

# Ultrastructure of the subacromial bursa in painful shoulder syndromes

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Summary. In order to determine if inflammation of the subacromial bursa or "bursitis" is as common an occurrence as believed in painful conditions of the shoulder, eight bursae were obtained during surgery from 3 cases of calcifying tendinitis, 2 of tight coracoacromial ligament, 2 of rotator cuff tear and 1 of acromioclavicular osteoarthritis. In the cases of calcifying tendinitis, tight coracoacronial ligament and rotator cuff tear the morphological changes mainly consisted of a numerical increase in cells throughout the bursal wall along with proliferation of endothelial cells in the vascular channels. All of the cells including the endothelial had densely packed, intermediate type filaments in their cytoplasm but no appreciable diminution of metabolic organelles. Lipid droplets were abundant in the extracellular connective tissue of the bursae from the cases with rotator cuff tear. In contrast to the other cases, the bursa from the case of acromioclavicular osteoarthritis showed widespread fibrin deposition in association with cell necrosis. In none of the cases did inflammatory leukocytic cells infiltrate bursal tissue.

We conclude, that the subacromial bursa tends to undergo proliferative or degenerative changes in rotator cuff tendinopathies but bursal inflammation with polymorphonuclear cell infiltrate does not occur commonly.

Key words: Subacromial bursa – Ultrastructure of Bursa – Bursitis

#### Introduction

From a clinical point of view, the subacromial bursa is the most important deep bursa of the body (Bywaters 1979). This large bursa is situated beneath the deltoid muscle, the acromion and the coracoacromial ligament, and

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it overlies the rotator cuff tendons especially the supraspinatus. It usually does not communicate with the glenohumeral joint.

Inflammation of the subacromial bursa is often implicated as a source of shoulder pain, and "bursitis" is an all too frequently used diagnosis even when a primary disease of a contiguous structure is evident (Bland et al. 1977). Indeed, subacromial bursitis is deemed to be synonymous with various rotator cuff tendinitides (Simon 1975; Buckingham 1981). The term "bursitis", denotes either clinically discernible effusion in the bursal cavity, or, in the absence of fluid accumulation, distinct pathological changes indicating inflammation of the saccular wall (Canoso 1981). The pathological alterations, however, are seldom examined in cases where the painful shoulder has been treated surgically and there is a clear lack of information about the bursa at the ultrastructural level.

The present study was undertaken to delineate the ultrastructure of the subacromial bursa in eight painful shoulder conditions. In all of these cases, the bursa was not implicated as the primary site of pain in the preoperative diagnosis but its secondary involvement was not excluded.

## Materials and methods

Biopsies of the subacromial bursa were obtained during surgery. Altogether, tissues from 8 cases: 3 of calcifying tendinitis; 2 with tight coracoacromial ligament; 2 with tear of the rotator cuff and 1 suffering from acromioclavicular osteoarthritis, were examined. The age, sex and occupation of those patients are listed in Table 1, along with duration of symptoms, the preoperative diagnosis and gross appearance of the bursa observed at the time of surgery.

Table 1.

| Case     | Occupation          | Duration<br>of<br>symptoms | Pre-op<br>cortisone | Pre-op<br>diagnosis                      | Appearance<br>of the bursa<br>during surgery |
|----------|---------------------|----------------------------|---------------------|--|--|
| GB, 41 ♀ | Nurse               | 5 years                    | None                | Calcifying tendinitis                    | Unremarkable                                 |
| RB, 44 ♂ | Administrator       | 10 days                    | None                | Same                                     | Unremarkable                                 |
| JR, 37 ♂ | Computer technician | 1 year                     | None                | Same                                     | Unremarkable                                 |
| JP, 28 ♂ | Mechanic            | 6 months                   | 2                   | Tight<br>coracoacromial<br>ligament      | Unremarkable                                 |
| RL, 59 ♀ | Nun                 | 2 years                    | 3                   | Same                                     | Thickened and edematous                      |
| JS, 35 ♂ | Mover               | 5 years                    | None                | Rupture rotator cuff                     | Somewhat<br>thickened                        |
| JM, 59 ♀ | Housewife           | 1 year                     | 2                   | Same                                     | Unremarkable                                 |
| RL, 34 3 | Carpenter           | 1 year                     | 2                   | Acromio-<br>clavicular<br>osteoarthritis | Thick and reddened with villous surface      |

The number of cortisone injections which several patients received preoperatively at the site of pain has also been mentioned in Table 1.

A few fragments from each specimen were fixed in 10% neutral buffered formalin and embedded in paraffin. Sections were stained with hematoxylin and eosin, Masson's trichrome and von Kossa stains.

The rest of the specimen was processed for electron microscopy. Tiny pieces of tissues were fixed in one-half strength Karnovsky's fixative for 2–4 h at 4° C, washed in 0.1 m sodium cacodylate buffer and post-fixed in 1% osmium tetroxide. The pieces were dehydrated serially in graded ethyl alcohol and finally in propylene oxide before being embedded in epon-araldite. Thick sections were stained with toluidine blue while thin sections for electron microscopical use were stained with uranyl acetate and lead citrate.

To establish the morphological appearance of a normal subacromial bursa, specimens were taken during autopsy when no history of shoulder ailment was found in the patient's chart. Altogether 67 bursae from postmortem cases, ranging in age from a 5-month old infant to persons in their eighties, were fixed, embedded in paraffin and stained routinely for light microscopy. None of the specimens was processed for electron microscopy as the autopsies were done 6–8 h or longer after death.

## Results

We found during surgery as well as in postmortem cases that the subacromial bursa was clearly demarcated from the overlying muscle but was firmly attached to the peritendinous connective tissue underneath. A plane of cleavage between the bursa and the epitenon of the supraspinatus was not discernible either grossly or histologically.

The light microscopic examination of the normal bursa often demonstrated two clearly distinguishable zones. The superficial zone which was at places thrown into folds, was a narrow compact band of connective tissue containing one or more layers of cells in pseudostratification (Fig. 1). The cells were usually ovoid with their long axis parallel to the surface. The deep zone consisted of loose connective tissue containing cells which were broadly similar to those in the superficial zone but showing more variations in size and shape. Vascular channels were scattered in this zone.

For the sake of convenience, the gross and microscopic appearances of the subacromial bursa in each disease condition are described separately.

# Calcifying tendinitis

The duration of symptoms among 3 patients ranged from 10 days to 5 years; the subacromial bursa, however, appeared during surgery to be unremarkable in all the cases. None of these patients had received local injections of corticosteroids preoperatively (Table 1).

Histologically, the zonal disposition of the bursa was similar to that in the normal specimens. The cellular component, however, appeared increased and many of the vascular channels were engorged. In a 44-year old male who had only 10 days of acute pain before he was operated for removal of calcium, the bursa showed microscopic foci of calcium (Fig. 2) which were not detected by preoperative radiograms. The bursal tissue surrounding calcific deposits was devoid of inflammatory cell infiltrate.

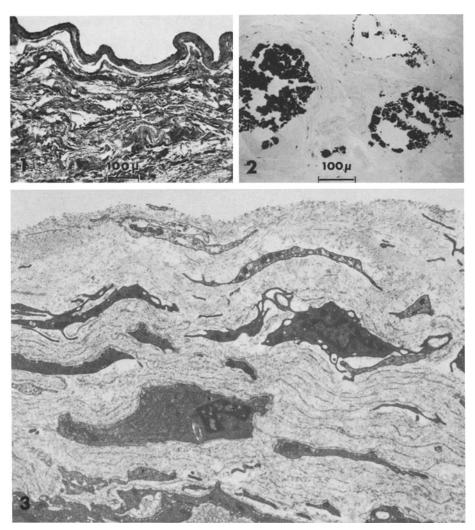
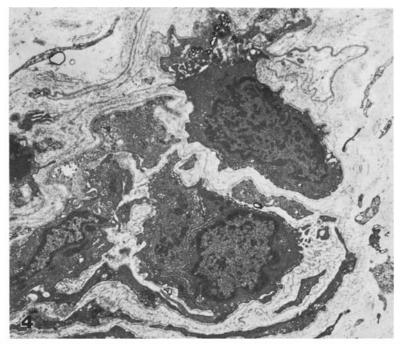


Fig. 1. The bursa obtained from a 45-year old male who died accidentally, shows a compact band of superficial zone with cells in pseudostratification and a deep layer of loose connective tissue. Hematoxylin and eosin

The magnifications of light micrographs are indicated on the prints. The electron micrographs are from sections which were stained with uranyl acetate and lead citrate.

Fig. 2. Microscopic foci of calcification are present in the bursa of a patient with calcifying tendinitis. The bursal tissue around calcific deposits is devoid of inflammatory infiltrate. Von Kossa

Fig. 3. In a grossly unremarkable bursa from a case of calcifying tendinitis, both Type A and Type B cells are seen in the adjacent region of the surface.  $\times$  8,160



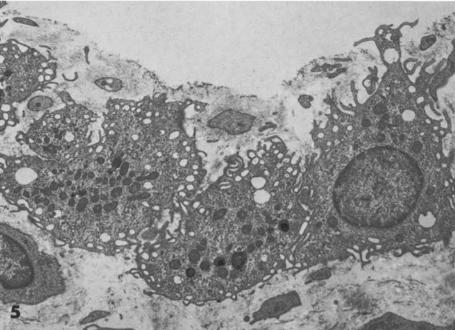


Fig. 4. In a bursa from a case of calcifying tendinitis, the basal lamina of a contorted vascular channel is in multiple layers and forming redundant folds.  $\times 7,200$ 

Fig. 5. In a grossly edematous bursa from a case with tight coracoacromial ligament, the superficial zone shows Type A cells in a row.  $\times 8,100$ 

The demarcation between superficial and deep zones were not conspicuous in tissues processed for electron microscopy. The ultrastructure of the superficial zone consisted of transversely oriented cells and cell processes embedded in collagenous and microfibrillar connective tissue (Fig. 3). The cells corresponded to A and B types of synovial cells, the former primarily characterized by filopodia, cytoplasmic vacuoles and many mitochondria, while the latter had well-developed cisternae of rough endoplasmic reticulum. The cells in the deep zone were larger, and tended to demonstrate a combination of features from both types of cells. Intermediate-type filaments were abundant in the cytoplasm, and the nuclei generally had an irregular contour.

The enlarged endothelial cells of vascular channels showed marked irregularity in their size and shape. There was proliferation of the basal lamina which was generally multilayered forming redundant folds (Fig. 4). Pericytes were prominent. The intercellular space was mainly occupied by collagen fibres in random arrangement, inconspicuous strands of microfibrils and small clumps of amorphous débris.

## Tight coracoacromial ligament

Of the 2 cases, the 28-year old male patient had a relatively short duration of symptoms for 6 months, but had local corticosteroid injections twice before coming to surgery. The subacromial bursa was grossly unremarkable. In contrast, the bursa in the older patient, a 59-year old female who had 3 steroid injections during a 2-year period of pain in her right shoulder, appeared thickened and edematous (Table 1). Histologically, the deep zone of some fragments from the edematous bursa had a myxoid appearance. Ultrastructurally, although the cellular characteristics were broadly similar in two specimens, they were accentuated in the edematous bursa. Type A cells were prominent in the superficial zone (Fig. 5). Intermediate filaments were abunadent in the cell cytoplasm (Fig. 6), and an occasional cell demonstrated a pair of centrioles. The nuclei of several cells contained a fibrous lamina along the inner membrane. The endothelial cells showed marked proliferation and enlargement, often obliterating the lumen (Fig. 7).

## Rotator cuff tear

The younger male patient with a longer duration of symptoms showed a somewhat thickened bursa during surgery, while that of the older patient who had 2 steroid injections in 1 year, was unremarkable (Table 1). Histologically, lipid globules were abundant at and beneath the superficial zone in both the cases (Fig. 8), and those were easily recognizable with the electron microscope. The cellular features were similar to what we have already described above, including those of nuclei showing a fibrous lamina along the inner membrane.

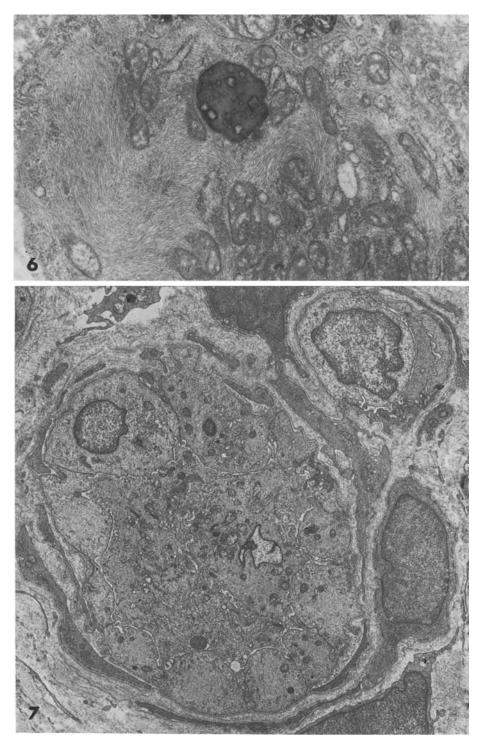
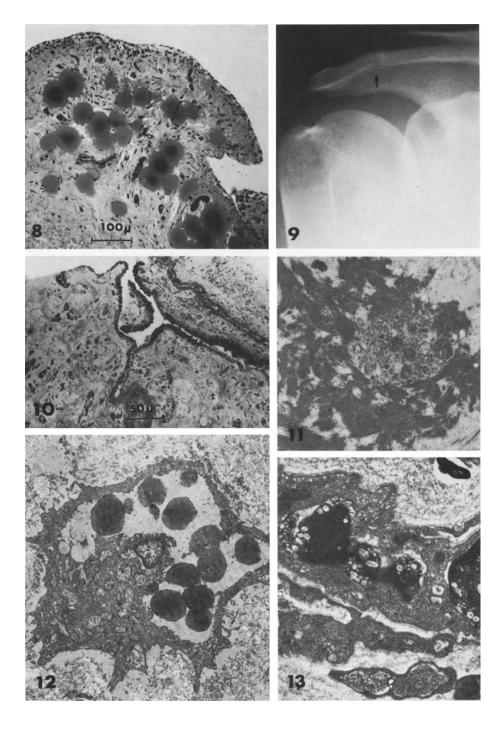


Fig. 6. The cells in the same bursa show abundant intermediate type filaments along with many mitochondria.  $\times 25,500$ 

Fig. 7. The lumen of a vascular channel is obliterated by proliferating endothelial cells. The cytoplasm of these cells contains abundant intermediate filaments.  $\times 7,600$ 



## Acromioclavicular osteoarthritis

Radiograms cleary showed a narrowing of the acromioclavicular cartilage space and marginal osteophyte formation (Fig. 9) in the right shoulder of a 34-year old male who had a 1-year history of pain for which he was given 2 steroid injections (Table 1). At surgery, the bursa looked thick and reddened with a rough surface. The corresponding appearance, histologically, consisted of a thick coating of fibrinoid material on villous protuberances alternating with deep clefts (Fig. 10). There were areas of the superficial zone which were relatively acellular. Deposits resembling fibrin appeared to entrap cell processes (Fig. 11), and many cells showed degenerative changes (Fig. 12). The bursal and endothelial cells otherwise showed features as we have already described. The lumen of one vessel appeared to be plugged by amorphous electron-dense substance containing numerous vesicular bodies (Fig. 13).

## Discussion

In the normal bursa, the surface is covered by a single layer of cells which overlie a zone a of connective tissue containing vascular channels (Rubens-Duval 1972). The cells in the bursa are similar to those in the synovium in which two main cell types have been described. Type A or M cells show the features of a phagocyte which has long filopodia on the surface and vacuoles and lysosomal bodies in the cytoplasm. Golgi apparatus is prominent in the paranuclear region. Type B or F cells, on the other hand, resemble fibroblasts. They contain well-developed cisternae of rough endoplasmic reticulum (Barland et al. 1962; Ghadially 1978). Frequently, however, a number of cells show features of both the types of cells, and these "intermediate" cells contain rough endoplasmic reticulum as well as the Golgi apparatus (Ghadially 1978). The cells of subacromial bursae in our study showed

- Fig. 8. In a bursa from a case of rotator cuff rupture, lipid droplets of various sizes are clustered in the bursal tissue. Toluidine blue
- Fig. 9. Radiograph of the shoulder of the patient with acromioclavicular osteoarthritis shows narrowing of the aeromioclavicular cartilage space and marginal osteophyte formation (arrow)
- Fig. 10. The bursa from the case of acromioclavicular osteoarthritis shows villous protrusions on the surface alternating with clefts. The entire surface is covered by a layer of fibrinoid material. Toluidine blue
- Fig. 11. Beneath the surface of the same bursa, a cellular process with barely recognizable organelles is trapped within fibrin deposits.  $\times 17,000$
- Fig. 12. The cytoplasm of a degenerative cell contains inclusions, possibly of lipidic nature.  $\times 7{,}600$
- Fig. 13. The lumen of a vessel is plugged by amorphous electron-dense substance which contains numerous vesicular bodies.  $\times$  14.400

predominant features of either Type A or Type B, but more commonly a combination of the two.

From the various changes undergone by the subacromial bursa that we have described, it can be concluded that the morphological alterations could be characterized as either proliferative or degenerative. The proliferative changes were seen primarily in cases of calcifying tendinitis, tight acromioclavicular ligament and rotator cuff tear. There was a numerical increase of cells in both the superficial and deep zones along with that of endothelial cells in vascular channels. Some of the ultrastructural features of these proliferating cells were difficult to interpret. The densely packed intracytoplasmic microfilaments were found within bursal as well as endothelial cells. An excessive deposition of filaments has been considered a sign of degeneration in cartilage cells when accompanied by paucity of organelles (Ghadially 1975). The cells in the present study, however, had a large number of metabolic organelles including the Golgi apparatus along with the microfilaments. An occasional cell would even contain a pair of centrioles which are never a feature of a degenerative cell. Furthermore, some of the bursal cells demonstrated a thick fibrous lamina along the inner nuclear membrane. This is believed to occur in cells which are in the process of repair and has been described in synovial cells from a case of chronic synovitis with hyperplasia of synovial membrane (Ghadially et al. 1974).

The degenerative changes were pervasive in the bursa obtained from the patient with acromioclavicular osteoarthritis. Fibrinoid deposits effaced cells in the superficial zone. Many cells in the deep zone were overtly necrotic. Lipid inclusions within cytoplasm, vacuolation of endoplasmic reticulum and of mitochondrial matrix were regularly seen in most of the cells. The ultrastructural characteristics of proliferating cells found, for example, in cases of calcifying tendinitis were infrequently present.

It can be concluded, then, that painful shoulder syndromes primarily affecting structures such as rotator cuff tendons, acromioclavicular joint or coracoacromial ligament, would alter the ultrastructure of the subacromial bursa, probably because of anatomical proximity. None of the bursae had leukocytic infiltration of the saccular wall, even in those which appeared edematous and congested during surgery. Equally devoid of inflammatory cells was the bursa containing multiple calcific foci, although it is generally believed that calcium rupturing into the bursa from a contiguous tendon produces acute bursitis (Ghormley 1939). Bywaters (1965) has rightly emphasized the importance of bursae in the causation of symptoms. The term "bursitis", however, is so vaguely defined that refinement of both clinical and morphological criteria is necessary to improve diagnostic relevance.

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