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Review

The Subacromial Bursa in Rotator Cuff Disease: Biological Insights From Preclinical *In Vitro* and *In Vivo* Studies—A Systematic Review

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Abstract

Background: The subacromial bursa (SAB) is traditionally viewed as a source of inflammation in rotator cuff (RC) disease, often leading to routine bursectomy; recent evidence suggests its potential as a biological resource. This systematic review aims to evaluate the biological properties of SAB and its role in RC pathophysiology and repair by synthesizing evidence from preclinical *in vitro* and *in vivo* studies. **Methods:** A systematic search was conducted across PubMed, Scopus, and Web of Science (January 2015–December 2026) following Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines. *In vitro* and *in vivo* preclinical studies investigating SAB were included. Main characteristics and findings of each included article were extracted. Risk of bias (RoB) was assessed using the Systematic Review Centre for Laboratory animal Experimentation (SYRCLE) tools. **Results:** Twenty-seven studies were included (21 *in vitro*, 3 *in vivo*, 3 combined). *In vitro* studies confirmed that SAB-derived cells possess mesenchymal stem cell-like properties, including high proliferative capacity and multilineage differentiation potential, often superior to bone marrow-derived cells. SAB cells demonstrated "biological plasticity", responding to anabolic stimuli and pharmacological agents. *In vivo* models suggested that SAB preservation or cell transplantation facilitates histological and mechanical maturation of repaired tendons. **Conclusions:** From preclinical evidence, SAB emerged as a biologically active tissue that responds to inflammation and serves as a source of mesenchymal progenitor cells and bioactive factors, with the potential to enhance tendon healing. However, the available preclinical evidence is limited by methodological weaknesses and risks of bias. High-quality studies are needed to confirm efficacy and optimal indications its application in RC repair.

Keywords: Subacromial bursa, shoulder, preclinical studies, systematic review.

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Introduction

Rotator cuff (RC) disorders represent one of the most common causes of shoulder pain and functional impairment, affecting 2.4–14% of the general working-age population with some evidence suggesting greater prevalence with increasing age [1]. Shoulder pain has been reported to affect up to 20–30% of individuals over their lifetime, with RC pathology representing the most frequent underlying diagnosis [2]. The leading causes of shoulder pain include tendinopathy and partial or full-thickness tears [3]. Epidemiological studies have shown a significant increase in the prevalence of RC tears with age [4], impacting approximately 25% of individuals in their 60s years old and about 45% of those in their 70s years old [5]. Adhesive capsulitis, also known as frozen shoulder, affects approximately 2–5% of the population, typically between 40 and 60 years of age, with a higher prevalence in women and patients with metabolic comorbidities [6,7]. Glenohumeral instability is less common but predominantly affects young active males following traumatic events [8]. Subacromial impingement syndrome is a major contributor to chronic shoulder pain and is often associated with RC defects [9]. Glenohumeral osteoarthritis, a less prevalent condition, is nevertheless a significant cause of pain and disability in the elderly [10]. The RC anatomy combines four muscles—supraspinatus, infraspinatus, teres minor, and subscapularis—and their corresponding tendons for connecting to bones. Together, these muscles stabilize the glenohumeral joint and facilitate coordinated shoulder movement. The dynamic stabilizing action of these muscles is crucial for maintaining the

humeral head centered within the glenoid fossa during arm elevation and rotation. Due to the complex biomechanical environment of the shoulder and the high mechanical loads experienced by the RC tendons, this anatomical unit is particularly susceptible to degenerative changes and structural failure [11]. Within the subacromial space, several peritendinous structures may influence both the pathogenesis and healing of RC disease. Among these, the subacromial bursa (SAB) has historically been considered primarily as a lubricating synovial structure that facilitates smooth gliding. Anatomically, the SAB lies between the acromion, the coracoacromial ligament, the deltoid muscle, and the RC tendons, where it plays an important role in reducing friction during shoulder movement. The structure consists of an inner synovial membrane containing synoviocytes and a vascularized subintimal layer and is surrounded by fibrous connective tissue. Although traditionally considered a passive mechanical interface and a source of inflammation, it is now increasingly recognized as a metabolically active and biologically dynamic tissue due to its ability to release cytokines, growth factors, bioactive mediators and mesenchymal stem cells (MSCs). The SAB exerts paracrine effects on surrounding tissues. It contributes to the regulation of the local inflammatory and regenerative environment. In the presence of RC pathology, however, the SAB frequently undergoes structural and biological changes.

Histological studies have demonstrated synovial hypertrophy, increased vascularity, and infiltration of inflammatory cells within the bursal tissue. Molecular analyses have further revealed increased expression of inflammatory mediators and cytokines associated with RC degeneration and subacromial pain syndromes [12]. For many years, surgeons have regarded the SAB as a pain-modulator tissue because of these inflammatory characteristics. To date, bursectomy has become a standard component of arthroscopic RC repair procedures, to enhance visualization of the surgical area and to remove potentially inflammatory tissue from the subacromial space [13]. In recent years, however, the perception of the SAB has changed considerably. Emerging evidence has demonstrated that this tissue contains a substantial population of progenitor cells and MSCs with significant proliferative and differentiation potential [14,15].

These findings have led to a new understanding of the SAB, shifting the conventional view of it as mere surgical waste to its potential as a valuable biological resource with the capacity to facilitate tendon regeneration [16–18]. Several authors have proposed the preservation, mobilization, or reimplantation of bursal tissue as a biological augmentation strategy during arthroscopic RC repair [19].

Given the growing body of literature investigating this debated topic, a comprehensive evaluation of the available preclinical evidence is warranted. Therefore, the purpose of this systematic review is to evaluate and synthesize the current evidence regarding the role of SAB in shoulder and RC pathology, including *in vitro* and *in vivo* studies. By systematically analyzing the available literature, this review aims to better characterize the biological properties and to clarify its potential role in the pathophysiology and treatment of RC disorders.

Materials and Methods

Search Strategy

This systematic literature review was conducted by performing a comprehensive search of three electronic databases (PubMed, Scopus and Web of Science; www.pubmed.gov, www.scopus.com and www.webofscience.com), in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) and PRISMA-S guidelines [20,21]. The review was organized using the PICO framework to provide a thorough assessment of the existing evidence. The Population (P) included both *in vitro* and *in vivo* articles of RC disease focusing on the SAB. The Intervention (I) was defined as the assessment of the biological, regenerative, or pharmacological properties of SAB-derived cells or tissue. Comparison (C) involved control groups (e.g., bone marrow-derived cells or bursa-resected models), while the Outcomes (O) included bursal cellular characterization, gene expression analysis, or biological responses to stimuli and histological features or biomechanical properties of the repaired tendon. The search criteria used for each database are reported in **Supplementary Table 1** of the Supplementary File. The first electronic search was conducted on 25 November 2025 and rerun on 22 July 2026, covering studies published or made available online from January 2015 up to December 2026, including early-access/in-press articles already assigned to 2026 issues and in English language. The inclusion criteria comprised *in vitro*, *in vivo*, *ex vivo*, and *cadaveric* studies investigating the SAB and specifically assessing its role in RC disease.

Studies published in languages other than English, review articles, book chapters, clinical trials and studies not involving an SAB model (*in vitro* or *in vivo*) were excluded.

Two reviewers (VB and MT) independently and in a blinded manner screened and evaluated the retrieved articles using Rayyan web software (accessed 25 November 2025; Rayyan Systems, Inc. Cambridge, MA, USA)[22], a web-based platform designed for systematic and narrative reviews. The software was used to identify and remove duplicate records, and to assess the eligibility of articles based on their titles and abstracts. Furthermore, the reference lists of the included studies were manually screened to identify additional relevant publications. Any disagreements were resolved through discussion. The study selection process is illustrated in Fig. 1.

Data Extraction

VB and MT extracted the key characteristics of the selected papers.

For *in vitro* studies, the data extracted included: the author and year of publication, the aim, the cell source, the cell passage, the number, age and sex of donors, the outcome measures, the culture duration, tear/disease with size or severity, interventions and main findings.

For *in vivo* studies, the following data were collected: the author and year of publication, the animal model, the species, strain, sex and number of animals, the experimental group, the established experimental times, the outcome measures and main findings. In case of missing data, the term Not Reported (NR) was used.

A third and fourth authors (GC and EV) verified the accuracy and completeness of the data. Any disagreements were resolved through discussion. When necessary, a fifth author (MC) was consulted.

Main findings extracted from *in vitro* and *in vivo* studies were uploaded in AI-assisted technology tool for generating infographics (NotebookLM, Google LLC, USA, April 2026 version).

Assessment of Risk of Bias (RoB)

For the *in vitro* studies, the Modified Systematic Review Centre for Laboratory animal Experimentation (SYRCLE)'s RoB tool for *in vitro* research was applied [23]. The checklist utilized in this study comprises ten items, each assigned a risk rating of low, high, or unclear based on the comprehensiveness of the reporting. The assessment was conducted by two independent authors (VB and MT), and any disagreements were resolved through consensus or by a third author (GG).

For the *in vivo* studies, the SYRCLE tool for animal research was applied [24], which includes a 10-item checklist. The RoB was categorized as low, high, or unclear, based on the visibility of the items in question, categorized as reported, NR, or reported unclearly, respectively. For the modified SYRCLE and SYRCLE tools, **Supplementary Tables 2** and **3** report the domain-level decision rules adopted for the RoB appraisals.

The assessment was conducted by two independent authors (VB and MT), and any disagreements were resolved through consensus or by a third author (GG). Inter-rater agreement was calculated using Cohen's kappa (κ), yielding a κ value of 0.77 reflecting good reliability between assessors.

Results

Search Strategy

The review process began with a comprehensive literature search performed using the selected search criteria. This process yielded a total of 18 scientific papers from PubMed (www.pubmed.gov), 18 from Web of Science (www.webofscience.com), and 26 from Scopus (www.scopus.com). The references obtained were imported into the web-based software program, Rayyan. This was used to remove duplicates, resulting in the exclusion of 29 records. Following the removal of duplicates, 33 records remained for screening.

The preliminary evaluation of the abstracts resulted in the exclusion of 14 articles, encompassing two reviews, one editorial comment, two book chapters, one foreign language, one clinical trial, one duplicate and six papers that did not pertain to SAB. The comprehensive catalogue of excluded papers, accompanied by the underlying rationales, is delineated in **Supplementary Table 4** (supplementary file). The remaining 19 articles were then subjected to full-text screening.

This process enabled the identification and subsequent inclusion of an additional 8 articles from the reference lists of the included studies, as illustrated by the PRISMA flow chart (Fig. 1).

Following the application of the screening process, 27 articles satisfied the eligibility criteria for inclusion and were thus incorporated into this review: 3 articles were sourced from PubMed, 4 from Web of Science, and 20 from Scopus.

The papers included a total of 21 *in vitro* studies, constituting 78% of the total, 3 *in vivo* studies, accounting for 11%, and 3 papers (11%) that combined *in vitro* and *in vivo* experiments.

Data Extraction

Table 1, Table 2 and **Supplementary Table 5** present the data extracted from the *in vitro* and *in vivo* studies following the search strategy.

In particular, Table 1 reports data from 21 *in vitro* studies + 3 from the combined studies for a total of 24 items; Table 2 reports data from 3 *in vivo* studies + 3 from the combined studies for a total of 6 items.

The results are reported separately for a more detailed discussion of *in vitro* and *in vivo* studies.

In Vitro Studies

These preclinical *in vitro* studies investigated the role of SAB in RC pathologies. According to the inclusion criteria, all studies employed SAB tissue or SAB-derived cells: most included studies performed mesenchymal cell and tissue characterizations in combination with other tissues or RC tendons, or to test cellular efficacy when used as cellular substrate in association with scaffolds and growth factors in tissue engineering approaches. Fig. 2 summarizes the main results obtained in the included *in vitro* studies.

SAB-Derived Cell Characterizations

Thirteen studies investigated the biological properties of SAB-derived cells using *in vitro* models. These studies collectively addressed cell isolation methods, characterization of mesenchymal stem cell–like properties, proliferative capacity, differentiation potential, interactions with tendon tissue, and the molecular environment of the SAB in the context of RC pathology.

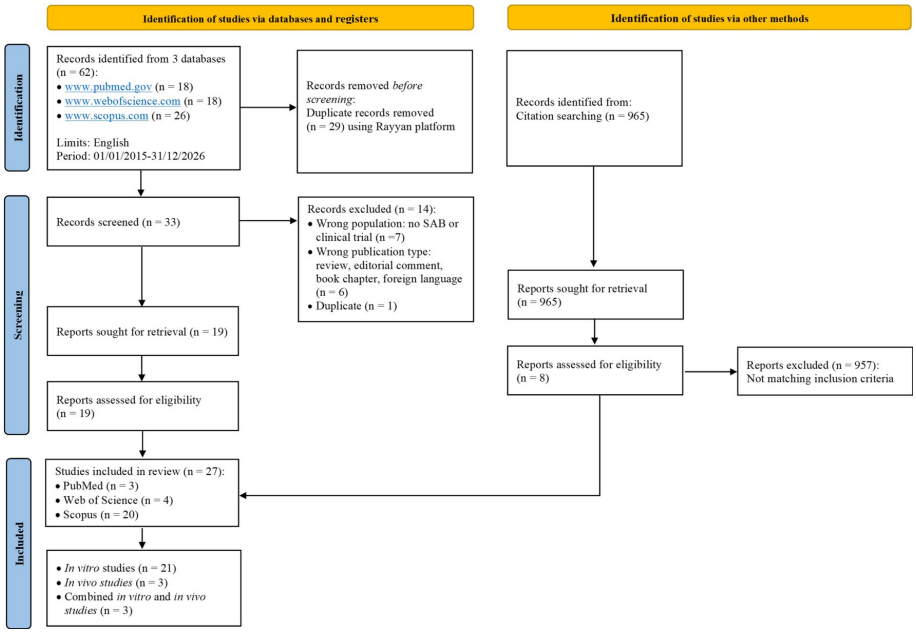


Fig. 1. Systematic review flowchart. Search strategy according to the PRISMA guidelines [20,21].

Samples were obtained exclusively from human donors across all studies. When reported, cell passages generally ranged from 1 to 3; however, approximately 50% of the studies in this group did not specify the passage number. Experimental durations varied between 2 and 21 days, reflecting predominantly short-term culture conditions.

Morikawa *et al.* [25] compared different isolation techniques and demonstrated that mechanical and enzymatic processing (chopping and collagenase, respectively) yields higher numbers of nucleated cells compared to whole tissue culture or homogenization, suggesting these approaches are the most efficient for maximizing SAB cell yield.

Baldino *et al.* [26] further characterized the SAB, identifying two anatomically distinct regions: a muscle-associated portion, which is more adipose and vascularized, and a tendon-associated portion, which is more fibrous and less vascularized. Both regions could generate viable cell cultures, although tendon-derived cells showed greater proliferative capacity. These findings highlight the intrinsic heterogeneity of the bursal tissue.

Several studies confirmed that SAB-derived cells exhibit MSC–like characteristics, including strong proliferative potential and the ability to differentiate along multiple lineages. Muench *et al.* [27] reported that these properties remain consistent across different patient profiles, supporting the robustness of the bursa as a cell source. Klatte-Schulz *et al.* [28] investigated the response of cells to mechanical loading, finding that morphology was unaffected.

Comparative studies with bone marrow–derived cells further support the biological relevance of SAB-derived cells. Steinert *et al.* [29] demonstrated comparable differentiation capacity between the two cell populations, while showing a faster early proliferative response in SAB cells. Similarly, Morikawa *et al.* [30] reported greater proliferation and colony-forming ability in SAB-derived cells over time. Landry *et al.* [31] also showed that SAB cells form colonies more rapidly than bone marrow–derived cells, suggesting potential practical advantages in regenerative applications.

Tamburini *et al.* [32] investigated the interaction between SAB and RC tendon using a co-culture model. Their findings demonstrated an upregulation of extracellular matrix components and tendon-related markers in both tissues, with a more pronounced effect observed under co-culture conditions, suggesting an active paracrine interaction between the bursa and tendon. In fact, this study provides evidence that exposure to interleukin (IL)-1β, as an inflammatory stimulus, leads to a significant increase of inflammatory mediators across all experimental conditions. These findings suggest that the SAB can actively participate in and potentially amplify the inflammatory processes associated with RC disease.

The influence of patient-related factors and RC tear characteristics on SAB-derived cells has been specifically investigated. Muench *et al.* [33] found no significant association between patient demographics and cellular proliferation or biological properties, suggesting that SAB-derived cells maintain consistent behavior across different clinical profiles. In contrast, Morikawa *et al.* [34] reported that tear size negatively correlates with cellular activity, with larger tears associated with reduced colony-forming capacity. This finding was further supported by subsequent analyses from the same group, particularly in tissues sampled from regions directly overlying the tendon [35]. At the molecular level, the SAB demonstrates significant alterat-

Table 1. Data extraction from *in vitro* studies.

Category	Author [Ref]	Aim	Cell source	No. of donors	Outcome measures	Culture duration	Main findings
SAB-derived cell characterization	Morikawa *et al.* [25] 2020	Comparison of 4 isolation methods (chopping, collagenase, homogenization or whole tissue)	Human SAB	7 pts	Nucleated Cell Count, Flow Cytometry (CD105, CD90, CD73, CD45, CD31)	7 d	Chopping and collagenase isolation produced significantly more cells than whole tissue.
	Baldino *et al.* [26] 2020	Classification of muscle and tendon SAB tissue using intraoperative and in vitro characteristics	Human SAB (over muscle and tendon sites)	48 pts	Cellular proliferation (nucleated cell count), Flow cytometry (CD73, CD90, CD105, CD31, CD45, CD 271) and histology (H&E)	3 w	Muscle SAB was mostly fatty and vascularized and tendon SAB mostly fibrous and low vascularized. Forty-one of 48 tendon SAB and 37 of 47 muscle SAB produced confluent plates at 3 w. Tendon SAB had greater proliferation.
	Muench *et al.* [27] 2020	Investigation on mesenchymal potential of SAB cells	Human SAB	16 pts	Cellular proliferation, flow cytometry (CD73, CD90, CD105, CD45 and CD31), multilineage differentiation	2 d	The SAB is a reliable source of cells with high proliferative potential. Age, body mass index, and AHD had no significant influence on cellular measures of SAB cells.
	Klatte-Schulz *et al.* [28] 2021	Analysis of whether and how SAB cells respond to physiological and pathological mechanical loading (3 d at 1 Hz for 1 or 4 h/d at 1, 5 and 10% strain)	Human SAB	6 pts	Proliferation, ELISA (MMP-1, MMP-2, MMP-3, TIMP and COL I), YAP/Actin staining, pMLC, Western Blot, qRT-PCR for COL I, II, III, Aggrecan, Decorin, Versican, Fibromodulin, ITGA, ITGA2, MMP-2, MMP-3, TIMP1, TIMP2	6 d	No morphological and proliferation differences after loading. 1 and 5% loading caused a denser actin cytoskeleton, YAP activation, decrease of TIMP2. COL1A1 and MMP-2 increased with higher/longer straining. No changes in COL3A1, Versican and fibromodulin, MMP-1.
	Steinert *et al.* [29] 2015	Characterization of SAB cells and comparison their MSC properties to bone marrow	Human SAB and BMSCs	10 pts	Cell proliferation assay, immunohistochemistry and flow cytometry (CD44, CD90, CD105, CD73, CD106, CD34, CD133, Stro-1, ALP, FGF), qRT-PCR (AGN, DEC, SOX9), IHH (COL2A1, LPL, PPARγ2, ALP, COL1, Cbfa1), histology and immunohistochemistry (PAS, MG, VG, Azan, CD44, CD90, CD105, Stro-1)	1, 3, 5, 7, 9, 12, and 17 d	SAB cells have significantly higher proliferation on d 3 while BMSCs surpass on the d 7–17. Similar CD expression and multilineage differentiation capacities.

Table 1. Continued.

Category	Author [Ref]	Aim	Cell source	No. of donors	Outcome measures	Culture duration	Main findings
	Morikawa *et al.* [30] 2019	Comparison of the potency of mesenchymal stem cells between SAB and cBMA	Human SAB and cBMA	13 pts	Proliferation, CFU, qRT-PCR (COL II, aggrecan, ALP, COL I, fatty acid binding protein 4 and PPARγ)	3, 5, 7, and 10 d	SAB cells showed increased proliferation over time with higher values, CFU and differentiation ability than cBMA.
	Landry *et al.* [31] 2020	Evaluation of time required for colonies to develop from cBMA and SAB	Human cBMA from proximal humerus and SAB	cBMA: 92 pts SAB: 54 pts	CFU, Flow cytometry (CD73, CD90, CD105 and CD45) and cellularity (only for cBMA).	Up to colonies formation.	Average time to form colonies: cBMA: 7.1 ± 1.5 d SAB: 4.4 ± 1.0 d For cBMA, age associated with more d to form colonies. For SAB, age is associated with longer time to form colonies.
	Tamburini *et al.* [32] 2021	Test a tissue co-culture of RC tendon and SAB in normal and inflammatory conditions	Human RC tendon and SAB	Part 1: 6 pts Part 2: 7 pts	qRT-PCR (COL1, COL3, Tenascin-C, Decorin, Scx, FABP4, ALP, SOX9); biochemical assays NO, ELISA (PGE2, MMP-3, MMP-13) and histology (H&E)	21 d	COL 1 and 3, tenascin C, decorin, and Scx increased at 21 d individually and in co-culture. On 21 d, tenascin-C, decorin, and Scx in the SAB and COL3 and decorin in the tendon increased in co-culture than individually. In part 2: significant increases in inflammatory markers and NO in all conditions.
	Muench *et al.* [33] 2020	Investigation of the influence of pts demographics and RC tear on SAB cells	Human SAB	73 pts	Proliferation assay, flow cytometry (CD73, CD90, CD105, CD45 and CD31) and CFU	3 w	No correlation between pts's or RC tear characteristics and SAB cell proliferation.
	Morikawa *et al.* [34] 2021	Investigation of the influence of pts demographics and tear size on SAB characteristics	Human SAB	30 pts	Flow cytometry (CD105, CD90, CD73, CD45, CD31); CFU; nucleated cell count; Trilineage differentiation	21 d	Tear size negatively correlated with CFUs. No other pts demographics influenced cells.
	Morikawa *et al.* [35] 2021	Investigation of the influence of pts demographics and tear size on SAB characteristics	Human SAB (over muscle and tendon sites)	19 pts	Count and CFUs	14 d	Tear size negatively correlated with CFUs from SAB from tendon.

Table 1. Continued.

Category	Author [Ref]	Aim	Cell source	No. of donors	Outcome measures	Culture duration	Main findings
	Ko *et al.* [36] 2019	Investigation into whether miR-29 is relevant to the development of SAB fibrosis in RC lesion	Human SAB	57 pts (22 with RC tears and stiffness and 35 without)	qRT-PCR (miR-29a, miR-29b, miR-29c and COL) and immunohistochemistry (COL1A1, COL3A1, COL4A1, ADAM12, IL-1β, IL-6, IL-8), histology (H&E, Masson's Trichrome) and histomorphometry (SAB membrane thickness and fibrotic tissue area); in situ hybridization for miR-29a	NR	SAB from RC tears with stiffness had higher synovial membrane thickness, fibrotic tissue area and vessel number, with significant increases COL1A1, COL3A1, COL4A1, ADAM12, IL-1β, IL-6 and IL-8, but decreased miR-29a.
	Minkwitz *et al.* [37] 2021	Comparison of SAB from control healthy (shoulder instability) and different pathologies (osteochondral pathology, partial or full-thickness ST tear)	Human SAB	Healthy: 5 pts osteochondral: 7 pts partial tear: 6 pts full-thickness tear: 16 pts	qRT-PCR (IL-1β, IL-6, TNF-α, COX2, IL-10, TGF-β1, MIF, PENK, GAP43, PGP9.5, MMP-1, MMP-9, BMP2); Histology and immunohistochemistry (H&E, Alcian Blue, αSMA, CD45, CD68 and S100)	Immediate analysis	In the pathology groups more monocytes/macrophages are present. In ostochondral pathology higher numbers of vessels, nerves, IL-6, IL-1β, TNF-α and COX2. MMP-1 increased in full-thickness tears. Expression of pro-inflammatory cytokines correlated with the severity of the tear.
Bursal responses to biochemical and synthetic agents	Muench *et al.* [38] 2021	Evaluation of the effect of IGF-1 on SAB cells over time.	Human SAB	4 pts (3 male, 1 female)	Cellular proliferation; Migration assay and Cell counting	24, 48, 72 and 96 h	Untreated SAB had greater proliferation when compared with IGF-1 and insulin within the first 48 h, although this effect was found to subside by 96 h.
	Karsmarski *et al.* [39] 2022	Evaluation of the functional in vitro effect of activated serum on SAB cells	Human ST and SAB	NR	Proliferation assay, ELISA (PDGF, IGF-1, VEGF, IL-1β, IL-1Ra)	2 d	Activated serum enhanced SAB and ST cell proliferation and increased PDGF, IGF-1 and VEGF, without increasing IL-1β and IL1Ra.
	Hawthorne *et al.* [40] 2025	Examination of effects of PRP and AAS on the polarization of macrophages in co culture of tendon and SAB	Human RC tendon and SAB	6 pts	qRT-PCR (IL-6, IL-12, TNF-α, arginase-1, IL-10, TGF-β) ELISA (same markers) Flow cytometry (CD80, CD86, CD64, CD16, CD32, CD163, CD206) NO assay	16 d	AAS and PRP+ AAS contributed to a large shift to macrophage polarization to the anti-inflammatory M2 compared with PRP.

Table 1. Continued.

Category	Author [Ref]	Aim	Cell source	No. of donors	Outcome measures	Culture duration	Main findings
	Feng *et al*. [41] 2019	Characterization of SAB from tendinopathic and healthy pts and investigation of EGCG role in reducing inflammation	Human RC tendon and SAB	12 symptomatic pts and 3 asymptomatic pts	Histology (H&E), qRT-PCR (pain mediators, cytokines, MMPs, S100, growth factors, Tenocyte assays), IHC/IF (CD34, CD68, PGM1, CD3, CD20, CD138, Substance P, IL-6, IL-1β, Scx) Western blotting (IL-1β and IL-6)	1 and 2 d	Histology: neoangiogenesis, macrophages and mast cells in tendinopathic SAB. qRT-PCR: higher level of TAC1, MCP1, PANX1, P2X7, TNF-α, IL-6, MMP-1 and S100 proteins than healthy SAB. Western blot: higher IL-6 and IL-1β than healthy. IHC/IF: higher substance P, IL-6, and IL-1β. EGCG reduced TAC1, MCP1, PANX1, P2X7, TNF-α, IL-1β, IL-6, MMP-1, MMP-13, VEGF and NGF.
	Hawthorne *et al*. [42] 2024	Quantification of cellular senescence in pathological ST and SAB and investigation of the in vitro efficacy of the senolytic dasatinib + quercetin (D + Q)	Human SAB and ST	41 pts	Part1: IHC (p16INK4a and p21), qRT-PCR (p16INK4a and p21) and SASP (IL-6, IL-8, MMP-3, MCP-1) Part 2: qRT-PCR (p16INK4a and p21) SASP (IL-6, IL-8, MMP-3, MCP-1), qRT-PCR (COL1A1, COL3A1, Scx, decorin, and tenascin C), and ELISA (IL-6, IL-8, MMP-3 and MCP-1)	Part 1 + 2: 38 d	Patiet's age-dependent increase in markers of SAB and tendon senescence (p21; IL-6, IL-8, MMP-3). D + Q treatment in tendons: decreased p21, IL-6, and IL-8 and increased COL1A1. D + Q treatment in SAB: decreased p21, and IL-8 and increased COL1A1.
	Muench *et al*. [43] 2021	Evaluation of effects of Mg on human SAB, osteoblasts, and tenocytes on nonabsorbable suture tape	Human SAB, bone fragments and tendons	3 pts	Adhesion and proliferation assays	2 d	Mg increased SAB adhesion compared to untreated sutures; no significant increase for osteoblasts and tenocytes. No differences in proliferation in any populations.
	Muench *et al*. [44] 2023	Evaluation of the effect of Mg, PRP or both on SAB adhesion and proliferation	Human SAB	4 pts	Cellular adhesion and proliferation assays	2 d	Mg increased cellular adhesion. Mg with or without PRP increased proliferation.

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Table 1. Continued.

Category	Author [Ref]	Aim	Cell source	No. of donors	Outcome measures	Culture duration	Main findings
	Muench *et al.* [45] 2020	Evaluation of DBM, as scaffold, for cBMA, SAB, tenocytes, and osteoblasts	Human cBMA from proximal humerus, SAB, tenocytes and osteoblasts from knee	1 pts for cBMA, SAB and tenocyte and 1 pts for osteoblasts	Cell count, adhesion and proliferation assays, flow cytometry (CD105, CD90, CD73, CD45, CD31), histology (H&E), SEM	2 d	Osteoblasts higher proliferation on DBM than other cells.
	Lebaschi *et al.* [46] 2022 (both *in vitro* and *in vivo*)	Evaluation of SAB cell proliferation on fibrin beads	Murine SAB	10 C57Bl/6 mice	Viability assays at different concentration onto fibrin beads	0, 1, 3, 5 d	Live cells distributed in all 3 different concentrations at all time points.
	Marshall *et al.* [47] 2024 (both in vitro and in vivo)	Identification of the MSC bursa population and its use in in vitro co-culture model of inflammation and bursa–tendon crosstalk, including the response to dexamethasone	Rat SAB, ST and infraspinatus tendons	Sprague Dawley Rats	Coculture bursa–tendon fibroblasts with IL-1β; qRT-PCR (Alp, Lpl, Adipoq, Pparg, Acan, COL2A1, SOX9, Scx, Tnmd, COX2, IL-1β, IL-6, IL-10, TGF-β, SDF-1α, TNF-α, Runx2, COL1, COL3, Bgn, MMP-1, MMP-8, MMP-13, Timp1, Timp2, Timp3), transcriptomics, histology (Alizarin Red, Alcian Blue, Green Neutral Lipid Stain, Masson's trichrome), immunocytochemistry (CD73, CD90, AF488 and AF594), flow cytometry (CD90, CD45, CD29), viability (Live/Dead), NO release.	3, 7, 14 and 21 d	Rat SAB contains MSC population capable of osteogenic, adipogenic, chondrogenic and tendogenic differentiation. SAB remain viable for at least 8 d and respond to IL-1β with increased NO and inflammatory genes, a response that can be reduced by dexamethasone.
	Dyrna *et al.* [48] 2018 (both *in vitro* and *in vivo*)		Human SAB and bone marrow from proximal humeral head	5 pts	Yield, CFU	7 d	No statistical analysis performed.

Table 2. Data extraction from *in vivo* studies.

Author [Ref]	Animal model	Species, strain, sex, number	Exp. groups	Exp. time	Outcome measures	Main findings
Lebaschi *et al.* [46] 2022	ST detachment and repair.	12-w-old male C57Bl/6 mice, 245	Part 1: 2 groups: Study group: repair + fibrin with 100,000 SBCs Control group: repair + fibrin without cells Part 2: 4 groups: Tissue group: fresh SAB (2 × 2 mm); Cell group: 100,000 SBCs in fibrin; Fibrin group: fibrin without cells; Control group: standard repair without augmentation	2 and 4 w	Biomechanics (failure force) and histological analysis (cell count and proteoglycan content)	Study 1: failure force in the study group significantly higher than controls at 2 and 4 w but lower than native tendons. Lower cell density in the study group at 2 and 4 w compared with the controls. No differences in proteoglycan content. Study 2: no difference in failure force between cell and tissue groups and between fibrin and control at 2 and 4 w.
Marshall *et al.* [47] 2024	ST tenotomy with or without bursectomy and ST tenotomy + repair with or without bursectomy	Sprague–Dawley male/female rats, 54	In both models 2 groups: BP BR	Tenotomy: 3, 7 and 14 d Tenotomy + repair: 14, 28 and 56 d	Histology (Movat's pentachrome, H&E); Biomechanics (strength, stiffness, and toughness); Micro-CT (bone area, area fraction, cross-sectional thickness and trabecular bone volume, volume fraction, number, thickness, separation); qPCR (COX2, IL-1β, IL-6, IL-10, TGF-β, SDF-1α, TNF-α, COL1, COL3, SCX, SOX9, Runx2, Timp, MMP-13); Transcriptomics	*IL1B*, *IL10*, *CXCL12*, and *TNFA* gene expression increased BP compared with BR. Tensile strength and stiffness not different from control when BP, but in the bursectomy group, strength and stiffness lower when the BP. BP upregulated ECM-related gene expression (COL1; COL3; SCX) compared to the BR. Trabecular bone volume and trabecular number decreased in BR. Trabecular thickness was decreased in BP compared with sham control or BR.
Dyrna *et al.* [48] 2018	Patellar tendon defects	NOD Scid gamma male/female mice, 72	Group A: human SBCs (150,000 cells) in fibrin gel; Group B: human humeral head BMSCs (150,000 cells) in fibrin gel. Group C: fibrin gel without cells; Group D: naturally healing control.	1, 2, 5, or 8 w	Multiphoton Microscopy (cell engraftment and survival, healing thickness); IHC (COL I, III, IV, tenascin-C, human nuclear antigen and F4/80 macrophage); Biomechanics (static stress relaxation and failure load)	No differences in tendon rupture rate and between male/female mice. SBCs had greater survival and greater engraftment into host tissue compared to BMSCs. The presence of cells (SBCs or BMSCs) increases the thickness of healing tissue compared to controls and greater infiltration and three-dimensional integration into tendon tissue. No differences in macrophage recruitment, no group completely restored the mechanical properties of the native tendon.

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Table 2. Continued.

Author [Ref]	Animal model	Species, strain, sex, number	Exp. groups	Exp. time	Outcome measures	Main findings
Sun *et al*. [49] 2021	Bilateral ST tendon defects with and without BP	Sprague–Dawley male rats, 25	Right side→ defect + BP Left side→ defect + BR	3 and 9 w	Histological and IHC analysis (H&E, Modified Bonar Score, COL I and III) Biomechanical testing (maximal load-to-failure and stiffness)	At 3 w better alignment in the BP; at 9 w superior tendon quality compared with the BR. BP is significantly better maximum load at 3 and 9 w and better stiffness at 9 w than BR.
Tanimura *et al*. [50] 2026	ST tendon detachment and repair with and without BP	Sprague-Dawley rats (n = 71, 35 males, 36 female) and ScxGFP transgenic rats (12 female)	4 groups: intact, SAB injury-only, BP, BR.	1, 3 and 6 w	scRNA-seq, histological and histomorphometric analyses (H&E, cellularity, vascularity, collagen fiber scores) Immunohistochemical (GFP, SOX9, PDGFRα and CD34) analysis, Imaging of the second harmonic generation (COL fiber orientation), Biomechanical testing (ultimate stress and load to failure and stiffness)	SAB contains progenitors (PDGFRα+, CD34+) that can differentiate into Scx+/SOX9+ and facilitate tendon-bone healing. COL fiber orientation higher in the BP RC repair; at 3 w BP RC repair significantly higher ultimate load to failure, stiffness and stress.
Miura *et al*. [51] 2024	Bilateral ST full-thickness RC tear with and without BP	Lewis male rats, 21	Right side→ defect + BR Left side→ defect + BP	3 and 6 w	Histology (H&E, Masson's trichrome) Immunohistochemistry (COLIII, decorin, SCX) RT-qPCR (COL I, COLIII, tenomodulin, decorin, SCX, MMP-13, IL-1β, IL-6,iNOS, TGF-β, IL-10, Arg-1)	BP promotes more effective tendon healing, with adequate progression from inflammation to repair, increased vascularisation and thickening of the tendon.

ions in the presence of RC pathologies. Furthermore, Ko *et al*. [36] reported increased expression of pro-inflammatory and fibrosis-related factors in patients with RC tears and stiffness, along with structural changes, such as increased tissue thickness, vascularity, and fibrosis.

Similarly, Minkwitz *et al*. [37] observed increased inflammatory cell infiltration and elevated cytokine expression in different pathological conditions compared to shoulder instability, that was considered as control. These changes were found to correlate with disease severity, suggesting a progressive activation of inflammatory and degenerative pathways even within the bursal microenvironment.

Bursal Responses to Biochemical and Synthetic Agents

Eight studies investigated the biological response of SAB-derived cells to various biochemical stimuli, including growth factors, natural anti-inflammatory compounds, autologous serums, senolytics, and metallic ions. Regarding cell culture characteristics, reported cell passages ranged from 1 to 3. However, there is a significant lack of standardized reporting in this area, as 63% of the studies in this group did not specify the passage number. Experimental durations were highly variable, spanning a range from 2 to 38 days.

The influence of anabolic factors and blood-derived products on SAB cell behavior was a primary focus, particularly regarding their potential to modulate the local environment. While basic growth factors like insulin and insulin-like growth factor-1 (IGF-1) showed a limited or transient effect on cell proliferation compared to untreated controls [38], the use of activated serum demonstrated a more robust capacity to enhance cellular activity and the secretion of endogenous trophic factors [39]. In co-culture models, Hawthorne *et al*. [40] found that autologous activated serum (AAS) proved superior to standard Platelet-Rich Plasma (PRP) in promoting a significant shift toward anti-inflammatory macrophage phenotypes, suggesting a potent paracrine role in the resolution of local inflammation.

Natural compounds such as epigallocatechin-3-gallate (EGCG) were shown by Feng *et al*. [41] to broadly downregulate the expression of various catabolic enzymes and pro-inflammatory mediators that characterize pathological bursal tissue.

Evidence of age-dependent cellular senescence was found within the SAB by Hawthorne *et al*. [42]. Their work demonstrated that the administration of senolytic agents (Dasatinib + Quercetin) successfully cleared these senescent markers and promoted a shift toward a more regenerative, matrix-producing phenotype.

The compatibility of SAB cells with biomaterials, surgical tape materials and ions suggests their potential utility in augmented repair strategies. Magnesium ions were found to significantly enhance the physical interaction of SAB cells with their environment, with Muench *et al*. [43,44] reporting increased cellular adhesion and promoted proliferation even when applied to synthetic surgical tapes. Muench *et al*. [45] demonstrated that SAB cells effectively colonize and grow when seeded onto demineralized bone scaffolds, confirming their viability as a local cell source even for tissue engineering approaches.

Three works, combining *in vitro* and *in vivo* experiments, isolated and expanded SAB cells for subsequent *in vivo* implantation [46–48]. In these studies, the cellular sources included murine, rat, and human SAB cells to be transplanted in an autologous manner [46,47] or into immunodeficient (Non-obese diabetic/severe combined immunodeficiency (NOD/SCID)) mice [48]. Cellular characterization was performed up to 7 days of culture to confirm mesenchymal properties, including yield, viability, colony-forming unit (CFU) and biological activity when in pathophysiological conditions.

In Vivo Studies

All included studies used small rodent animal models. Out of the six included works (3 with only *in vivo* and 3 combining *in vitro* and *in vivo* procedures), 2 articles used a mice model (33 %) [46,48] and four used a rat model (67 %) [47,49–51]. Models with C57Bl/6 and NOD Scid mice were adopted for investigating the role of SAB-derived cell transplantation or whole intact bursa in the healing of tendon defects or repair; cells were loaded into a fibrin clot and transplanted up to 8 weeks. Post-explant analysis included histology, immunohistochemistry, biomechanics and multiphoton microscopy. Interestingly, Dyrna *et al* [48] compared the healing properties of SAB or bone marrow derived cells also between male and female animals: they found superior survival and engraftment of bursal cells compared to bone marrow ones without differences between animal's sex, while mechanical properties of repaired tendons resulted inferior to those of native tendon.

Four studies [47,49–51] employing rats, Sprague-Dawley and Lewis strains, adopted the supraspinatus tendon detachment model: all aimed at comparing the tendon healing and repair with or without bursectomy. Two studies analyzed sex differences in healing adopting both male and female animals [48,50]. Experimental times ranged from 3 days up to 9 weeks. Analyses included histology, immunohistochemistry, biomechanics, Micro-computed tomography (microCT), Quantitative Real-Time Polymerase Chain Reaction (qPCR) and transcriptomics.

Overall, as also depicted in Fig. 3, the studies converged on the hypothesis that SAB preservation, through appropriate inflammatory and repair phases, positively impacts and contributes to the histological maturation of the repaired tendons. From a biomechanical point of view, half studies found better mechanical competence of repaired tendon, when the bursal tissue was left *in situ* [47,49].

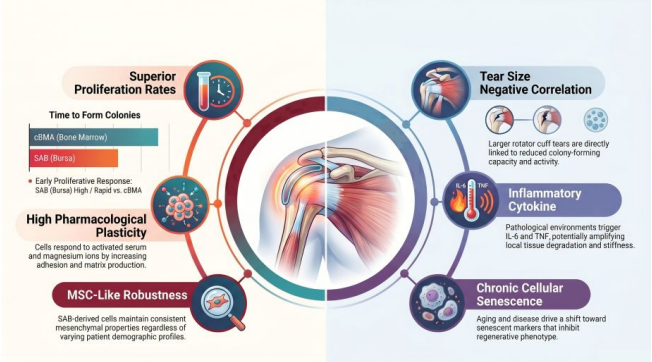


Fig. 2. Comprehensive infographic of data extracted from *in vitro* studies. The visualization illustrates the key findings discussed in Section "*In Vitro* Studies", highlighting the balance between regenerative potential and pathological burdens. This image was generated using NotebookLM (April 2026 version; Google LLC, Mountain View, CA, USA).



Fig. 3. Comprehensive infographic of data extracted from *in vivo* studies. The infographic illustrates the research models, analytical techniques, and key outcomes discussed in Section "*In Vivo* Studies". This image was generated using NotebookLM (April 2026 version).

Assessment of RoB

The summary analysis presents the overall RoB for both *in vitro* (Fig. 4, **Supplementary Table 6**) and *in vivo* (Fig. 5, **Supplementary Table 7**) studies, based on the final percentage distribution across all assessed domains.

For *in vitro* studies, the highest proportion of high-risk judgments was observed in item 3 "Detailed explanation of intervention" (96%), indicating a consistent methodological weakness across nearly all included studies in this domain. Elevated levels of risk were also identified in item 1 "Sample size calculation" (75%), item 5 "Details of comparison group" (71%), and item 10 "Other sources of bias" (58%). These items evidenced mainly the lack of an *a priori* power analysis to determine the minimum sample size that ensures the statistical significance of results, the inconsistency in the choice of control groups and the presence of supposed conflict of interests.

In contrast, items 7 "Blinding" and 8 "Incomplete outcome data" showed as low risk (75% and 71%, respectively), reflecting greater methodological consistency. The remaining items showed a more heterogeneous distribution between low, high, and unclear risk, indicating variable reporting quality across studies.

For the *in vivo* studies, several domains demonstrated a consistently high RoB. Items 1 "Sequence generation", 3 "Allocation concealment" and 5 "Blinding of caregivers/investigators" showed the highest proportion of high-risk judgments (83% each), highlighting critical shortcomings in these methodological areas. Items 4 "Random housing" and 7 "Blinding of outcome assessor" also presented substantial high-risk rates (67%), while item 6 "Random outcome assessment" was characterized by a mixed pattern, with 50% of studies rated as unclear risk and the other half at high risk. Furthermore, items 8 "Incomplete outcome data" and 9 "Selective outcome reporting" demonstrated the most robust methodological profile, with all studies classified as low RoB (100%), and item 10 "Other sources of bias" was predominantly rated as low risk (83%).

Discussion

The present systematic review aims to summarize the preclinical evidence regarding the role of SAB in shoulder and RC pathology for the final goal to clarify its potential role in the pathophysiology and treatment of RC disorders.

Most studies reveal that a large population of viable MSCs is contained within the bursal tissue: these cells fulfil the MSC minimal criteria as referred in the International Society for Cellular Therapy positioning statement by Do-

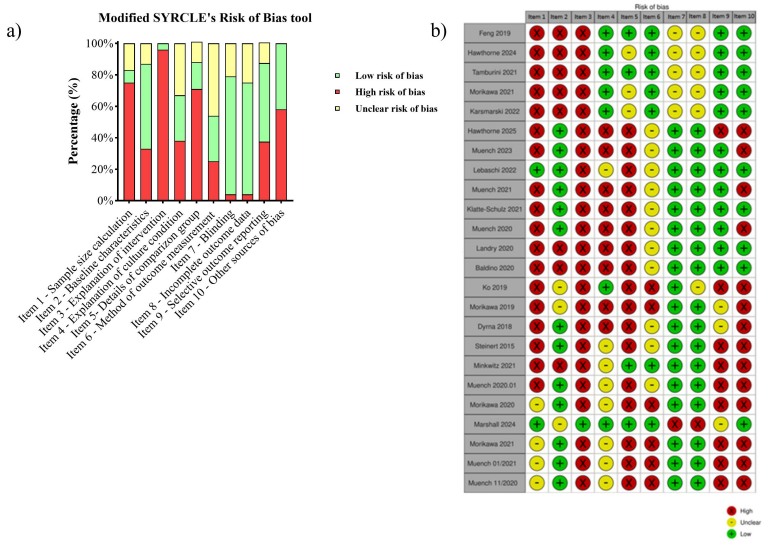


Fig. 4. *In vitro* RoB results. Overall RoB (**a**) and assessments for each *in vitro* study (**b**), as determined by using the Modified SYRCLE's RoB tool [23]. Each item is reported as having a low (green), high (red) or unclear (yellow) RoB. Graphs in (**a**) and (**b**) were realized with GraphPad Prism (v.11.0.1, GraphPad Software LLC, San Diego, CA, USA) and robvis tool [52], respectively.

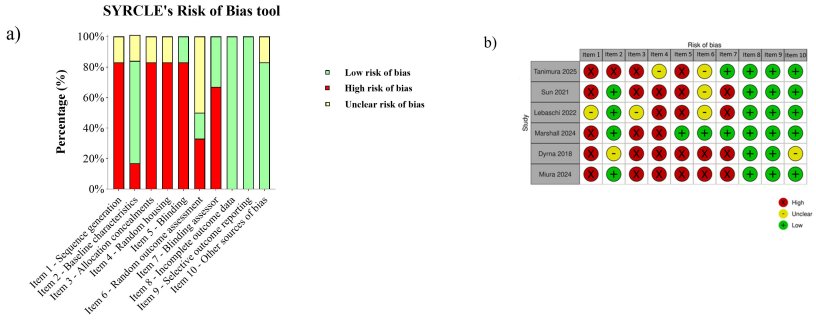


Fig. 5. *In vivo* RoB results. Overall RoB (**a**) and assessment for each *in vivo* study (**b**), as determined by using the SYRCLE's RoB tool [24]. Each item is reported as having a low (green), high (red) or unclear (yellow) RoB. Graphs in (**a**) and (**b**) were realized with GraphPad Prism (v.11.0.1) and robvis tool [52], respectively.

minici *et al*. [53] in term of cluster of differentiation (CD) expression, adherence to plastic and multilineage differentiation capacity. Additionally, SAB derived cells displayed higher CFU and proliferative potential than concentrated bone marrow derived cells: these findings are also confirmed from *in vivo* studies, where superior survival and engraftment of bursal cells compared to bone marrow ones was found.

Retrieved *in vivo* studies are few, all performed in rodents, and employ mainly two different experimental models of surgically induced RC disease: tendon detachment followed by standard repair or the creation of tendon tears. In addition, tendon defect dimensions aren't reported, and thus the variability inherent to the utilized experimental models constrains cross-study comparisons, precluding the establishment of a robust evidence-based framework.

Moreover, the review shows the lack of studies conducted in larger animal models, such as sheep or goats and pigs: despite they share anatomical analogy with humans and thus are preferred for clinical translatability, they have notable disadvantages represented by extensive costs associated with housing and surgical procedures [54].

In general, animal models for RC repair are limited by profound biomechanical differences between the human and quadrupedal anatomy. In fact, unlike the human scapulohumeral joint which has a wide range of motion, quadrupedal joints have restricted movement patterns, as the limb remains largely integrated with the trunk. Due to these limitations, and the ethical barriers surrounding the use of non-human primates—the only species with comparable mechanics—*in vivo* studies are few [55].

Furthermore, the diagnostic gold standard for evaluating the SAB and the whole RC—Magnetic Resonance Imaging (MRI)—is prohibitively expensive and frequently unavailable in standard animal research facilities. This technical and economic constraint significantly hampers the

longitudinal monitoring of soft-tissue healing in preclinical models [56–59]. From our review, none of the *in vivo* studies used MRI or ultrasound evaluations for evaluating the outcome measures.

Within the RoB assessments, in the present review the validated SYRCLE's RoB tool for *in vivo* studies was adopted and the modified SYRCLE's RoB tool for *in vitro* studies, adjusted but not validated for application to *in vitro* research, was applied. The quality of the current evidence resulted limited by methodological gaps, mainly affected by selection and detection bias: most of *in vitro* and *in vivo* research inadequately performed and reported allocation, concealment, randomization and blinding measures as well as the *a priori* statistical analysis to determine the sample size. This issue is particularly important for fulfilling the ethical requirement of reduction within the 3Rs principle (Replacement, Reduction and Refinement). In addition, more than half of the *in vitro* studies were rated at high RoB for the "other sources of bias" domain, related to the presence of conflict of interests and/or funding received. Although these conflicts of interest and funding sources were transparently disclosed, their presence was nonetheless considered a potential source of bias according to the risk-of-bias tool criteria, as they may still have influenced the design, conduct, or interpretation of the study results.

Consequently, although the mobilisation or reimplantation of bursal tissue remains a promising biological augmentation strategy, further high-quality studies are needed to clarify its definitive role in the pathophysiology and treatment of RC disorders. Beside the relatively low quality, all *in vitro* and *in vivo* works support the benefits of using bursal tissue and cells, but preclinical evidence does not yet endorse its routine use. This discrepancy may be partly explained by the multifactorial causes of RC disease, which include intrinsic factors such as age- and sex-related tendon degeneration and decreased vascularity, as well as extrinsic mechanical stresses, overuse or traumas. In preclinical settings, sex determinants are underestimated: among the *in vivo* studies, 50% (3 out of 6 papers) used both male and female animals, but only one study [48] correlated the retear rate with animal's sex, finding no differences. Among *in vitro* studies, quite all authors isolated SAB derived cells from male and female patients, but only few determined relationships between sex with outcomes [33,34], finding no correlation. The patients' parameters, found to be inversely linked to SAB cells potentials, were the tear size [34,35,37] and patient's age [31]. The major limitation in the current literature is the "male-default": the omission of female determinants prevents from the understanding how bursa-derived cells interact within a female-specific environment, that frequently exhibit distinct pathological patterns, such as a higher prevalence of adhesive capsulitis comorbidities or altered collagen remodeling [60,61]. While it is true that the prevalence of RC injuries is higher in men, primarily for mechanical and occupational reasons, this gap narrows with increasing age, suggesting that hormonal factors related to menopause accelerate tendon degeneration in women. Regarding the healing and recovery after RC surgery, women also experience higher levels of disability compared to men, with poorer functional scores and longer healing times [62–65].

The main limitation of the present review is the inclusion of works edited after 2015 and limited to the last 10 years and lack of the protocol registration, at inception, in an international database, such as PROSPERO for increasing transparency. However, PROSPERO includes two registries for systematic reviews on human participants and animals, respectively, excluding *in vitro* studies. Another limitation is the adoption of a not validated RoB tool for *in vitro* assays. To the author's knowledge, the only validated RoB tool for *in vitro* assays is available in dentistry (QUIN Tool—Quality Assessment Tool for *In Vitro* Studies) [66]. Many others are proposed in different fields, such as ToxR-Tool (Toxicological data Reliability Assessment Tool) [67], INVITES-IN (*In Vitro* Experimental Studies Internal Validity) [68], OHAT (RoB Rating Tool) [69] and the modified SYRCLE's [23]. Since the present review isn't related to dentistry, QUIN's domain structure is not directly transferable to our study population. After having considered all the above-mentioned alternative, the modified SYRCLE's tool is selected because it allows methodological consistency between the *in vivo* and *in vitro* arms of the review, using a parallel domain structure.

In this review, the modified SYRCLE's RoB is performed to be in line with animal studies. Despite these limitations, the adoption of RoB tools for both *in vitro* and *in vivo* papers ensure the methodological appraise of quality.

This review provides a comprehensive overview of the preclinical scenario about SAB characteristics and potential in RC repair, with the aim of supporting its use in conjunction with therapeutic strategies for the RC pathology.

Conclusions

In conclusion, the SAB appears to be biologically active tissue. It plays a dual role in RC disease: while it can respond to the inflammatory environment, it also serves as a vital source of mesenchymal progenitor cells and bioactive factors. The current evidence suggests that preserving and utilizing bursal tissue during RC repair may promote tendon healing. However, the present review also highlights the relatively low quality of the available preclinical evidence, which is characterized by limited methodological rigor and high-to-unclear risks of bias, thereby hampering the translational potential of these findings. Further clinical trials are necessary to determine whether these biological properties translate into superior structural outcomes, to determine the field of applicability within the wide range of RC pathologies, and the long-term functional recovery for patients.

List of Abbreviations

AAS, autologous activated serum; CD, cluster of differentiation; CFU, colony-forming unit; EGCG, epigallocatechin-3-gallate; microCT, Micro-computed tomography; MRI, Magnetic Resonance Imaging; MSC, mesenchymal stem cell; NR, Not Reported; NOD/SCID, Non-obese diabetic/severe combined immunodeficiency; PRISMA, Preferred Reporting Items for Systematic Reviews and Meta-Analyses; PRP, Platelet-Rich Plasma; RoB, Risk of Bias; SYRCLE, Systematic Review Centre for Laboratory animal Experimentation; αSMA, alpha smooth muscle actin; ADAM, a disintegrin and metalloproteinase; Adipoq, adiponectin; AF488, Alexa Fluor 488; AGN, Angiotensinogen/Angiotensin; AHD, Acromiohumeral distance; ALP, alkaline phosphatase; ARCR, arthroscopic rotator cuff repair; Azan, azocarmine aniline blue; Bgn, biglycan; BMP, bone morphogenetic protein; BMSCs, bone marrow-derived mesenchymal stem cells; Cbfa1, core-binding factor subunit alpha 1; cBMA, Concentrated Bone Marrow Aspirate; COL, collagen; COX, Cyclooxygenase; d, day/days; D + Q, dasatinib + quercetin; DBM, demineralized bone matrix; d/h, days / hours; DEC, Diethylcarbamazine; FABP, fatty acid binding protein; FGF, Fibroblast Growth Factor; GAP, growth associated protein; h, hour/hours; H&E, hematoxylin and eosin; Hz, Hertz; IHH, Indian Hedgehog (Cell signaling protein); IHC/IF, immunohistochemistry/immunofluorescence; IGF, insulin-like growth factor-1; IL, interleukin; IL-1Ra, Interleukin-1 Receptor Antagonist; iNOS, inducible nitric oxide synthase; ITG1, Beta-1 Integrin (ITGB1); LPL, lipoprotein lipase; MCP, monocyte chemoattractant protein; MG, Masson's trichrome; Mg, magnesium; MIF, macrophage migration inhibitory factor; miR, microRNA; MMP, matrix metalloproteinase; mo, month/months; NGF, nerve growth factor; NO, nitric oxide; p16INK4, Inhibitor of Cyclin-Dependent Kinase 4; P2X7, P2X Purinoceptor 7; PANX, pannexin; PAS, periodic acid–Schiff; PDGF, platelet-derived growth factor; PENK, proenkephalin; PG, prostaglandin; PGM1, Phosphoglucomutase-1; PGP, protein gene product; pMLC, phosphorylated myosin light chain; PPAR, peroxisome proliferator-activated receptor; pts, patient/patients; qRT-PCR/qPCR, Quantitative Real-Time Polymerase Chain Reaction; RC, rotator cuff; Runx, runt-related transcription factor; S100, S100 proteins (Calcium-binding protein family); SAB, subacromial bursa; SASP, senescence-associated secretory phenotype; Scx, scleraxis; SDF, Stromal Cell-Derived Factor; SEM, scanning electron microscopy; SOX, SRY-box transcription factor; ST, supraspinatus tendon; Stro, stromal precursor antigen; TAC1, Tachykinin Precursor 1; TGF, transforming growth factor; TIMP, tissue inhibitor of metalloproteinases; TNF, tumor necrosis factor; TNMD, Tenomodulin; VEGF, vascular endothelial growth factor; VG, Van Gieson; YAP, Yes-associated protein; w, week/weeks; ARG, Arginase; BP, bursa preservation; BR, bursa resection; ECM, Extracellular Matrix; GFP, Green Fluorescent Protein; PDGFR, Platelet-Derived Growth Factor Receptor; SBC, subacromial bursa cell; scRNA-seq, Single-cell RNA sequencing.

Availability of Data and Materials

The raw data generated in this systematic review, including the list of excluded papers and the results of the RoB assessments, are provided in **Supplementary Tables 4, 6,** and **7**.

Author Contributions

VB, GG and MT contributed to the design of this work; GC and EV contributed to the interpretation of data; VB, MC, GC, EV and MT analyzed the data and drafted the work; GG, MC and MT revised critically for important intellectual content. All authors read and approved the final manuscript. All authors agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

Ethics Approval and Consent to Participate

Not applicable, as this study is a review article and does not involve human participants or animal subjects.

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The Graphical Abstract and infographics in Figs. 2,3 were generated using NotebookLM (Google LLC, USA; April 2026 version).

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Conflict of Interest

The authors state that they have no competing financial interests or personal relationships that could have affected the work described in this paper.

Supplementary Material

Additional supporting information, including the detailed search strategy (**Supplementary Table 1**), domains and judgment criteria applied for *in vitro* and *in vivo* studies (**Supplementary Tables 2** and **3**), the list of excluded studies with reasons (**Supplementary Table 4**), additional methodological details of *in vitro* studies (**Supplementary Table 5**) and the raw data of the RoB assessment (**Supplementary Tables 6** and **7**) can be found in the online version of this article as Supplementary Material, at https://doi.org/10.22203/eCM.v059a18.

References

[1] Littlewood C, May S, Walters S. Epidemiology of rotator cuff tendinopathy: a systematic review. Shoulder & Elbow. 2013; 5: 256–265. https://doi.org/10.1111/sae.12028.

[2] Luime JJ, Koes BW, Hendriksen IJ, Burdorf A, Verhagen AP, Miedema HS, *et al*. Prevalence and incidence of shoulder pain in the general population; a systematic review. Scandinavian Journal of Rheumatology. 2004; 33: 73–81. https://doi.org/10.1080/03009740310004667.

[3] Mitchell C, Adebajo A, Hay E, Carr A. Shoulder pain: diagnosis and management in primary care. BMJ: British Medical Journal/British Medical Association. 2005; 331: 1124–1128. https://doi.org/10.1136/bmj.331.7525.1124.

[4] Minagawa H, Yamamoto N, Abe H, Fukuda M, Seki N, Kikuchi K, *et al*. Prevalence of symptomatic and asymptomatic rotator cuff tears in the general population: From mass-screening in one village. Journal of Orthopedics. 2013; 10: 8–12. https://doi.org/10.1016/j.jor.2013.01.008.

[5] Yamamoto A, Takagishi K, Osawa T, Yanagawa T, Nakajima D, Shitara H, *et al*. Prevalence and risk factors of a rotator cuff tear in the general population. Journal of Shoulder and Elbow Surgery. 2010; 19: 116–120. https://doi.org/10.1016/j.jse.2009.04.006.

[6] Ramirez J. Adhesive Capsulitis: Diagnosis and Management. American Family Physician. 2019; 99: 297–300.

[7] Zreik NH, Malik RA, Charalambous CP. Adhesive capsulitis of the shoulder and diabetes: a meta-analysis of prevalence. Muscles Ligaments and Tendons Journal. 2016; 6: 26–34. https://doi.org/10.11138/mltj/2016.6.1.026.

[8] Hoelen MA, Burgers AM, Rozing PM. Prognosis of primary anterior shoulder dislocation in young adults. Archives of Orthopaedic and Trauma Surgery. 1990; 110: 51–54. https://doi.org/10.1007/BF00431367.

[9] Garving C, Jakob S, Bauer I, Nadjar R, Brunner UH. Impingement Syndrome of the Shoulder. Deutsches Ärzteblatt International. 2017; 114: 765–776. https://doi.org/10.3238/arztebl.2017.0765.

[10] Ibounig T, Simons T, Launonen A, Paavola M. Glenohumeral osteoarthritis: an overview of etiology and diagnostics. Scandinavian Journal of Surgery. 2021; 110: 441–451. https://doi.org/10.1177/14574969209350 18.

[11] Longo UG, Berton A, Khan WS, Maffulli N, Denaro V. Histopathology of rotator cuff tears. Sports Medicine and Arthroscopy Review. 2011; 19: 227–236. https://doi.org/10.1097/JSA.0b013e318213bccb.

[12] HajAssaad A, Willacy R, Wilson R. A Systematic Review of the Histological and Molecular Changes in the Subacromial Bursa in Rotator Cuff Disease. Journal of Surgical Orthopaedic Advances. 2020; 29: 1–4.

[13] Ma Y, Jiang W, Qian P, Guo JJ. Removal or retention: evolving views on possible roles of the subacromial bursa in rotator cuff disease. EFORT Open Reviews. 2025; 10: 851–864. https://doi.org/10.1530/EOR-2024-0183.

[14] Utsunomiya H, Uchida S, Sekiya I, Sakai A, Moridera K, Nakamura T. Isolation and characterization of human mesenchymal stem cells derived from shoulder tissues involved in rotator cuff tears. The American Journal of Sports Medicine. 2013; 41: 657–668. https://doi.org/10.1177/0363546512473269.

[15] Dalal AH, Grogan SP, Bijanki V, Nwadike B, D'Lima DD, Alberton LA. Bursal Tissue Harvested During Rotator Cuff Repair Contains Viable Mesenchymal Stem Cells. Arthroscopy, Sports Medicine, and Rehabilitation. 2024; 6: 100947. https://doi.org/10.1016/j.asmr.2024.100947.

[16] Montgomery SR, Petrigliano FA, Gamradt SC. Biologic augmentation of rotator cuff repair. Current Reviews in Musculoskeletal Medicine. 2011; 4: 221–230. https://doi.org/10.1007/s12178-011-9095-6.

[17] Rodeo SA. Biologic augmentation of rotator cuff tendon repair. Journal of Shoulder and Elbow Surgery. 2007; 16: S191–S197. https://doi.org/10.1016/j.jse.2007.03.012.

[18] Rynecki ND, Pereira DS. The Role of Mesenchymal Stem Cells in Augmenting Rotator Cuff Repairs. Bulletin of the Hospital for Joint Diseases. 2018; 76: 232–237.

[19] Freislederer F, Dittrich M, Scheibel M. Biological Augmentation With Subacromial Bursa in Arthroscopic Rotator Cuff Repair. Arthroscopy Techniques. 2019; 8: e741–e747. https://doi.org/10.1016/j.eats.2019.03.010.

[20] Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffmann TC, Mulrow CD, *et al*. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. British Medical Journal. 2021; 372: n71. https://doi.org/10.1136/bmj.n71.

[21] Rethlefsen ML, Kirtley S, Waffenschmidt S, Ayala AP, Moher D, Page MJ, *et al*. PRISMA-S: an extension to the PRISMA Statement for Reporting Literature Searches in Systematic Reviews. Journal of the Medical Library Association. 2021; 109: 174–200. https://doi.org/10.5195/jmla.2021.962.

[22] Ouzzani M, Hammady H, Fedorowicz Z, Elmagarmid A. Rayyan—a web and mobile app for systematic reviews. Systematic Reviews. 2016; 5: 210. https://doi.org/10.1186/s13643-016-0384-4.

[23] De la Rosa González A, Guerra-Ojeda S, Camacho-Villa MA, Valls A, Alegre E, Quintero-Bernal R, *et al*. Effect of Probiotics on Gastrointestinal Health Through the Aryl Hydrocarbon Receptor Pathway: A Systematic Review. Foods. 2024; 13: 3479. https://doi.org/10.3390/foods13213479.

[24] Hooijmans CR, Rovers MM, de Vries RB, Leenaars M, Ritskes-Hoitinga M, Langendam MW. SYRCLE'S risk of bias tool for animal studies. BMC Medical Research Methodology. 2014; 14: 43. https://doi.org/10.1186/1471-2288-14-43.

[25] Morikawa D, Muench LN, Baldino JB, Kia C, Johnson J, Otto A, *et al*. Comparison of Preparation Techniques for Isolating Subacromial Bursa-Derived Cells as a Potential Augment for Rotator Cuff Repair. Arthroscopy. 2020; 36: 80–85. https://doi.org/10.1016/j.arthro.2019.07.024.

[26] Baldino JB, Muench LN, Kia C, Johnson J, Morikawa D, Tamburini L, *et al*. Intraoperative and *In Vitro* Classification of Subacromial Bursal Tissue. Arthroscopy. 2020; 36: 2057–2068. https://doi.org/10.1016/j.arthro.2020.03.039.

[27] Muench LN, Kia C, Berthold DP, Uyeki C, Otto A, Cote MP, *et al*. Preliminary Clinical Outcomes Following Biologic Augmentation of Arthroscopic Rotator Cuff Repair Using Subacromial Bursa, Concentrated Bone Marrow Aspirate, and Platelet-Rich Plasma. Arthroscopy, Sports Medicine, and Rehabilitation. 2020; 2: e803–e813. https://doi.org/10.1016/j.asmr.2020.07.019.

[28] Klatte-Schulz F, Bormann N, Voss I, Melzer J, Schmock A, Bucher CH, *et al*. Bursa-Derived Cells Show a Distinct Mechano-Response to Physiological and Pathological Loading *in vitro*. Frontiers in Cell and Developmental Biology. 2021; 9: 657166. https://doi.org/10.3389/fcell.2021.657166.

[29] Steinert AF, Kunz M, Prager P, Göbel S, Klein-Hitpass L, Ebert R, *et al*. Characterization of bursa subacromialis-derived mesenchymal stem cells. Stem Cell Research & Therapy. 2015; 6: 114. https://doi.org/10.1186/s13287-015-0104-3.

[30] Morikawa D, Johnson JD, Kia C, McCarthy MBR, Macken C, Bellas N, *et al*. Examining the Potency of Subacromial Bursal Cells as a Potential Augmentation for Rotator Cuff Healing: An *In Vitro* Study. Arthroscopy. 2019; 35: 2978–2988. https://doi.org/10.1016/j.arthro.2019.05.024.

[31] Landry A, Levy BJ, McCarthy MBR, Muench LN, Uyeki C, Berthold DP, *et al*. Analysis of Time to Form Colony Units for Connective Tissue Progenitor Cells (Stem Cells) Harvested from Concentrated Bone Marrow Aspirate and Subacromial Bursa Tissue in patients Undergoing Rotator Cuff Repair. Arthroscopy, Sports Medicine, and Rehabilitation. 2020; 2: e629–e636. https://doi.org/10.1016/j.asmr.2020.07.013.

[32] Tamburini LM, Levy BJ, McCarthy MB, Kriscenski DE, Cote MP, Applonie R, *et al*. The interaction between human rotator cuff tendon and subacromial bursal tissue in co-culture. Journal of Shoulder and Elbow Surgery. 2021; 30: 1494–1502. https://doi.org/10.1016/j.jse.2020.09.025.

[33] Muench LN, Baldino JB, Berthold DP, Kia C, Lebaschi A, Cote MP, *et al*. Subacromial Bursa–Derived Cells Demonstrate High Proliferation Potential Regardless of Patient Demographics and Rotator Cuff Tear Characteristics. Arthroscopy. 2020; 36: 2794–2802. https://doi.org/10.1016/j.arthro.2020.06.008.

[34] Morikawa D, Hawthorne BC, McCarthy MBR, Bellas N, Johnson JD, Trudeau MT, *et al*. Analysis of Patient Factors Affecting *In Vitro* Characteristics of Subacromial Bursal Connective Tissue Progenitor Cells during Rotator Cuff Repair. Journal of Clinical Medicine. 2021; 10: 4006. https://doi.org/10.3390/jcm10174006.

[35] Morikawa D, LeVasseur MR, Luczak SB, Mancini MR, Bellas N, McCarthy MBR, *et al*. Decreased Colony-Forming Ability of Subacromial Bursa-Derived Cells During Revision Arthroscopic Rotator Cuff Repair. Arthroscopy, Sports Medicine, and Rehabilitation. 2021; 3: e1047–e1054. https://doi.org/10.1016/j.asmr.2021.03.010.

[36] Ko JY, Lian WS, Tsai TC, Chen YS, Hsieh CK, Kuo CW, *et al*. MicroRNA-29a Mitigates Subacromial Bursa Fibrosis in Rotator Cuff Lesion with Shoulder Stiffness. International Journal of Molecular Sciences. 2019; 20: 5742. https://doi.org/10.3390/ijms20225742.

[37] Minkwitz S, Thiele K, Schmock A, Bormann N, Nguyen TH, Moroder P, *et al*. Histological and molecular features of the subacromial bursa of rotator cuff tears compared to non-tendon defects: a pilot study. BMC Musculoskeletal Disorders. 2021; 22: 877. https://doi.org/10.1186/s12891-021-04752-1.

[38] Muench LN, Tamburini L, Kriscenski D, Landry A, Berthold DP, Kia C, *et al*. The Effect of Insulin and Insulin-like Growth Factor 1 (IGF-1) on Cellular Proliferation and Migration of Human Subacromial Bursa Tissue. Arthroscopy, Sports Medicine, and Rehabilitation. 2021; 3: e781–e789. https://doi.org/10.1016/j.asmr.2021.01.019.

[39] Karsmarski OP, Hawthorne BC, Cusano A, LeVasseur MR, Wellington IJ, McCarthy MB, *et al*. Activated Serum Increases *In Vitro* Cellular Proliferation and Growth Factor Expression of Musculoskeletal Cells. Journal of Clinical Medicine. 2022; 11: 3442. https://doi.org/10.3390/jcm11123442.

[40] Hawthorne BC, Engel S, McCarthy MBR, Cote MP, Mazzocca AD, Coyner KJ. Biologic Adjuvants to Rotator Cuff Repairs Induce Anti-inflammatory Macrophage 2 Polarization and Reduce Inflammatory Macrophage 1 Polarization *In Vitro*. Arthroscopy. 2025; 41: 32–41. https://doi.org/10.1016/j.arthro.2024.04.031.

[41] Feng H, He Z, Twomey K, Ilaltdinov AW, Leong D, Wang Y, *et al*. Epigallocatechin-3-gallate suppresses pain-related and proinflammatory mediators in the subacromial bursa in rotator cuff tendinopathy. Discovery Medicine. 2019; 27: 63–77.

[42] Hawthorne BC, Wellington IJ, Sabitsky JT, Murphy KV, Karsmarski OP, Thomas RO, *et al*. Human Rotator Cuff Tears Reveal an Age-Dependent Increase in Markers of Cellular Senescence and Selective Removal of Senescent Cells With Dasatinib + Quercetin Increases Genetic Expression of COL1A1 *In Vitro*. Arthroscopy. 2024; 40: 34–44. https://doi.org/10.1016/j.arthro.2023.05.036.

[43] Muench LN, Kriscenski DE, Tamburini LM, Berthold DP, Rupp MC, Mancini MR, *et al*. Augmenting Suture Tape Used in Rotator Cuff Surgery With Magnesium Increases *In vitro* Cellular Adhesion of Human Subacromial Bursal Tissue. Arthroscopy, Sports Medicine, and Rehabilitation. 2021; 3: e1975–e1980. https://doi.org/10.1016/j.asmr.2021.09.020.

[44] Muench LN, Tamburini L, Kriscenski DE, Berthold DP, Rupp MC, Cote MP, *et al*. The effect of augmenting suture material with magnesium and platelet-rich plasma on the *in vitro* adhesion and proliferation potential of subacromial bursa-derived progenitor cells. JSES International. 2023; 7: 2367–2372. https://doi.org/10.1016/j.jseint.2023.06.027.

[45] Muench LN, Berthold DP, Kia C, Lebaschi A, Cote MP, McCarthy MB, *et al*. Concentrated bone marrow aspirate and subacromial bursa-derived cells demonstrate similar cellular adhesion and proliferation potential on demineralized bone matrix scaffolds for biologic augmentation of rotator cuff repair. Muscles Ligaments Tendons Journal. 2020; 10: 48–56. https://doi.org/10.32098/mltj.01.2020.06.

[46] Lebaschi A, Kriscenski DE, Tamburini LM, McCarthy MBR, Obopilwe E, Uyeki CL, *et al*. Subacromial bursa increases the failure force in a mouse model of supraspinatus detachment and repair. Journal of Shoulder and Elbow Surgery. 2022; 31: e519–e533. https://doi.org/10.1016/j.jse.2022.05.009.

[47] Marshall BP, Ashinsky BG, Ferrer XE, Kunes JA, Innis AC, Luzzi AJ, *et al*. The subacromial bursa modulates tendon healing after rotator cuff injury in rats. Science Translational Medicine. 2024; 16: eadd8273. https://doi.org/10.1126/scitranslmed.add8273.

[48] Dyrna F, Zakko P, Pauzenberger L, McCarthy MB, Mazzocca AD, Dyment NA. Human Subacromial Bursal Cells Display Superior Engraftment Versus Bone Marrow Stromal Cells in Murine Tendon Repair. American Journal of Sports Medicine. 2018; 46: 3511–3520. https://doi.org/10.1177/0363546518802842.

[49] Sun Y, Kwak JM, Kholinne E, Koh KH, Tan J, Jeon IH. Subacromial bursal preservation can enhance rotator cuff tendon regeneration: a comparative rat supraspinatus tendon defect model study. Journal of Shoulder and Elbow Surgery. 2021; 30: 401–407. https://doi.org/10.1016/j.jse.2020.05.025.

[50] Tanimura S, Tokunaga T, Kawakami J, Tian X, Goshogawa H, Tsuyama T, *et al*. Enthesis-related progenitors recruited from the subacromial bursa contribute to rotator cuff healing in rats. Journal of Shoulder and Elbow Surgery. 2026; 35: 1089–1102. https://doi.org/10.1016/j.jse.2025.08.016.

[51] Miura Y, Endo K, Sekiya I. Histological and biochemical changes in a rat rotator cuff tear model with or without the subacromial bursa. Tissue Cell. 2024; 88: 102370. https://doi.org/10.1016/j.tice.2024.102370.

[52] McGuinness LA, Higgins JPT. Risk-of-bias VISualization (robvis): An R package and Shiny web app for visualizing risk-of-bias assessments. Research Synthesis Methods. 2021; 12: 55–61. https://doi.org/10.1002/jrsm.1411.

[53] Dominici M, Le Blanc K, Mueller I, Slaper-Cortenbach I, Marini F, Krause D, *et al*. Minimal criteria for defining multipotent mesenchymal stromal cells. The International Society for Cellular Therapy position statement. Cytotherapy. 2006; 8: 315–317. https://doi.org/10.1080/14653240600855905.

[54] Rios JL, Sapède D, Djouad F, Rapp AE, Lang A, Larkin J, *et al*. Animal Models of Osteoarthritis Part 1-Preclinical Small Animal Models: Challenges and Opportunities for Drug Development. Current Protocols. 2022; 2: e596. https://doi.org/10.1002/cpz1.596.

[55] Codispoti G, Carniato M, Brogini S, Romanelli A, Martini L, Giavaresi G, *et al*. Decellularized biological matrices for the repair of rotator cuff lesions: a systematic review of preclinical *in vivo* studies. Frontiers in Bioengineering and Biotechnology. 2024; 12: 1345343. https://doi.org/10.3389/fbioe.2024.1345343.

[56] Wachsmuth L, Mensen A, Barca C, Wiart M, Tristão-Pereira C, Busato A, *et al*. Contribution of preclinical MRI to responsible animal research: living up to the 3R principle. Magnetic resonance materials in physics, biology, and medicine. 2021; 34: 469–474. https://doi.org/10.1007/s10334-021-00929-w.

[57] Remoleda JL, Khalil M, Gompels LL, Wylezinska-Arridge M, Vincent T, Gsell W. Imaging technologies for preclinical models of bone and joint disorders. EJNMMI Research. 2011; 1: 11. https://doi.org/10.1186/2191-219X-1-11.

[58] Litwak A, Sarabia A, Tamboline M, Mori S, Xu S. Overcoming Low-Field Limitations: High-Quality *Ex Vivo* Soft Tissue Imaging with Compact MRI Systems. Current Protocols. 2026; 6: e70288. https://doi.org/10.1002/cpz1.70288.

[59] Bilgen M. Feasibility and merits of performing preclinical imaging on clinical radiology and nuclear medicine systems. International Journal of Molecular Imaging. 2013; 2013: 923823. https://doi.org/10.1155/2013/923823.

[60] Magnusson SP, Hansen M, Langberg H, Miller B, Haraldsson B, Westh EK, *et al*. The adaptability of tendon to loading differs in men and women. International Journal of Experimental Pathology. 2007; 88: 237–240. https://doi.org/10.1111/j.1365-2613.2007.00551.x.

[61] Daher M, Parmar T, Boufadel P, Zalaquett Z, Fares MY, Abboud JA. Gender Disparities in Shoulder Pain and Shoulder Surgery: A Current Concepts Review. Journal of Clinical Medicine. 2026; 15: 1886. https://doi.org/10.3390/jcm15051886.

[62] Smith KM, Hotaling JM, Presson AP, Zhang C, Horns JJ, Cannon-Albright LA, *et al*. The Effect of Sex Hormone Deficiency on the Incidence of Rotator Cuff Repair: Analysis of a Large Insurance Database. Journal of Bone and Joint Surgery American Volume. 2022; 104: 774–779. https://doi.org/10.2106/JBJS.21.00103.

[63] Gruber MS, Bischofreiter M, Brandstätter P, Hochreiter J, Sadoghi P, Ortmaier R. Age- and Gender-Related Differences in the Morphology of Cuff Tear Arthropathy: A Cross Sectional Analysis. Journal of Functional Morphology and Kinesiology. 2023; 8: 8. https://doi.org/10.3390/jfmk8010008.

[64] Daniels SD, Stewart CM, Garvey KD, Brook EM, Higgins LD, Matzkin EG. Sex-Based Differences in Patient-Reported Outcomes After Arthroscopic Rotator Cuff Repair. Orthopaedic Journal of Sports Medicine. 2019; 7: 2325967119881959. https://doi.org/10.1177/2325967119881959.

[65] Fancher AJ, Mok AC, Vopat ML, Templeton K, Kimbrel BK, Tarakemeh A, *et al*. Comparing Sex-Specific Outcomes After Rotator Cuff Repair: A Meta-analysis. Orthopaedic Journal of Sports Medicine. 2022; 10: 23259671221086259. https://doi.org/10.1177/23259671221086259.

[66] Sheth VH, Shah NP, Jain R, Bhanushali N, Bhatnagar V. Development and validation of a risk-of-bias tool for assessing *in vitro* studies conducted in dentistry: The QUIN. Journal of Prosthetic Dentistry. 2024; 131: 1038–1042. https://doi.org/10.1016/j.prosdent.2022.05.019.

[67] Hartung T, Hoffmann S, Whaley P. Assessing risk of bias in toxicological studies in the era of artificial intelligence. Archives of Toxicology. 2025; 99: 3065–3090. https://doi.org/10.1007/s00204-025-03978-5.

[68] Svendsen C, Whaley P, Vist GE, Husøy T, Beronius A, Consiglio ED, *et al*. Protocol for designing INVITES-IN, a tool for assessing the internal validity of *in vitro* studies. Evidence-Based Toxicology. 2023; 1: 1–15. https://doi.org/10.1080/2833373x.2023.2232415.

[69] Paiva Barbosa V, Bastos Silveira B, Amorim Dos Santos J, Monteiro MM, Coletta RD, De Luca Canto G, *et al*. Critical appraisal tools used in systematic reviews of *in vitro* cell culture studies: A methodological study. Research Synthesis Methods. 2023; 14: 776–793. https://doi.org/10.1002/jrsm.1657.

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