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High-Dose Melatonin in Fracture Healing: A Comprehensive Narrative Review

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Amendments - This protocol was registered with the International Platform of Registered Systematic Review and Meta-Analysis Protocols (INPLASY) on 3 September 2025 and was last updated on 30 September 2025.

INTRODUCTION

Review question / Objective This comprehensive narrative review critically evaluates current evidence regarding high-dose melatonin (≥30 mg/day) for enhancing fracture healing, addressing molecular mechanisms, preclinical efficacy, clinical translation challenges, and implications for orthopedic practice. The primary objective is to synthesize available literature examining melatonin's therapeutic potential in bone regeneration, with emphasis on pharmacological dosing strategies that may overcome limitations of physiological concentrations. Secondary objectives include identifying translational barriers from preclinical to clinical application, evaluating safety profiles of high-dose protocols, and characterizing advanced delivery systems that may optimize therapeutic outcomes.

Rationale Despite robust preclinical evidence demonstrating melatonin's osteogenic properties, clinical translation remains limited due to poor oral bioavailability (10-15%) and rapid clearance (half-life 30-60 minutes). Most clinical research has focused on low-dose regimens (1-10 mg/day), which may not achieve therapeutic tissue concentrations necessary for meaningful MT2 receptor occupancy and osteogenic effects. Delayed union and nonunion affect 5-19% of fractures, particularly in tibial fractures, creating substantial socioeconomic burdens. High-dose melatonin protocols (≥30 mg/day) show promise in preclinical models but lack systematic evaluation in orthopedic populations, representing a significant knowledge gap requiring comprehensive synthesis.

Condition being studied Fracture healing, bone regeneration, orthopedic trauma, delayed union, nonunion, osteoporosis-related fractures, age-

related bone healing impairment, exercise-induced bone damage, medication-induced bone healing inhibition (NSAIDs, corticosteroids, opioids), radiation-induced bone injury, compromised bone healing in elderly populations, senescence-associated bone deterioration.

METHODS

Search strategy Systematic literature search across PubMed/MEDLINE, Web of Science, Scopus, and Cochrane Library (January 2020-September 2025) using Boolean operators: ("melatonin" OR "N-acetyl-5-methoxytryptamine") AND ("fracture healing" OR "bone regeneration" OR "bone repair" OR "osteogenesis" OR "bone formation" OR "callus formation") AND ("high dose" OR "pharmacological dose" OR "therapeutic dose" OR "supraphysiological"). Additional MeSH terms included MT1/MT2 receptors, RANK/RANKL/OPG axis, antioxidant properties, osteoblast differentiation. Reference lists and forward citation tracking supplemented primary searches.

Participant or population Human patients with acute fractures (tibial, femoral, humeral, others), elderly individuals with age-related bone healing impairment, athletes with exercise-induced bone stress, patients with compromised bone healing (diabetes, osteoporosis, medication-induced), orthopedic trauma populations, individuals with delayed union or nonunion. Animal models including rodents (rats, mice), rabbits, and other species used in bone healing research. In vitro studies utilizing mesenchymal stem cells, osteoblasts, osteoclasts, bone marrow-derived cells, and bone tissue cultures.

Intervention High-dose melatonin supplementation (≥30 mg/day), pharmacological melatonin protocols, exogenous melatonin administration via oral, parenteral, topical, or local delivery routes. Advanced delivery systems including melatonin-loaded hydrogels, electrospun nanofibers, biodegradable microspheres, silk fibroin nanofibers, three-dimensional bioprinted polymers. Combination therapies incorporating melatonin with bone morphogenetic proteins, growth factors, antioxidant compounds, or stem cell therapies. Various dosing regimens from single-dose protocols to extended treatment periods.

Comparator Placebo controls, low-dose melatonin (<30 mg/day), physiological melatonin concentrations (<10 mg/day), standard orthopedic care, untreated control groups, alternative bone healing interventions (bone morphogenetic proteins, parathyroid hormone, bisphosphonates), other antioxidant compounds, conventional fracture management protocols, sham treatments in animal models.

Study designs to be included Randomized controlled trials, non-randomized controlled trials, observational studies (cohort, case-control), preclinical investigations (in vivo animal studies, in vitro cell culture studies), systematic reviews, meta-analyses, narrative reviews addressing high-dose melatonin protocols, case series with adequate sample sizes (≥10 participants), pilot studies evaluating safety and feasibility of high-dose regimens.

Eligibility criteria Inclusion: Original research evaluating melatonin's effects on bone healing processes, English language publications, studies focusing on high-dose regimens (≥30 mg/day) or mechanistic investigations relevant to pharmacological doses, contemporary publications (2020-2025) ensuring current relevance, adequate methodology and clear outcome reporting. Exclusion: Studies utilizing exclusively non-bone tissue models, investigations limited to physiological dosages without mechanistic relevance, duplicate publications, letters to editors, conference abstracts lacking complete datasets, studies with inadequate methodology or unclear outcome measures.

Information sources PubMed/MEDLINE (primary database for biomedical literature), Web of Science (multidisciplinary citation database), Scopus (comprehensive research database), Cochrane Library (systematic reviews and clinical trials), reference lists of included studies, forward citation tracking, grey literature searches including thesis databases, conference proceedings, regulatory agency reports, clinical trial registries (ClinicalTrials.gov, ISRCTN).

Main outcome(s) Bone formation markers (alkaline phosphatase, osteocalcin, RUNX2 expression), radiographic healing assessments (RUST scores, cortical bridging, callus formation), biomechanical strength parameters (ultimate load, stiffness, energy absorption), histological measures (bone volume fraction, trabecular architecture, osteoblast/osteoclast counts), molecular targets (MT1/MT2 receptor activation, Wnt/β-catenin signaling, BMP pathway modulation), safety profiles and adverse events documentation.

Additional outcome(s) Antioxidant enzyme activities (SOD, catalase, glutathione peroxidase),

inflammatory markers (TNF-α, IL-1β, IL-6, NF-κB expression), angiogenesis indicators (VEGF expression, vascular density), bone turnover markers (CTX, P1NP), DNA damage assessment (comet assay), cellular senescence markers (SASP factors), pharmacokinetic parameters (bioavailability, clearance, tissue concentrations), functional outcomes (pain scores, quality of life measures), treatment compliance and tolerability.

Data management Standardized data extraction forms capturing study characteristics, participant demographics, intervention protocols, outcome measures, and key findings. Two independent reviewers will perform data extraction with discrepancies resolved through consensus discussion. Data will be organized using reference management software with version control and secure backup systems. Quality control measures include cross-verification of extracted data and regular team meetings to ensure consistency.

Quality assessment / Risk of bias analysis Study quality assessment using appropriate validated tools: Cochrane Risk of Bias tool for randomized controlled trials, Newcastle-Ottawa Scale for observational studies, SYRCLE's risk of bias tool for animal studies. Assessment domains include randomization procedures, allocation concealment, blinding of participants and outcome assessors, incomplete outcome data, selective reporting, and other potential sources of bias. Quality ratings will inform evidence synthesis and interpretation of findings.

Strategy of data synthesis Narrative synthesis approach organizing evidence according to thematic categories: molecular mechanisms, preclinical efficacy, clinical translation challenges, pharmacokinetic barriers, advanced delivery systems, and future research directions. Emphasis on identifying knowledge gaps and translational research opportunities. Evidence tables will summarize study characteristics and findings. Where appropriate, qualitative synthesis will explore patterns, relationships, and contradictions across studies while acknowledging heterogeneity in study designs, populations, and interventions.

Subgroup analysis Subgroup analyses planned for: dosing regimens (30-50 mg vs >50 mg/day), delivery routes (oral vs parenteral vs local), patient populations (elderly vs younger adults, compromised vs normal healing), fracture types (appendicular vs axial skeleton), study designs (RCTs vs observational studies), duration of treatment (acute vs chronic protocols), species in animal studies (rodents vs larger animals).

Sensitivity analysis Sensitivity analyses will examine the robustness of findings by: excluding studies with high risk of bias, analyzing only randomized controlled trials versus all study designs, excluding studies with small sample sizes (<20 participants), examining effects of different outcome measurement methods, assessing impact of industry funding versus independent research, evaluating geographical variations in research findings.

Language restriction English language publications only, due to resource limitations for translation and to ensure accurate interpretation of complex scientific terminology. This limitation will be acknowledged as a potential source of publication.

Country(ies) involved Brazil.

Other relevant information The dissemination strategy for this high-dose melatonin pilot study targets multiple stakeholders through academic publication in orthopedic journals, presentations at international conferences, and engagement with practice guideline committees.

Keywords bone healing; fracture; melatonin; osteogenesis; pharmacokinetics; tissue regeneration; high-dose therapy; MT2 receptor; antioxidant; anti-inflammatory; RANK/RANKL/OPG; bone formation; callus; biomechanical strength.

Dissemination plans Development of clinical practice guidelines or recommendations for orthopedic practitioners. Creation of educational materials for medical students and residents. Collaboration with professional organizations to disseminate findings through newsletters and webinars. Social media engagement through professional networks to reach broader clinical and research communities.

Contributions of each author

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