be partially vacuolized.*

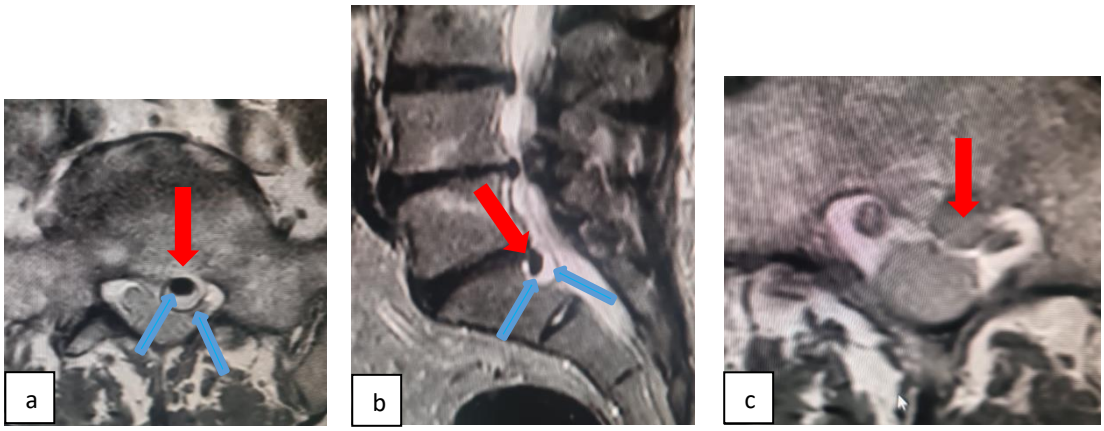


Fig 2. ***a****): T2 axial scan: Partial vacuolization of the hernia (hypointense signal) (**red arrow**) with hyperintense "target" peripheral portion with high water content in relation to colliquation phenomena (**blue arrows**); **b**): Sagittal T2 STIR MRI: partial vacuolation of the hernia (**red arrow**), peripheral colliquation (**blue arrows**); **c**): More caudal T2 axial MRI scan: the herniated disc presents a disc-like intensity in its most caudal portion (**red arrow**).*

DISCUSSION

Spontaneous healing of herniated discs was first reported in 1984 (5, 6). In 1990 Saal et al. (7) selected patients diagnosed with herniated discs by Computed Tomography (CT) in order to explore their natural history. This demonstrates that the larger the dimensions of the disc protrusions, the greater the signs of resorption, while the smaller ones had lower resorption ratios.

In recent years, researchers have paid increasing attention to the resorption of herniated discs. A meta-analysis showed that the average incidence of symptomatic resorption of herniated discs was 62-66% in 38 clinical studies reported over the last 30 years (8). A recent retrospective analysis also showed that 59.06% of patients treated conservatively had resorption of the herniated disc (9).

Furthermore, the North American Spine Society's evidence-based clinical guidelines for the diagnosis and treatment of lumbar disc herniation with radiculopathy (10) emphasized that with the advancement of natural history, the herniated disc material of most patients may spontaneously shrink or degenerate. At the same time, as the size of the hernia decreases, the clinical symptoms also improve. The phenomenon of herniated disc resorption is, therefore, not random, and the clinical symptoms of most patients can be alleviated or even disappear without necessarily having to resort to surgery.

It is generally accepted that herniated discs are more prone to regression than other types of disc disease. In 2015, Chiu et al. systematically evaluated the probability of resorption of different types of herniated discs, concluding that the majority of herniated discs can be spontaneously absorbed after conservative treatment and that extruded herniations had higher regression rates (8, 11).

Resorption may be favored when the herniated disc contains most of the nucleus pulposus, while the presence of cartilaginous tissue inhibits the resorption of the herniation. To demonstrate this, Iwabuchi et al. (12, 13) used plain MRI to predict the effect of herniated disc composition on resorption based on signal intensity in T1 and T2 weightings. Failure of spontaneous absorption of herniated discs is mainly related to cartilage protrusion (14).

The effect of the disc component on resorption may depend mainly on whether it promotes vascularization, while hyaline cartilage fragments tend to resist revascularization and resorption. Therefore, if the MRI shows that the herniation is predominantly made up of the nucleus pulposus, this could favor resorption; however, a high percentage of cartilaginous tissue can make conservative treatment of the herniated disc difficult and may not be effective.

It is now known that the intervertebral disc dissociated from the epidural space can cause an autoimmune reaction, which leads to an inflammatory reaction and to the formation of surrounding granulation tissue, which manifests itself as a ring peripherical to the herniated disc tissue. MRI after administration of a paramagnetic contrast medium can perfectly document this mechanism. The evident increase in the edges of the herniated tissue suggests the possibility of spontaneous absorption, which is also considered an important factor in evaluating spontaneous regression of the herniated disc (15-17).

Ring enhancement is believed to be related to the vascularization of the lumbar disc herniation and the formation of inflammatory granulation tissue (18-19), and the neovascularization and inflammatory response of the herniated disc are

the key factors for resorption (20). Therefore, magnification around the protrusion can be used as an imaging manifestation of new blood vessel formation and inflammation in the prominent tissue, which may predict resorption.

Three mechanistic hypotheses have been reported in the literature, which could be jointly involved in the resolution and disappearance of the herniated disc. The first mechanism is the retraction of the protrusion, which can occur without separation of the protrusion from the fibrous ring (21). The second mechanism is the gradual dehydration and contraction of the herniated nucleus pulposus, which causes the retraction of the protuberances in the annulus fibrosus (22). A third mechanism, which has received widespread attention, states that fragments of herniated disc material enter the epidural space, triggering an autoimmune response that includes inflammatory cell infiltration and neovascularization. The autoimmune system recognizes enial protrusions as “foreign” in the vertebral epidural vascular space, which in turn triggers a cascade of inflammatory responses, including neovascularization, matrix protease activation, increased levels of inflammatory mediators, phagocytosis of inflammatory cells and enzymatic degradation (1, 7, 8).

Macrophages are a key immune regulator that triggers the resorption of extruded herniated discs. Macrophage infiltration and activation are critical steps in the resorption process. In herniated disc fragments, there is increased expression of IL-12 and IFN-γ compared to protruded discs (23). IFN-γ produced by Th1 lymphocytes recruits and activates more macrophages (24). Specifically, contact of the extruded hernia tissue with the systemic circulation leads to lymphocyte activation and secretion of IFN-γ, promoting macrophage recruitment. High expression of IFN-γ in herniated disc may represent a specific immune response against the herniated tissue (25). These findings suggested that the mode of immune activation in the extruded hernia involves the infiltration and activation of macrophages.

Vascular growth has been recognized as an essential feature of spontaneous absorption of extruded hernias (26, 27). Histological examination revealed neovascularization at the site of a herniated disc (27, 28). Mediators inducing neovascularization of the extruded hernia mainly include TNF-α, VEGF, basic fibroblast growth factor (bFGF), and platelet-derived growth factor (PDGF) (15, 28).

The activity of different types of macrophages and secreted pro-angiogenic mediators are the main regulators of neovascularization in the inflammatory response. In the case presented, the partial vacuolation of the hernia peripherally to which colliquated tissue is recognizable to signify the ongoing presence of a process of degeneration or organic disintegration of the extruded disc hernia appears to be predictive signs of possible dehydration of the herniated disc extruded with possible resolution of sciatica symptoms without having to resort to surgical intervention, thus giving the possibility of managing the patient conservatively.

These MRI findings appear to be rarely encountered in daily clinical practice. Therefore, we wanted to highlight them in this note as a predictive finding of possible natural healing of an extruded disc herniation in a patient suffering from acute left sciatica.

CONCLUSIONS

Extruded disc herniation is one of the most common vertebral column diseases. With the support of radiological technology, especially MRI, doctors can scientifically and reasonably diagnose the presence of extruded disc herniation and then provide patients with feasible and effective treatment strategies.

The extrusion of the disc material creates conditions favorable to the infiltration of macrophages and neovascularization, leading to possible natural healing. However, if the tissue of the herniated disc contains a greater percentage of cartilage, this does not favor the infiltration of macrophages and the growth of blood vessels, thus preventing the occurrence of reabsorption of the extruded hernia. In the rare case presented by us, the characteristics of the MRI are predictive for possible natural healing of the extruded hernia, therefore clinically addressable with conservative therapy or even without intervening while waiting for healing, thus avoiding the need for surgical intervention.

REFERENCES

1. Hornung AL, Barajas JN, Rudisill SS, et al. Prediction of lumbar disc herniation resorption in symptomatic patients: a prospective, multi-imaging and clinical phenotype study. *The Spine Journal*. 2023;23(2):247-260. doi:https://doi.org/10.1016/j.spinee.2022.10.003
2. Hornung AL, Baker JD, Mallow GM, et al. Resorption of Lumbar Disk Herniation: Mechanisms, Clinical Predictors, and Future Directions. *JBJS reviews*. 2023;11(1). doi:https://doi.org/10.2106/JBJS.RVW.22.00148
3. Liu D, Bai Y, Zhang G, Chen L, Xu W, Liu J. [Research progress of resorption of lumbar disc herniation]. *Zhongguo Xiu Fu Chong Jian Wai Ke Za Zhi*. 2022;36(10):1312-1316. doi:https://doi.org/10.7507/1002-1892.202204105
4. Lee J, Kim J, Shin JS, et al. Long-Term Course to Lumbar Disc Resorption Patients and Predictive Factors Associated with Disc Resorption. *Evidence-Based Complementary and Alternative Medicine*. 2017;2017:1-10.

doi:https://doi.org/10.1155/2017/2147408

5. Guinto Jr F, Hashim H, Stumer M. CT demonstration of disk regression after conservative therapy. *AJNR American journal of neuroradiology*. 1984;5(5).
6. Zhong M, Liu JT, Jiang H, et al. Incidence of Spontaneous Resorption of Lumbar Disc Herniation: A Meta-Analysis. *Pain Physician*. 2017;20(1):E45-E52.
7. SAAL JA, SAAL JS, HERZOG RJ. The Natural History of Lumbar Intervertebral Disc Extrusions Treated Nonoperatively. *Spine*. 1990;15(7):683-686. doi:https://doi.org/10.1097/00007632-199007000-00013
8. Chiu CC, Chuang TY, Chang KH, Wu CH, Lin PW, Hsu WY. The probability of spontaneous regression of lumbar herniated disc: a systematic review. *Clinical rehabilitation*. 2015;29(2):184-195. doi:https://doi.org/10.1177/0269215514540919
9. Ma Z, Yu P, Jiang H, et al. Conservative Treatment for Giant Lumbar Disc Herniation: Clinical Study in 409 Cases. *Pain Physician*. 2021;24(5):E639-E648.
10. Kreiner DS, Hwang SW, Easa JE, et al. An evidence-based clinical guideline for the diagnosis and treatment of lumbar disc herniation with radiculopathy. *The spine journal : official journal of the North American Spine Society*. 2014;14(1):180-191. doi:https://doi.org/10.1016/j.spinee.2013.08.003
11. Macki M, Hernandez-Hermann M, Bydon M, Gokaslan A, McGovern K, Bydon A. Spontaneous regression of sequestrated lumbar disc herniations: Literature review. *Clinical Neurology and Neurosurgery*. 2014;120:136-141. doi:https://doi.org/10.1016/j.clineuro.2014.02.013
12. Iwabuchi M, Murakami K, Ara F, Otani K, Kikuchi SI. THE PREDICTIVE FACTORS FOR THE RESORPTION OF A LUMBAR DISC HERNIATION ON PLAIN MRI. *FUKUSHIMA JOURNAL OF MEDICAL SCIENCE*. 2010;56(2):91-97. doi:https://doi.org/10.5387/fms.56.91
13. Shan Z, Fan S, Xie Q, et al. Spontaneous Resorption of Lumbar Disc Herniation Is Less Likely When Modic Changes Are Present. *Spine*. 2014;39(9):736-744. doi:https://doi.org/10.1097/brs.0000000000000259
14. Yamashita K, Hiroshima K, Kurata A. Gadolinium-DTPA-Enhanced Magnetic Resonance Imaging of a Sequestered Lumbar Intervertebral Disc and Its Correlation with Pathologic Findings. *Spine*. 1994;19(Supplement):479-482. doi:https://doi.org/10.1097/00007632-199402001-00021
15. Sakai T, Tsuji T, Asazuma T, Yato Y, Matsubara O, Nemoto K. Spontaneous resorption in recurrent intradural lumbar disc herniation. *Journal of Neurosurgery: Spine*. 2007;6(6):574-578. doi:https://doi.org/10.3171/spi.2007.6.6.11
16. Komori H, Okawa A, Haro H, Muneta T, Yamamoto H, Shinomiya K. Contrast-Enhanced Magnetic Resonance Imaging in Conservative Management of Lumbar Disc Herniation. *Spine*. 1998;23(1):67-73. doi:https://doi.org/10.1097/00007632-199801010-00015
17. Youichi Kawaji, Uchiyama S, Yagi E. Three-dimensional evaluation of lumbar disc hernia and prediction of absorption by enhanced MRI. *Journal of Orthopaedic Science*. 2001;6(6):498-502. doi:https://doi.org/10.1007/s007760100004
18. Löhr M, Lebenheim L, Berg F, et al. Gadolinium enhancement in newly diagnosed patients with lumbar disc herniations are associated with inflammatory peridiscal tissue reactions – Evidence of fragment degradation? *Clinical Neurology and Neurosurgery*. 2014;119:28-34. doi:https://doi.org/10.1016/j.clineuro.2014.01.008
19. Haro H, Kato T, Komori H, Osada M, Shinomiya K. Vascular endothelial growth factor (VEGF)-induced angiogenesis in herniated disc resorption. *Journal of Orthopaedic Research*. 2002;20(3):409-415. doi:https://doi.org/10.1016/s0736-0266(01)00150-4
20. Orief T, Orz Y, Attia W, Almusrea K. Spontaneous Resorption of Sequestrated Intervertebral Disc Herniation. *World Neurosurgery*. 2012;77(1):146-152. doi:https://doi.org/10.1016/j.wneu.2011.04.021
21. Kim ES, Oladunjoye AO, Li JA, Kim KD. Spontaneous regression of herniated lumbar discs. *Journal of Clinical Neuroscience*. 2014;21(6):909-913. doi:https://doi.org/10.1016/j.jocn.2013.10.008
22. Tsarouhas A, Soufla G, Katonis P, Pasku D, Vakis A, Spandidos DA. Transcript levels of major MMPs and ADAMTS-4 in relation to the clinicopathological profile of patients with lumbar disc herniation. *European Spine Journal*. 2010;20(5):781-790. doi:https://doi.org/10.1007/s00586-010-1573-9
23. Park JB, Chang H, Kim YS. The Pattern of Interleukin-12 and T-Helper Types 1 and 2 Cytokine Expression in Herniated Lumbar Disc Tissue. *Spine*. 2002;27(19):2125-2128. doi:https://doi.org/10.1097/00007632-200210010-00009
24. Shamji MF, Setton LA, Jarvis W, et al. Pro-inflammatory cytokine expression profile in degenerative and herniated human intervertebral disc tissues. *Arthritis & Rheumatism*. 2010;62(7):NA-NA. doi:https://doi.org/10.1002/art.27444
25. Autio RA, Karppinen J, Niinimäki J, et al. Determinants of Spontaneous Resorption of Intervertebral Disc Herniations. *Spine*. 2006;31(11):1247-1252. doi:https://doi.org/10.1097/01.brs.0000217681.83524.4a
26. Ozaki S, Muro T, Ito S, Mizushima M. Neovascularization of the outermost area of herniated lumbar intervertebral discs. *Journal of Orthopaedic Science*. 1999;4(4):286-292. doi:https://doi.org/10.1007/s007760050105
27. Yasuma T, Arai K, Yamauchi Y. The Histology of Lumbar Intervertebral Disc Herniation. *Spine*. 1993;18(13):1761-1765. doi:https://doi.org/10.1097/00007632-199310000-00008
28. Tolonen J, Grönblad M, Virri J, Seitsalo S, Rytömaa T, Karaharju EO. Platelet-derived growth factor and vascular endothelial growth factor expression in disc herniation tissue: an immunohistochemical study. *European Spine Journal*. 1997;6(1):63-69. doi:https://doi.org/10.1007/bf01676576

Case Report

SURGICAL TREATMENT IN HAWKINS I TYPE TALUS NECK FRACTURES: A CASE REPORT

P. Rao, J. Troiano, A. Pantalone, D. Bruni and R. Buda

Clinic of Orthopaedics and Traumatology, Department of Medicine and Aging Sciences, "G. d 'Annunzio" University of Chieti-Pescara, Chieti, Italy

**Correspond to:*
Roberto Buda, MD
Clinic of Orthopaedics and Traumatology,
Department of Medicine and Aging Sciences,
"G. d 'Annunzio" University of Chieti-Pescara,
Chieti, Italy
e-mail: roberto.buda@unich.it

ABSTRACT

Talus fractures are a rare occurrence, accounting for about 0.5% of all fractures. These fractures are classified into 4 types, according to Hawkins. Hawkins type 1 fractures are compound fractures; due to their low incidence, it is difficult to evaluate long-term results and prognosis. This kind of fracture could be treated conservatively, but the importance of surgical treatment should not be underestimated. The present article describes a case of Hawkins type I talar neck fracture, early recognized and successfully surgically treated by percutaneous screws. At a 12-month follow-up, the patient reported returning to full-time employment, resuming all activities without pain or functional limitation. The decision-making in Hawkins type I talus neck fractures is not well defined because of the rarity of the incidence and the low number of cases in the literature. This case report supports the success and good outcomes of a prompt surgical approach in Hawkins Type-1 fractures.

KEYWORDS: *talar neck fracture, Hawkins type I, conservative treatment, surgical treatment*

INTRODUCTION

The talus is the 2nd largest bone of the foot and is covered by 60% cartilage. It has no direct muscle insertions (1) and is characterized by a limited vascular supply (2, 3). Talar fractures are a rare occurrence, accounting for 0.5% of all fractures, but should not be underestimated due to a possible severe prognosis. The average age of these injuries is between 30 and 38 years, with a male/female ratio of 3 to 1. Talus neck fractures represent half of all talus fractures and result from forced foot extension (4, 5).

Ischemic necrosis of the talus body complicates half of talus neck fractures due to the interruption of the main nutritional arteries, which penetrate the bone in the neck (6, 7, 8). According to Hawkins classification (9), talar neck fractures can be classified into four types, each with a different percentage risk of avascular necrosis. TYPE 1: compound fracture of the neck of the talus and without dislocation of the tibio-talar and sub-talus. Risk of osteonecrosis (NAV) 0-15%. TYPE 2: displaced fracture of the neck of the talus with subluxation or dislocation of the sub-talus. NAV 20-50%. TYPE 3: displaced fracture of the neck of the astragalus with subluxation or dislocation of the sub-talar and tibia-talus. NAV 70-100%. TYPE 4: displaced fracture of the neck of the astragalus with subluxation or dislocation of the sub-talus, tibio-talar, and talus-navicular. NAV 100%. The present case regards a 21-year-old man affected by a talus fracture

Received: 6 September 2023
Accepted: 12 October 2023

Hawkins type 1. The therapeutic algorithm is debated due to the rarity of fractures. After a detailed presentation, the case will be discussed.

CASE PRESENTATION

A 21-year-old man came to the emergency room for orthopedic counseling. He reported sprained trauma of his right ankle due to an accidental fall from the stairs of his home, describing a movement of dorsal hyperflexion of the foot during the landing phase. An edematous and tumid ankle was observed, with widespread pain at the tarsal level. The patient, a tattoo artist by profession, was a heavy smoker (> 20 cigarettes/day) and had no history of comorbidities. Anteroposterior, oblique, and lateral ankle radiographs showed a compound talus fracture (Fig. 1).

The ankle was immobilized with a high-knee cast. A CT scan with 3D reconstructions was performed to study the fracture better and its joint involvement, showing a compound fracture of the neck of the talus (Fig. 2). Taking into account the ankle edema, the patient's heavy smoking history and his high functional demand, an osteosynthesis treatment was opted for. The fracture was synthesized with two cannulated screws with a washer of 4 x 45 mm and 4 x 40 mm in anteromedial percutaneous access (Fig. 3). The patient was discharged with a ban on loading on the operated limb, a plaster cast, and anti-thromboembolic prophylaxis. The plaster cast was removed after 30 days, and the patient was allowed to walk with a skimming load for the following 30 days. After 60 days from surgery, complete weight bearing was permitted, but sports were not allowed for another month. The patient had no clinical and radiographic signs of avascular necrosis 120 days after surgery, with an AOFAS score of 100/100. A new clinical and radiographic evaluation was performed about 1 year later (Fig. 4, 5). The patient reported returning to his full-time employment, resuming all activities in the absence of pain, and showing extreme satisfaction with the treatment received.

Fig. 1. *Pre-operative X-ray.*

Fig. 2. *Sagittal view of pre-operative CT scan.*

Fig. 3. *Post-operative X-ray.*

Fig. 4. *X-ray under load one year after surgery.*

Fig. 5. *Clinical image one year after surgery.*

DISCUSSION

The rarity of Hawkins I fractures means their management is not well established. Furthermore, it is not easy to assess the long-term prognosis. Although infrequent, these injuries may have devastating complications. Therefore, their evaluation requires a good understanding of the biology, vascularization, and anatomy of the talus (10). Type I neck fracture may be eligible for non-surgical treatment (11). Conservative treatment is a rational choice if the fracture is compound and the joint surfaces are aligned. The non-surgical treatment requires immobilization and a prolonged absence of load, with complications that can include loss of reduction of the fracture but also excessive stiffness of foot and ankle, algodystrophy, and osteopenia. For Kopp et al. (12), internal fixation is indicated in non-displaced Hawkins type 1 fractures. For Abdelkafy et al. (13), internal fixation of a simple talus neck fracture usually requires 2 screws in compression.

Morphometrically, it should be considered that the neck of the talus deviates medially with respect to the long axis of the talar body and plantarly. The aim is to position the screws to provide maximum interfragmentary compression at the fracture site. This leads to an increase in the probability of absolute stability with consequent reduction of complications such as avascular necrosis of the body of the talus. However, it is worth considering how the surgical approach to talar lesions should be performed by experienced surgeons. For Zeman et al. (14) the outcome of these procedures depends on the experience and skills of the surgeon and the system organization of the surgical department.

This case report deals with the surgical treatment of a Hawkins I fracture. The patient's smoking, his high functional demand, with the need to return to daily habits as quickly as possible have directed our decision-making toward a synthesis with 2 percutaneous screws. The patient started ankle mobilization at 30 days, freeing him from immobilization and starting the grazing load earlier. Swanson et al. (15) show that rigid internal fixation of type I talus neck fractures allows for early ankle mobilization. Conversely, conservative treatment presupposes a resumption of weight after 6-8 weeks (11). About 12 months after surgery and 120 days after surgery, our patient reported an AOFAS score of 100/100, proving extremely satisfied with the treatment received.

CONCLUSIONS

Type I talus neck fractures, according to Hawkins, are rare injuries that can lead to unpleasant and potentially complex complications. The decision-making on their treatment is not well established due to their low incidence and the rarity of case reports in Literature. This case report supports the success and positive outcomes of a prompt surgical approach in Hawkins Type-1 fractures.

REFERENCES

1. Bartoníček J, Rammelt S, Naňka O. Anatomy of the Subtalar Joint. *Foot and Ankle Clinics*. 2018;23(3):315-340. doi:https://doi.org/10.1016/j.fcl.2018.04.001
2. Prasad SA, Rajasekhar SSSN. Morphometric analysis of talus and calcaneus. *Surgical and Radiologic Anatomy*. 2018;41(1):9-24. doi:https://doi.org/10.1007/s00276-018-2101-6
3. Mulfinger GL, Trueta J. The blood supply of the talus. *The Journal of Bone and Joint Surgery British volume*. 1970;52-B(1):160-167. doi:https://doi.org/10.1302/0301-620x.52b1.160
4. McKeever FM. Fracture of the neck of the astragalus. *Archives of Surgery*. 1943;46(5):720-720. doi:https://doi.org/10.1001/archsurg.1943.01220110136022
5. Brockett CL, Chapman GJ. Biomechanics of the ankle. *Orthopaedics and Trauma*. 2016;30(3):232-238. doi:https://doi.org/10.1016/j.mporth.2016.04.015
6. Miller AN, Prasarn ML, Dyke JP, Helfet DL, Lorich DG. Quantitative Assessment of the Vascularity of the Talus with Gadolinium-Enhanced Magnetic Resonance Imaging. *The Journal of Bone & Joint Surgery*. 2011;93(12):1116-1121. doi:https://doi.org/10.2106/jbjs.j.00693
7. Canale ST, Kelly FB. Fractures of the neck of the talus. Long-term evaluation of seventy-one cases. *The Journal of Bone and Joint Surgery American Volume*. 1978;60(2):143-156.
8. Donnelly EF. The Hawkins Sign. *Radiology*. 1999;210(1):195-196. doi:https://doi.org/10.1148/radiology.210.1.r99ja08195
9. Hawkins LG. Fractures of the neck of the talus. *The Journal of Bone and Joint Surgery American Volume*. 1970;52(5):991-1002.
10. Beltran MJ, Mitchell PM, Collinge CA. Posterior to Anteriorly Directed Screws for Management of Talar Neck Fractures. *Foot & Ankle International*. 2016;37(10):1130-1136. doi:https://doi.org/10.1177/1071100716655434

11. Buckley R, Tough S, McCormack R, et al. Operative compared with nonoperative treatment of displaced intra-articular calcaneal fractures: a prospective, randomized, controlled multicenter trial. *J Bone Joint Surg Am*. 2002;84(10):1733-1744. doi:10.2106/00004623-200210000-00001
12. Kopp L, Obruba P, Riegl J, Meluzinová P, Edelmann K. Surgical Management of Talus Fractures: Mid-Term Functional and Radiographic Outcomes. *Acta Chirurgiae Orthopaedicae Et Traumatologiae Cechoslovaca*. 2013;80(2):165-170. doi:https://doi.org/10.55095/achot2013/026
13. Abdelkafy A, Imam MA, Sokkar S, Hirschmann M. Antegrade-Retrograde Opposing Lag Screws for Internal Fixation of Simple Displaced Talar Neck Fractures. *The Journal of Foot & Ankle Surgery*. 2014;54(1):23-28. doi:https://doi.org/10.1053/j.jfas.2014.09.046
14. Zeman J, Matějka J, Pavelka T. Surgical Treatment for Factures of the Neck and Body of the Talus. *Acta chirurgiae orthopaedicae et traumatologiae Cechoslovaca*. 2012;79(2):119-123. doi:https://doi.org/10.55095/achot2012/017
15. Swanson TV, Bray TJ, Holmes GB. Fractures of the talar neck. A mechanical study of fixation. *The Journal of Bone and Joint Surgery*. 1992;74(4):544-551. doi:https://doi.org/10.2106/00004623-199274040-00010.

Case Report

BILATERAL SIMULTANEOUS ATYPICAL FEMORAL PERIPROSTHETIC FRACTURE. A CASE REPORT AND REVIEW OF THE LITERATURE

L.N. Scaramuzzi[1], L. Scialpi[2], A. Carlet[3], I. Rausa[1], G. Sammarco [2], and G. Solarino[1]

[1]Orthopaedic & Trauma Unit, School of Medicine, University of Bari Aldo Moro, AOU Consorziale "Policlinico", Bari, Italy;
[2]Orthopaedic & Trauma Unit, Santissima Annunziata Hospital, Taranto, Italy;
[3]Orthopaedic & Trauma Unit, San Giacomo Hospital, Monopoli, Italy

**Correspondence to:*
Scaramuzzi Luciano Nicola, MD
Orthopaedic & Trauma Unit,
School of Medicine,
University of Bari Aldo Moro,
AOU Consorziale "Policlinico",
Piazza Giulio Cesare 11,
70124 Bari, Italy
e-mail: lucianoscaramuzzi@gmail.com

ABSTRACT

Atypical femoral fracture is one of the many complications after the long-term use of bisphosphonates. Long-term use of bisphosphonates (BPs) has been associated with a specific type of tensile-side femoral stress fracture known as Atypical Femoral Fracture (AFF). The American Society for Bone and Mineral Research has officially excluded periprosthetic femoral fractures (PFFs) from the definition of atypical femoral fractures (AFFs). However, several case reports correlate prolonged BP use with the occurrence of a type of PFF with an atypical pattern (atypical PFF, APFF). The aim of the present study is to report a case of bilateral APFF after prolonged BP use. A 77-year-old female with a history of long-term bisphosphonate use (over 15 years) and a total bilateral hip replacement 7 and 8 years ago, after minimal domestic trauma, reported a bilateral periprosthetic femoral fracture. Investigative results (Rx, histological examination biopsy) allowed us to classify these fractures as atypical. This case is consistent with the definition of bisphosphonate-related atypical femoral fracture around a well-fixed, total hip replacement. These findings challenge the current definition of atypical femoral fractures that excludes peri-prosthetic fractures.

KEYWORDS: *atypical femoral fracture, bisphosphonate, osteoporosis, bone, bone remodeling*

INTRODUCTION

Worldwide, osteoporosis causes more than 8.9 million fractures annually, resulting in an osteoporosis fracture every 3 seconds (1). Across Europe in 2019 (European Union, plus Switzerland & UK), 32 million individuals aged >50 are estimated to have osteoporosis, equivalent to 5.6% of the total European population aged >50, or approximately 25.5 million women (22.1% of women aged >50) and 6.5 million men (6.6% of men aged >50) (2).

Bisphosphonates are widely used in the treatment of osteoporosis to reduce fractures because they affect bone metabolism (3) and can cause necrosis of the jaws (4). Because of their long retention time in bone and uncommon side

Received: 4 October 2023
Accepted: 10 November 2023

effects, questions have been raised about the optimal duration of therapy. Prolonged bisphosphonate use has been related to the suppressing bone turnover, minimizing bone remodeling, and reducing bone healing capacity. This process will probably result in a frozen bone that is unable to repair the microcracks that may arise in the femoral shaft and could evolve into both incomplete and complete AFF. In fact, several retrospective studies have also suggested an association between bisphosphonate use and atypical femur fractures, although the results from larger observational studies are discordant (5, 6).

In a 2010 report from an international task force appointed by the American Society of Bone and Mineral Research to review this issue, major and minor criteria for atypical fractures were defined but did not include atypical periprosthetic femoral fractures (APFFs) (7).

Atypical femoral fractures usually occur at the subtrochanteric region or proximal to the mid-shaft of the femur because of high regional tensile stress. They are characterized by uni-cortical thickening (especially lateral) and transverse or short-oblique fracture patterns.

Because of the rarity of cases and limited awareness of this condition, to date, there are far fewer APFF studies than those on AFFs and typical periprosthetic femoral fractures (PFFs)

CASE REPORT

In November 2022, a 77-year-old woman presented with severe bilateral pain in both legs after minor trauma at home. She had been in treatment for over 15 years with bisphosphonates. In 2015 and 2016, she had undergone sequential bilateral Total Hip Arthroplasty for advanced osteoarthritis. Radiographs showed a Vancouver type-B1 periprosthetic fracture of both femurs (Fig. 1).

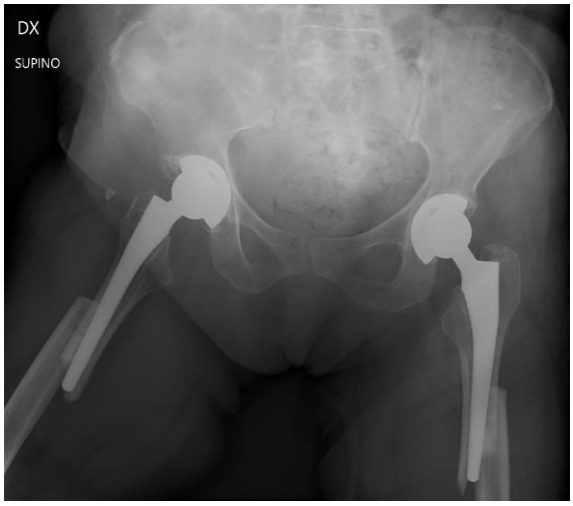


Fig. 1. *Vancouver type-B1 periprosthetic fracture of both femurs.*

A thickened femoral cortex or a ‘beak sign’ was noted bilaterally. The fractures were configured as an ‘inverted square root’, and the femoral stems were well fixed. Preoperatively, a laboratory workup was performed, including calcium, 25-OH vitamin D, bone alkaline phosphatase, and parathyroid hormone (PTH) levels.

Our patient underwent revision surgery involving the replacement of the femoral component with a revision of the long stem through an extended trochanteric osteotomy approach. At first, the procedure was performed only on one femur, after two days, the other side was operated on to allow for some recovery time between the two surgeries. A lateral approach to the hip, following the previous incision, was extended down to the knee. During the surgery, we could appreciate thickening of the cortex (Fig. 2).

Fig. 2. *Thickening of the femoral cortex.*

The fracture site was debrided, and a histological sample was taken. A revised long stem was then implanted (Fig. 3).

Fig. 3. *Postoperative X-ray.*

Intraoperative specimens were fixed in 4% formaldehyde, decalcified for 48 hours, and paraffin embedded. Tissue blocks were cut in 4mμ thick sections and stained with hematoxylin-eosin. The sample collected showed tissue characterized by marginal areas of necrotic bone with irregular borders, surrounded by active osteoclasts and lamellar bone with empty lacunae. The intertrabecular spaces were focally fibrotic, with scattered multinucleated osteoclast-like cells. In proximity to the rim of the fracture, some areas of bone remodeling were evident (Fig. 4).

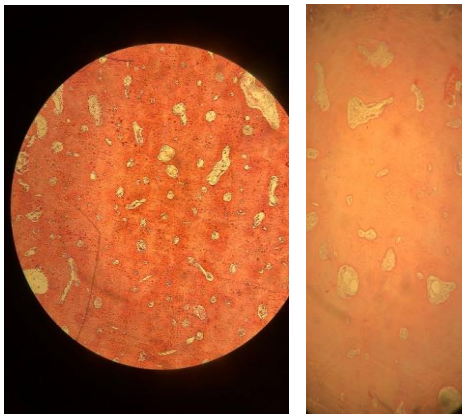


Fig. 4. *Scattered multinucleated osteoclast-like cells.*

Postoperatively, the patient maintained partial weight ambulation for 8 weeks and then advanced to full weight-bearing. In this case, calcium or vitamin D supplementation was not required. She regained the ability to walk without pain and any gait aid.

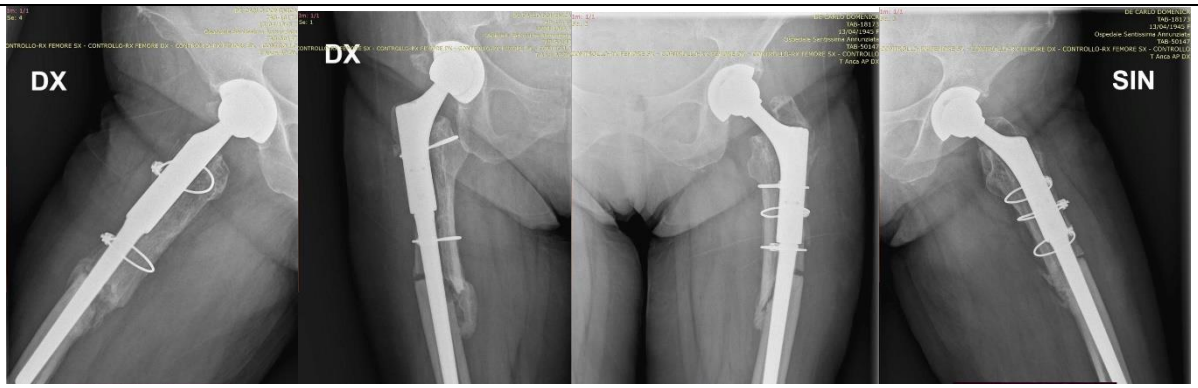


Fig. 5. *Post-operative X-rays (right and left).*

DISCUSSION

Epidemiology and diagnosis

As mentioned above, PFF is currently excluded from the definition of AFF based on the ASBMR task force reports (7, 8). According to the literature, there is a significant correlation between the use of bisphosphonates and atypical femoral fracture, particularly when the therapy is prolonged (> 5 years) (9, 10). These fractures currently represent a rare event; it is plausible that with the increase in annual performed THAs and the expanding indication for bisphosphonates use, the number of PFFs and, subsequently, APFFs could rise. It is crucial to diagnose an APFF, given that evidence supports a different type of approach and treatment for these patients.

In order to do that, an adequate collection of anamnestic data, with special attention to prior and current medications, is fundamental. It's also useful to investigate the mechanism of injury and possible occurrence of prodromal symptoms. An appropriate and early diagnosis of an APFF could also improve their outcomes. In fact, their treatment was more challenging, and their outcomes were worse than typical PFF because of the high rate of delayed healing, non-union, and fixation failure (11, 12).

Histological findings

The bilateral occurrence and delayed fracture healing support the hypothesis of an intrinsic bone deficiency over local stress factors. As different studies point out, bisphosphonates could lead to the formation of brittle hypermineralized bone that can suffer from low-impact stress. Indeed, drugs that decrease remodeling (i.e., antiresorptives) have been found to prevent bone healing through this mechanism. In a biopsy study by Miller and McCarthy (13), AFF patients had evidence of lower bone remodelling than expected, the rate increased with withdrawal of bisphosphonate therapy and commencement of teriparatide treatment.

Treatment strategies

Regarding treatment strategy, APFFs show poor fracture healing potential and require special attention. The management of atypical fractures is a big challenge, and the outcome is much poorer than that of typical fractures because of the delayed healing process, poor bone consolidation, difficulty of fracture fixation and high mortality rate (14, 15). Moreover, APFF union times were significantly longer than traditional AFF times (12). Indeed, fractures that were surgically treated took double the average time to union than those conservatively treated (16). Publications suggest that whenever identified, APFFs should be approached in a multidisciplinary way (16). Based on the radiographical pattern of the fracture, it is advisable to choose a surgical or conservative treatment: incomplete APFFs could benefit from both types, whereas complete ones require a surgical approach. Conservative treatment consists in avoiding weight bearing, in addition to medical management.

From a surgical point of view, treatment options for APFFs consist of fixation or revision. The choice between these approaches depends on different factors, as well as the surgeon's preference. Fixation consists of MIPO, a long-locked plate with or without cerclages, and structural graft. Adding a structural graft, granting the required construct stiffness and osteoconductive support for bone healing, provides both a mechanical and biological advantage. Otherwise, a more aggressive surgical approach could consist of a revision to a long stem. Medical management is based on clinical experience and a few uncontrolled studies (7, 8). Available options advocated by experts include the withdrawal of antiresorptive therapy, calcium, vitamin D supplementation, and consideration of anabolic therapy. In the USA, currently, the most utilized anabolic drug for osteoporosis is teriparatide, a protein consisting of the first 34 amino acids of

parathyroid hormone. There have been reports of some cases of healing following a daily subcutaneous injection of teriparatide in incomplete AFFs (13).

CONCLUSIONS

Accurate identification and diagnosis of APFFs are crucial for management planning and, eventually, the outcome. To this purpose, a precise medical anamnesis recollection of traumatic events and/or stressors is needed. It is also advisable to obtain a full laboratory workup, including calcium levels, 25-OH vitamin D, bone alkaline phosphatase, and parathyroid hormone (PTH) levels, since evidence has shown that, for these patients, oral supplementation or adjustment of blood levels could improve the outcome. Moreover, individualized preoperatory planning is important, also considering the option of conservative treatment for incomplete APFFs to reduce union time.

Furthermore, it has been shown to be beneficial to withdraw, where present, the antiresorptive therapy to restore the potential for bone remodeling and, therefore, strength. New research is also pointing forward to the use of anabolic drugs, of which teriparatide is the most widely known and used. Longer follow-ups of these patients would be useful in studying long-term outcomes.

Unfortunately, due to the rare occurrence of APFFs and the lack of consensus on their classification and treatment, further studies are definitely needed in order to define the optimal management option for this type of fracture. In this context, we hope to contribute with the presentation of this case report to the characterization of these clinical presentations and the deepening of our knowledge in this field.

REFERENCES

1. Johnell O, Kanis JA. An Estimate of the Worldwide Prevalence and Disability Associated with Osteoporotic Fractures. *Osteoporosis International*. 2006;17(12):1726-1733. doi:https://doi.org/10.1007/s00198-006-0172-4
2. Kanis JA, Norton N, Harvey NC, et al. SCOPE 2021: a new scorecard for osteoporosis in Europe. *Archives of Osteoporosis*. 2021;16(1). doi:https://doi.org/10.1007/s11657-020-00871-9
3. Scarano A, Crincoli V, Di Benedetto A, et al. Bone Regeneration Induced by Bone Porcine Block with Bone Marrow Stromal Stem Cells in a Minipig Model of Mandibular "Critical Size" Defect. *Stem Cells International*. 2017;2017:e9082869. doi:https://doi.org/10.1155/2017/9082869
4. Scarano A, Inchingolo F, Rapone B, Greco Lucchina A, Qorri E, Lorusso F. Role of Autologous Platelet Gel (APG) in Bone Healing: A Rabbit Study. *Applied Sciences*. 2021;11(1):395-395. doi:https://doi.org/10.3390/app11010395
5. Goh SK ., Yang KY, Koh JSB, et al. Subtrochanteric insufficiency fractures in patients on alendronate therapy. *The Journal of Bone and Joint Surgery British volume*. 2007;89-B(3):349-353. doi:https://doi.org/10.1302/0301-620x.89b3.18146
6. Lenart BA, Neviaser AS, Lyman S, et al. Association of low-energy femoral fractures with prolonged bisphosphonate use: a case control study. *Osteoporosis International*. 2008;20(8):1353-1362. doi:https://doi.org/10.1007/s00198-008-0805-x
7. Shane E, Burr D, Abrahamsen B, et al. Atypical Subtrochanteric and Diaphyseal Femoral Fractures: Second Report of a Task Force of the American Society for Bone and Mineral Research. *Journal of Bone and Mineral Research*. 2013;29(1):1-23. doi:https://doi.org/10.1002/jbmr.1998
8. Shane E, Burr D, Ebeling PR, et al. Atypical subtrochanteric and diaphyseal femoral fractures: Report of a task force of the american society for bone and mineral Research. *Journal of Bone and Mineral Research*. 2010;25(11):2267-2294. doi:https://doi.org/10.1002/jbmr.253
9. MacKenzie SA, Ng RT, Snowden G, Matilda, Duckworth AD, Chloe. Periprosthetic atypical femoral fractures exist and are associated with duration of bisphosphonate therapy. *The Bone & Joint Journal*. 2019;101-B(10):1285-1291. doi:https://doi.org/10.1302/0301-620x.101b10.bjj-2019-0599.r2
10. Koh A, Guerado E, Giannoudis PV. Atypical femoral fractures related to bisphosphonate treatment. *The Bone & Joint Journal*. 2017;99-B(3):295-302. doi:https://doi.org/10.1302/0301-620x.99b3.bjj-2016-0276.r2
11. Lee YK, Park CH, Kim KC, Hong SH, Ha YC, Koo KH. Frequency and associated factor of atypical periprosthetic femoral fracture after hip arthroplasty. *Injury*. 2018;49(12):2264-2268. doi:https://doi.org/10.1016/j.injury.2018.09.014
12. Robinson J de D, Leighton RK, Trask K, Bogdan Y, Tornetta P. Periprosthetic Atypical Femoral Fractures in Patients on Long-term Bisphosphonates. *Journal of Orthopaedic Trauma*. 2016;30(4):170-176. doi:https://doi.org/10.1097/bot.0000000000000508
13. Im GI, Lee SH. Effect of Teriparatide on Healing of Atypical Femoral Fractures: A Systemic Review. *Journal of Bone Metabolism*. 2015;22(4):183. doi:https://doi.org/10.11005/jbm.2015.22.4.183
14. Githens M, Garner MR, Firoozabadi R. Surgical Management of Atypical Femur Fractures Associated With Bisphosphonate Therapy. *Journal of the American Academy of Orthopaedic Surgeons*. 2018;26(24):864-871. doi:https://doi.org/10.5435/jaaos-d-16-00717

15. Lloyd AA, Gludovatz B, Riedel C, et al. Atypical fracture with long-term bisphosphonate therapy is associated with altered cortical composition and reduced fracture resistance. *Proceedings of the National Academy of Sciences of the United States of America*. 2017;114(33):8722-8727. doi:https://doi.org/10.1073/pnas.1704460114
16. Toro G, Ojeda-Thies C, Calabrò G, et al. Management of atypical femoral fracture: a scoping review and comprehensive algorithm. *BMC Musculoskeletal Disorders*. 2016;17(1). doi:https://doi.org/10.1186/s12891-016-1086-8

Review

NARRATIVE REVIEW ON POSITIONAL PLAGIOCEPHALY

E. Xhajanka[1], I.R. Bordea[2], P. Avantario[3], A.D. Inchingolo[3], A.M. Inchingolo[3], A. Laforgia[3], S.R. Tari[3], C. Bugea[3], F. Postiglione[3], F. Inchingolo[3] and G. Dipalma[3]

[1]Department of Dental Prosthesis, Medical University of Tirana, Rruga e Dibrës, U.M.T., Tirana, Albania
[2]Department of Oral Rehabilitation, Faculty of Dentistry, Iuliu Haţieganu University of Medicine and Pharmacy, Cluj-Napoca, Romania; bordea.ioana@umfcluj.com (I.R.B.)
[3]Department of Interdisciplinary Medicine, University of Bari "Aldo Moro", Bari, Italy

**Corrispondence to:*
Ioana Roxana Bordea, DDS
Department of Dental Prosthesis,
Medical University of Tirana,
Rruga e Dibrës, U.M.T.,
Tirana, Albania
e-mail: bordea.ioana@umfcluj.com

ABSTRACT

Plagiocephaly, characterized by asymmetrical skull distortion, affects infants and can manifest in various severities, impacting both aesthetics and cranial function. Historically linked to cultural practices like cradleboarding, modern medicine now focuses on its pathological aspects. Plagiocephaly is primarily categorized into synostotic plagiocephaly, caused by premature cranial suture fusion requiring surgical intervention, and positional plagiocephaly (PP), resulting from external pressures and managed through non-surgical methods. The prevalence of PP has increased since the 1990s due to the "Back to Sleep" campaign aimed at reducing sudden infant death syndrome, leading to prolonged supine positioning. PP affects approximately 20% of infants, with higher incidence in males, preterm infants, and those with limited neck mobility. Clinical significance extends beyond cosmetic concerns, potentially impacting cranial development, auditory and visual alignment, and cognitive development. Prevention involves maternal nutrition, safe sleeping practices, tummy time, repositioning, and parental education. Argenta's classification system categorizes PP severity, guiding intervention strategies from repositioning and physical therapy to helmet therapy and, rarely, surgical intervention. Challenges include parental awareness, cost, and adherence to preventive measures. Current research focuses on advanced diagnostic imaging, innovative treatments, and understanding long-term outcomes, emphasizing the importance of early detection and intervention for optimal cranial development.

KEYWORDS: *positional plagiocephaly, rigid mattress, flat head, semi-rigid mattress, infant well-being, infant cranial development, cranial deformation*

INTRODUCTION

Plagiocephaly, a condition characterized by an asymmetrical distortion of the skull, derives its name from the Greek words "plagios" (oblique) and "kefale" (head). This condition predominantly affects infants and can manifest in various forms and severities, impacting both the aesthetic and functional aspects of the cranial structure (1, 2). It is a condition that has been recognized and documented throughout medical history, with its clinical significance becoming increasingly prominent in recent decades due to changes in infant care practices. Historically, plagiocephaly was often attributed to cultural practices such as cradleboarding, where the infant's head was intentionally shaped by applying pressure through

Received: 19 October 2023
Accepted: 08 November 2023

binding or positioning. These cultural practices were widespread among various indigenous populations and were considered a norm for achieving desired cranial shapes. In modern medical practice, the focus has shifted to understanding the pathological aspects of plagiocephaly and its implications for infant health and development. Plagiocephaly can be broadly categorized into two primary forms: synostotic plagiocephaly and deformational or positional plagiocephaly (PP) (3-5).

- Synostotic plagiocephaly: This form of plagiocephaly is caused by the premature fusion of one or more cranial sutures, a condition known as craniosynostosis. The early fusion restricts the normal growth of the skull, leading to compensatory growth in other areas and resulting in an asymmetrical head shape. Synostotic plagiocephaly requires surgical intervention to correct the suture fusion and allow for normal cranial development (6-9).
- PP: this type occurs due to external pressures on the skull, typically from consistent positioning during sleep, limited movement, or other external forces. Unlike synostotic plagiocephaly, PP does not involve the premature fusion of cranial sutures and is generally managed through non-surgical interventions such as repositioning techniques, physical therapy, and, in some cases, orthotic devices (10-13). PP also can involve one or both sides of the occiput (Fig. 1).

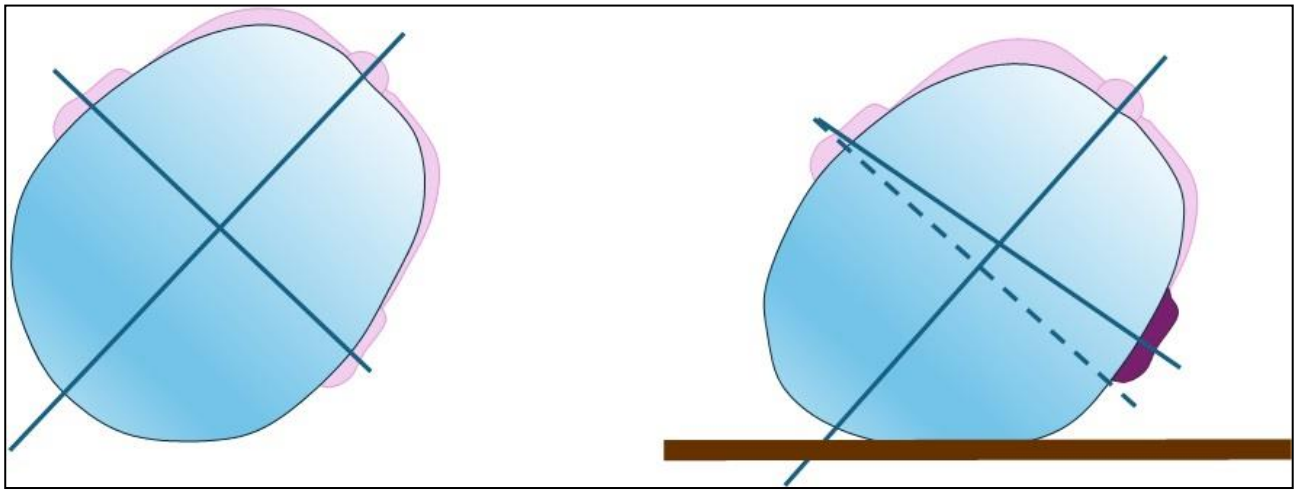


Fig. 1. *The pathogenetic mechanism of skull deformation.*

The prevalence of PP has significantly increased since the early 1990s, coinciding with the American Academy of Pediatrics' "Back to Sleep" campaign. This campaign, aimed at reducing the incidence of sudden infant death syndrome (SIDS), recommended that infants be placed on their backs to sleep. While this initiative successfully decreased SIDS rates, it inadvertently led to a rise in cases of PP due to prolonged supine positioning. Epidemiological studies suggest that PP affects nearly 20% of infants to varying degrees, making it a common concern for parents and healthcare providers. The condition tends to be more prevalent in male infants, preterm babies, and those with limited neck mobility, such as in cases of congenital muscular torticollis (14-18).

MATERIAL AND METHODS

The review was conducted following PRISMA, covering studies from January 2012 to October 2022 across six databases: PubMed, Web of Science, Google Scholar, Scopus, Cochrane Library, and ScienceDirect, focusing on English-language publications. The search used the Boolean keywords “positional plagiocephaly” and “prevention”. Inclusion criteria for the review were studies on babies and children aged 0 to 2 years, open access papers, and research on preventative interventions against PP, such as using a hard-surfaced mattress, lateralizing the infant during sleep, and tummy time activities. Two independent reviewers assessed the quality of the included studies based on selection criteria, outcome evaluation methods, and data analysis, with any disagreements resolved by a third researcher. The article reviews existing literature and studies on positional PP, including its categorization, prevalence, clinical significance, pathogenesis, risk factors, diagnosis, prevention strategies, and treatment interventions. It also discusses various preventive measures and their efficacy, challenges in prevention and treatment, and recent advancements in diagnostic

imaging and treatment approaches. The review includes analyzing data from epidemiological studies, clinical evaluations, and recent research trends in the field.

RESULTS

The initial search identified 530 articles, which were narrowed down to 416 after removing duplicates. After screening titles and abstracts, 247 publications were excluded, leaving 169 records. Following further eligibility assessment, 60 publications were found to be off-topic, resulting in 11 papers being included in the review for qualitative analysis.

DISCUSSION

The clinical significance of plagiocephaly extends beyond cosmetic concerns. While the aesthetic impact is often the primary concern for parents, there are potential functional implications that warrant medical attention. Severe cases of plagiocephaly can lead to asymmetries in the face and skull that may affect cranial development, auditory and visual alignment, and, in some instances, cognitive development. Early detection and intervention are crucial in mitigating these potential complications and promoting normal cranial growth and development (19, 20). Positional plagiocephaly develops due to continuous external forces exerted on an infant's malleable skull.

The increased prevalence of PP correlates with the "Back to Sleep" campaign by the American Academy of Pediatrics. While effective in reducing SIDS, this practice has led to an increase in PP cases, highlighting the need for balanced preventive strategies (21-23). The main risk factors for PP are male sex, multiple births, prematurity, assisted delivery, torticollis. Early diagnosis and intervention are crucial in managing PP. Pediatricians play a pivotal role in this process, employing a combination of clinical evaluation and diagnostic tools to identify and assess the severity of PP (24, 25). The clinical examination of an infant suspected of having positional PP begins with a visual assessment to identify any asymmetry in the head shape. Signs of PP may include flattening on one side of the occiput, misalignment of the ears, and, in severe cases, facial asymmetry. Following this, pediatricians perform palpation of the infant's skull to check for ridges along the cranial sutures, which could indicate craniosynostosis, a condition that needs to be differentiated from PP.

Measuring the cranial index (CI) and cranial vault asymmetry index (CVAI) with tools such as calipers or three-dimensional imaging provides a quantitative assessment of cranial asymmetry. Additionally, taking a thorough medical and family history helps to identify any genetic predispositions or family history of craniofacial conditions that might contribute to the development of PP (26). Effective prevention of positional PP involves a multifaceted approach that includes proper maternal nutrition, safe sleeping practices, and regular monitoring of the infant's head shape and positioning. Adequate intake of vitamin D and folic acid during pregnancy is essential for fetal bone and neural development. These nutrients help ensure the proper formation of the infant's skull and reduce the risk of deformities.

While placing infants on their backs to sleep remains a crucial measure to prevent SIDS, it is equally important to incorporate strategies that mitigate the risk of PP. Encouraging supervised tummy time during waking hours strengthens the neck, shoulder, and upper back muscles, promoting better head control and reducing the risk of positional flat spots. It is recommended to aim for at least 30 minutes of tummy time each day, gradually increasing as the infant grows. Regularly changing the infant’s head position during sleep and feeding can prevent constant pressure on one part of the skull. Techniques include alternating the direction the infant faces in the crib and varying feeding positions. Passive sleep curve mattresses are designed to distribute pressure evenly and support natural head shape development. These mattresses can be an effective preventive tool against PP. Educating parents and caregivers about the importance of repositioning, tummy time, and monitoring head shape is critical. Providing clear guidelines and practical tips can empower them to implement these strategies consistently (27).

Argenta’s classification system categorizes PP based on the severity and location of cranial asymmetry. Understanding this classification helps clinicians and caregivers tailor intervention strategies appropriately (27).

- type I: mild asymmetry restricted to the back of the skull. This is the most common and least severe form of PP, often resolving with simple repositioning techniques;
- type II: involves posterior cranial asymmetry with ear displacement but no frontal involvement. This type may require more active intervention, such as physical therapy or specialized pillows;
- type III: characterized by a parallelogram-shaped skull with frontal involvement. Infants with Type III PP may need more intensive therapies, including helmet therapy, to correct the asymmetry;

- type IV: includes facial asymmetry and more pronounced cranial deformity. This severity level often necessitates a combination of therapies and close monitoring to ensure effective correction;
- type V: the most severe form, featuring altered vertical skull growth and temporal area protrusion. Infants with type V PP typically require comprehensive treatment plans, including possible surgical intervention, to address the deformity (Fig. 2).

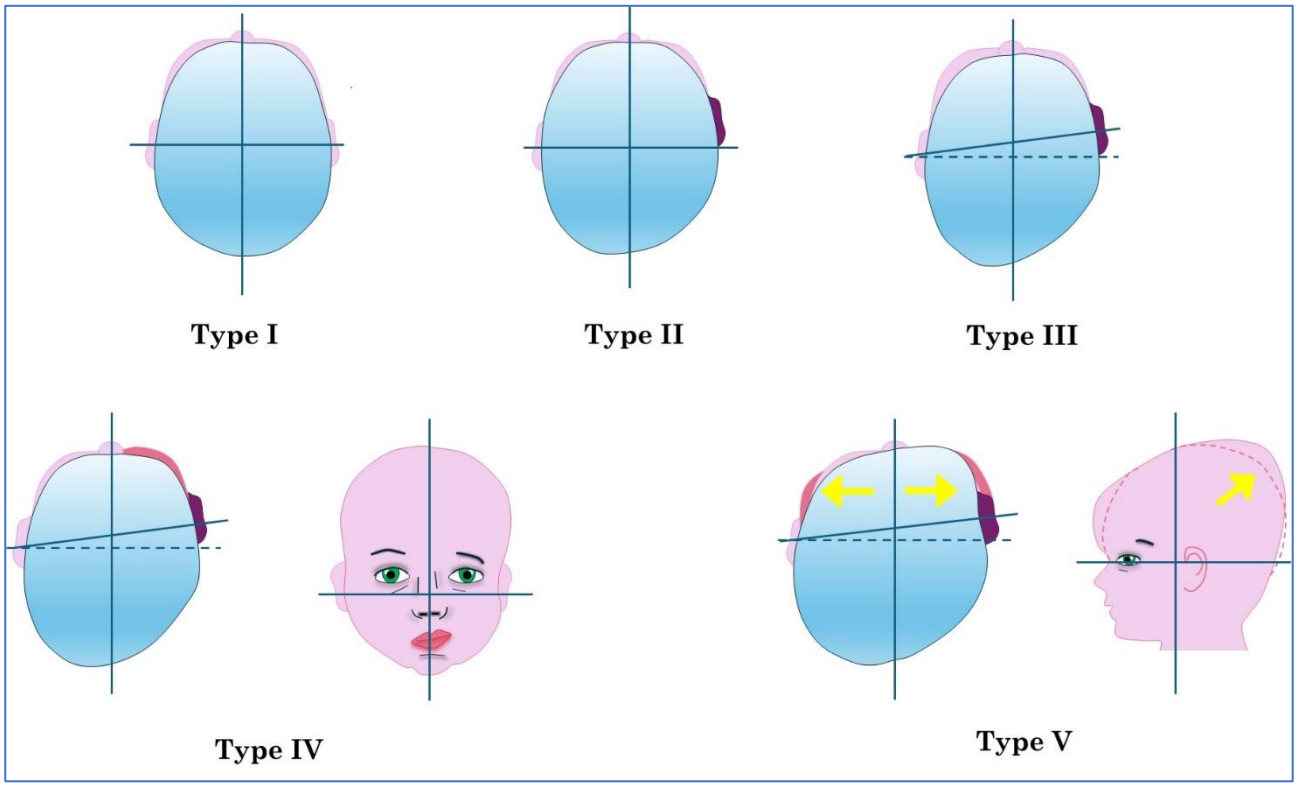


Fig. 2. *Classification into 5 types of plagiocephaly according to Argenta.*

Treatment strategies for positional PP vary depending on the severity of the condition and the infant's age, ranging from conservative measures to more intensive therapies. For mild to moderate cases of PP, repositioning techniques are often the primary strategy. This involves encouraging infants to turn their heads to the non-flattened side during sleep and play. Positional devices, such as wedges, rolls, or specially designed pillows, can help maintain the infant's head in the desired position (28). Additionally, environmental adjustments, like placing toys on the non-flattened side, encourage head turning in infants. Physical therapy improves neck muscle strength and head movement symmetry. Helmet therapy corrects moderate to severe PP with custom helmets despite potential discomfort, skin reactions, and social stigma (29-33). In severe cases, cranial vault remodeling surgery corrects asymmetry and ensures proper skull growth, especially in infants with craniosynostosis (32, 34–37).

Recent studies have shed light on the effectiveness of various preventive measures for positional PP, providing valuable insights into the most effective strategies for reducing its incidence and severity. Proper maternal nutrition, especially vitamin D and folic acid intake, lowers the incidence of PP by promoting fetal bone development and a stronger skull. Passive sleep curve mattresses effectively prevent and correct PP by evenly distributing pressure on the skull. Regular tummy time and repositioning techniques also prevent PP by strengthening neck and upper body muscles and reducing positional flat spots. However, challenges include ensuring parents and caregivers are informed about these strategies and overcoming the prohibitive cost of orthotic devices and specialized mattresses for some families. Educating parents and making interventions accessible to all infants are crucial for the effective prevention and treatment of PP (38). Consistent implementation of preventive measures by caregivers, including regular tummy time, repositioning techniques, and follow-up appointments with healthcare providers, is crucial for their effectiveness. Supporting and encouraging parents to follow these recommendations improves outcomes for infants with PP. Recent research focuses on better diagnostic methods, new treatment strategies, and understanding long-term outcomes (39, 40). New imaging technologies, like three - dimensional (3D) surface scanning and advanced ultrasound techniques, offer more accurate and non-invasive methods for diagnosing and monitoring PP, enabling earlier detection and timely intervention. Researchers are exploring

innovative treatment approaches, including custom-made orthotic devices and non-invasive therapies. Advances in 3D printing technology enable the creation of custom-made helmets and cranial orthoses tailored to the specific needs of each infant, offering improved comfort and effectiveness in correcting skull asymmetry. Studies are also investigating non-invasive therapies, such as low-level laser therapy, to promote skull growth and reduce asymmetry (41–44). Understanding the long-term outcomes of infants with PP is a key area of research. Longitudinal studies follow affected infants into childhood and adolescence to assess the impact of PP on cognitive development, academic performance, and psychosocial well-being (45). These studies help identify potential long-term effects and inform strategies for ongoing support and intervention (46, 47).

CONCLUSIONS

The increasing prevalence of positional PP underscores the need for effective prevention and intervention strategies. Pediatricians, parents, and caregivers must work together to ensure early detection and treatment. Simple, cost-effective measures, such as supervised tummy time, repositioning techniques, and the use of passive sleep curve mattresses, can significantly reduce the incidence and severity of PP. Early intervention and consistent monitoring are key to managing this condition and promoting optimal cranial development in infants. The type of mattress used is essential to prevent skull deformity resulting from plagiocephaly. The use of an orthopedic mattress is a modern aspect of PP management. As a result, pressure on the surface where the baby's skull might normally grow, directed by the expanding brain, could be better distributed.

Unlike other methods that rely on repositioning guidance or helmet orthotics, a sleep curve mattress is less expensive and does not require your child's compliance. From the study conducted by the Sant'Orsola hospital in Bologna, the Inglesina Welcome Pad® mattress, compared to other mattresses, facilitates the comfortable maintenance of the supine position and the alignment of the spine, promoting correct breathing of the child. Emerging research is exploring the role of genetic and epigenetic factors in the development of PP. Identifying genetic predispositions and understanding how environmental factors influence gene expression can provide insights into the underlying mechanisms of PP and guide the development of targeted prevention and treatment strategies.

Conflicts of Interest

The authors declare no conflicts of interest.

REFERENCES

1. Adina SS, Dipalma G, Bordea IR, et al. Orthopedic joint stability influences growth and maxillary development: clinical aspects. *PubMed*. 2020;34(3):747-756. doi:https://doi.org/10.23812/20-204-e-52
2. Aihara Y, Komatsu K, Dairoku H, Kubo O, Hori T, Okada Y. Cranial molding helmet therapy and establishment of practical criteria for management in Asian infant positional head deformity. *Child's Nervous System*. 2014;30(9):1499-1509. doi:https://doi.org/10.1007/s00381-014-2471-y
3. Bhatt RS, Hock A, White H, Jubran R, Galati A. The Development of Body Structure Knowledge in Infancy. *Child Development Perspectives*. 2016;10(1):45-52. doi:https://doi.org/10.1111/cdep.12162
4. Beuriat PA ., Szathmari A, Di Rocco F, Mottolese C. Deformational plagiocephaly: State of the art and review of the literature. *Neurochirurgie*. 2019;65(5):322-329. doi:https://doi.org/10.1016/j.neuchi.2019.09.003
5. Bialocerkowski AE, Vladusic SL, Wei Ng C. Prevalence, risk factors, and natural history of positional plagiocephaly: a systematic review. *Developmental Medicine & Child Neurology*. 2008;50(8):577-586. doi:https://doi.org/10.1111/j.1469-8749.2008.03029.x
6. Boere-Boonekamp MM, van der Linden-Kuiper TA, van Es P. [Preferential posture in infants; serious demands on health care]. *Ned Tijdschr Geneeskd* 1997;141(16):769-772.
7. Bridges SJ, Chambers, TL, Pople IK. Plagiocephaly and head binding. *Archives of Disease in Childhood*. 2002;86(3):144-145. doi:https://doi.org/10.1136/adc.86.3.144
8. Bruneteau RJ, Mulliken JB. Frontal Plagiocephaly: Synostotic, Compensational, or Deformational. *Plastic and Reconstructive Surgery*. 1992;89(1):21-31. doi:https://doi.org/10.1097/00006534-199289010-00005
9. Aarnivala H, Vuollo V, Harila V, et al. The course of positional cranial deformation from 3 to 12 months of age and associated risk factors: a follow-up with 3D imaging. *Eur J Pediatr*. 2016;175(12):1893-1903. doi:https://doi.org/10.1007/s00431-016-2773-z
10. Falguera-Uceda MI, Sanchez-Casanova S, Escudero-Duch C, Vilaboa N. A Narrative Review of Cell-Based Approaches for Cranial Bone Regeneration. *Pharmaceutics*. 2022;14(1):132-132. doi:https://doi.org/10.3390/pharmaceutics14010132
11. Cabrera-Martos I, Valenza MC, Benítez-Feliponi A, Robles-Vizcaíno C, Ruiz-Extremera A, Valenza-Demet G.

Clinical profile and evolution of infants with deformational plagiocephaly included in a conservative treatment program. *Child's Nervous System*. 2013;29(10):1893-1898. doi:https://doi.org/10.1007/s00381-013-2120-x
12. Cantore S, Ballini A, De Vito D, et al. Characterization of human apical papilla-derived stem cells. *PubMed*. 2017;31(4):901-910.
13. Ann HR, Jung YS, Lee KJ, Baik HS. Evaluation of stability after pre-orthodontic orthognathic surgery using cone-beam computed tomography: A comparison with conventional treatment. *The Korean Journal of Orthodontics*. 2016;46(5):301. doi:https://doi.org/10.4041/kjod.2016.46.5.301
14. COLLETT B, BREIGER D, KING D, CUNNINGHAM M, SPELTZ M. Neurodevelopmental Implications of "Deformational" Plagiocephaly. *Journal of Developmental & Behavioral Pediatrics*. 2005;26(5):379-389. doi:https://doi.org/10.1097/00004703-200510000-00008
15. Dassios T, Vervenioti A, Dimitriou G. Respiratory muscle function in the newborn: a narrative review. *Pediatric Research*. 2021;91(1):1-9. doi:https://doi.org/10.1038/s41390-021-01529-z
16. Dec W, Warren SM. Current Concepts in Deformational Plagiocephaly. *Journal of Craniofacial Surgery*. 2011;22(1):6-8. doi:https://doi.org/10.1097/scs.0b013e3182074e04
17. Di Chiara A, La Rosa E, Ramieri V, Vellone V, Cascone P. Treatment of Deformational Plagiocephaly With Physiotherapy. *Journal of Craniofacial Surgery*. 2019;30(7):2008-2013. doi:https://doi.org/10.1097/scs.0000000000005665
18. Losee JE, Corde Mason A. Deformational plagiocephaly: diagnosis, prevention, and treatment. *Clinics in Plastic Surgery*. 2005;32(1):53-64. doi:https://doi.org/10.1016/j.cps.2004.08.003
19. Goh JL, Bauer DF, Durham SR, Stotland MA. Orthotic (helmet) therapy in the treatment of plagiocephaly. *Neurosurgical Focus*. 2013;35(4):E2. doi:https://doi.org/10.3171/2013.7.focus13260
20. Margulis A, Hatuel, V, Weinberg A, Neuman, A, Israel Z, Wexler MR. [Plagiocephaly in children: etiology, differential diagnosis and helmet treatment]. *Harefuah*. 2024;136(7).
21. DeGrazia M, Giambanco D, Hamn G, Ditzel A, Tucker L, Gauvreau K. Prevention of Deformational Plagiocephaly in Hospitalized Infants Using a New Orthotic Device. *Journal of Obstetric, Gynecologic & Neonatal Nursing*. 2015;44(1):28-41. doi:https://doi.org/10.1111/1552-6909.12523
22. Priyadarshi M, Balachander B, Sankar MJ. Effect of sleep position in term healthy newborns on sudden infant death syndrome and other infant outcomes: A systematic review. *Journal of Global Health*. 2022;12. doi:https://doi.org/10.7189/jogh.12.12001
23. Hutchison BL, Thompson JMD, Mitchell EA. Determinants of Nonsynostotic Plagiocephaly: A Case-Control Study. *PEDIATRICS*. 2003;112(4):e316-e316. doi:https://doi.org/10.1542/peds.112.4.e316
24. Enlow DH, Moyers RE, Merow WW. *Handbook of Facial Growth*. W.B. Saunders Company; 1982.
25. Filisetti M, Cattarelli D, Bonomi S. Positional plagiocephaly from structure to function: Clinical experience of the service of pediatric osteopathy in Italy. *Early Human Development*. 2020;146:105028. doi:https://doi.org/10.1016/j.earlhumdev.2020.105028
26. González MJ, Schmitz KJ, Matos MI, López D, Rodríguez JR, Gorrín JJ. Folate supplementation and neural tube defects: a review of a public health issue. *Puerto Rico health sciences journal*. 1997;16(4):387-393.
27. Haas-Lude K, Wolff M, Will B, Bender B, Krimmel M. Clinical and imaging findings in children with non-syndromic lambdoid synostosis. *European Journal of Pediatrics*. 2013;173(4):435-440. doi:https://doi.org/10.1007/s00431-013-2186-1
28. Hutchison BL, Stewart AW, De Chalain TB, Mitchell EA. A randomized controlled trial of positioning treatments in infants with positional head shape deformities. *Acta Paediatrica*. 2010;99(10):1556-1560. doi:https://doi.org/10.1111/j.1651-2227.2010.01872.x
29. Mercuri E, Guzzetta A, Laroche S, et al. Neurologic examination of preterm infants at term age: Comparison with term infants. *The Journal of Pediatrics*. 2003;142(6):647-655. doi:https://doi.org/10.1067/mpd.2003.215
30. Michels A, El Bakkali N, Bastiaenen CH, de Bie RA, Colla CG, van. Periconceptional Folic Acid Use and the Prevalence of Positional Plagiocephaly. *Journal of Craniofacial Surgery*. 2008;19(1):37-39. doi:https://doi.org/10.1097/scs.0b013e31815c8a87
31. Michels AC, Van Den Elzen ME, Vies JSH, Van Der Hulst RR. Positional Plagiocephaly and Excessive Folic Acid Intake during Pregnancy. *The Cleft Palate-Craniofacial Journal*. 2012;49(1):1-4. doi:https://doi.org/10.1597/09-096
32. MIYABAYASHI H, NAGANO N, KATO R, et al. Reference Values for Cranial Morphology Based on Three-dimensional Scan Analysis in 1-month-old Healthy Infants in Japan. *Neurologia medico-chirurgica*. 2022;62(5):246-253. doi:https://doi.org/10.2176/jns-nmc.2021-0384
33. McGarry A, Dixon MT, Greig RJ, Hamilton DRL, Sexton S, Smart H. Head shape measurement standards and cranial orthoses in the treatment of infants with deformational plagiocephaly. *Developmental Medicine & Child Neurology*. 2008;50(8):568-576. doi:https://doi.org/10.1111/j.1469-8749.2008.03017.x
34. Moss ML. Twenty years of functional cranial analysis. *American Journal of Orthodontics*. 1972;61(5):479-485. doi:https://doi.org/10.1016/0002-9416(72)90152-2
35. Mulliken JB, Vander Woude DL, Hansen M, LaBrie RA, Scott MR, Mulliken JB. Analysis of Posterior

Plagiocephaly: Deformational versus Synostotic. *Plastic and Reconstructive Surgery*. 1999;103(2):371-380. doi:https://doi.org/10.1097/00006534-199902000-00003

36. Najarian SP. Infant cranial molding deformation and sleep position: Implications for primary care. *Journal of Pediatric Health Care*. 1999;13(4):173-177. doi:https://doi.org/10.1016/s0891-5245(99)90036-6
37. Nieberle F, Spoerl S, Lottner LM, et al. Direct Anthropometry Overestimates Cranial Asymmetry—3D Digital Photography Proves to Be a Reliable Alternative. *Diagnostics*. 2023;13(10):1707-1707. doi:https://doi.org/10.3390/diagnostics13101707
38. Knorr A, Giambanco D, Staude MV, et al. Feasibility and Safety of the Preemie Orthotic Device to Manage Deformational Plagiocephaly in Extremely Low Birth Weight Infants. *Advances in Neonatal Care*. 2019;19(3):226-235. doi:https://doi.org/10.1097/anc.0000000000000585
39. Smartt JM, Reid RR, Singh DJ, Bartlett SP. True Lambdoid Craniosynostosis: Long-Term Results of Surgical and Conservative Therapy. *Plastic and Reconstructive Surgery*. 2007;120(4):993-1003. doi:https://doi.org/10.1097/01.prs.0000278043.28952.e8
40. Robinson S, Proctor M. Diagnosis and management of deformational plagiocephaly. *Journal of Neurosurgery: Pediatrics*. 2009;3(4):284-295. doi:https://doi.org/10.3171/2009.1.peds08330
41. Turk AE, McCarthy JG, Thorne CH, Wisoff JH. The “back to sleep campaign” and deformational plagiocephaly: is there cause for concern?. *The Journal of Craniofacial Surgery*. 1996;7(1):12-18. doi:https://doi.org/10.1097/00001665-199601000-00006
42. van Adrichem LNA, van Vlimmeren LA, Čadanová D, et al. Validation of a Simple Method for Measuring Cranial Deformities (Plagiocephalometry). *Journal of Craniofacial Surgery*. 2008;19(1):15-21. doi:https://doi.org/10.1097/scs0b013e31815c93cb
43. Kahle P, Gallus M, Kierdorf H, Kierdorf U. Age estimation in the harbour seal (*Phoca vitulina*) based on the closure of skull sutures and synchondroses. *Anatomia Histologia Embryologia*. 2022;52(2):300-311. doi:https://doi.org/10.1111/ahe.12884
44. Kajita H, Sakamoto Y, Takenouchi T, Miwa T, Takahashi T. Is that cranial deformity really due to sleeping position? *Pediatrics International*. 2017;59(4):494-496. doi:https://doi.org/10.1111/ped.13216
45. Peitsch WK, Keefer CH, LaBrie RA, Mulliken JB. Incidence of Cranial Asymmetry in Healthy Newborns. *PEDIATRICS*. 2002;110(6):e72-e72. doi:https://doi.org/10.1542/peds.110.6.e72
46. Wilbrand JF, Wilbrand M, Malik CY, et al. Complications in helmet therapy. *Journal of Cranio-Maxillofacial Surgery*. 2012;40(4):341-346. doi:https://doi.org/10.1016/j.jcms.2011.05.007
47. Wolfson MR, Greenspan JS, Deoras KS, Allen JL, Shaffer TH. Effect of position on the mechanical interaction between the rib cage and abdomen in preterm infants. *Journal of Applied Physiology*. 1992;72(3):1032-1038. doi:https://doi.org/10.1152/jappl.1992.72.3.1032