Inframammary Fold: A Histologic Reappraisal

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The inframammary fold is a defining element in the shape and structure of the female breast. It should be preserved whenever possible in ablative procedures and recreated accurately when the breast is reconstructed after mastectomy. To date, no accurate anatomic description of this essential structure exists. Previous studies have suggested that the fold is produced by a supporting ligament running from the dermis in the fold region to a variety of locations on the rib cage. This clinic's experience with mastectomy, augmentation mammaplasty, and breast reconstruction does not support the existence of a ligamentous structure. To define the structure of the inframammary fold, 10 female and 2 male cadavers were studied. The anterior chest wall was removed en bloc and frozen in orthostatic position. Parasagittal sections were made of the inframammary fold with the chest wall intact. After decalcification of the ribs and routine histologic preparation, thin sections were stained with Gomori's trichrome. On light microscopic examination, no demonstrable ligamentous structure of dense regular connective tissue could be identified in the fold region in any of the 12 specimens. Superficial and deep fascial layers were uniformly observed anterior to the pectoralis major and serratus anterior muscles. The superficial fascia was connected to the dermis in the fold region in a variety of configurations. In some cases, the deep fascia fused with the superficial fascia and dermis at the fold level. In other cases, bundles of collagen fibers arising from the superficial fascial layer were found to insert into the dermis at the inframammary fold, slightly inferior to it, or both. These bundles were observed consistently in sections from the sternum to the middle axillary line. They were distinct from Cooper's suspensory ligaments, which are seen more superiorly in the glandular tissue. (*Plast. Reconstr. Surg.* 105: 549, 2000.)

The inframammary fold is a defining element in the shape and structure of the female breast. The fold is undetectable in the prepubescent breast, but with the onset of puberty, the inframammary fold begins to define the inferior extent of the breast. From the onset of breast development, the inframammary fold anchors the inferior pole of the breast to the chest wall, and with age, the breast begins to sag or becomes ptotic relative to this point. This structure has paramount importance in both aesthetic and reconstructive surgery of the breast, with considerable attention paid to its role in determining the technique used, in the difficulty in creating a natural-appearing inframammary fold,[1–4] and in the use of "skin-sparing" mastectomies to preserve the structure.[5–9] In augmentation mammaplasty, the inframammary fold provides a relatively well-hidden site for an incision to place a mammary implant. However, it is also responsible for the "double bubble" phenomenon seen in patients with ptosis who undergo transaxillary subpectoral augmentation.[10]

Descriptions of the inframammary fold abound, beginning with Sir Astley Cooper's[11] description in 1845. He stated that at "the abdominal margin, the gland is turned upon itself at its edge, and forms a kind of hem." Since that time, several studies have attempted to define its composition. To date, there is still not a complete anatomic description of this important structure, and it is essentially ignored in comprehensive breast textbooks.[12,13] Previous studies have focused on either gross anatomic observation after blunt dissection techniques,[14] blunt dissection followed by histologic confirmation of the connective tissue nature of the structure,[15] or gross dissection followed by histologic confirmation.[16] All of these methods introduce dissection artifact into the description of this structure.

However, our clinical experience with both augmentation mammaplasty and breast reconstruction after mastectomy does not support

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Received for publication February 24, 1999; revised June 28, 1999.

Presented at the American Association of Clinical Anatomists Annual Meeting, in Lexington, Kentucky, in June of 1998, and the Plastic Surgery Research Council Meeting, in Pittsburgh, Pennsylvania, in May of 1999.

the existence of a distinctive ligamentous or connective tissue structure defining the inframammary fold. In preliminary studies on fresh cadaver material, we could not demonstrate the presence of a ligamentous structure. The purpose of our study is to characterize the anatomic structures responsible for the formation of the inframammary fold without the introduction of dissection artifact. We hypothesize that the inframammary fold represents a fusion of superficial fascial layers over the chest wall between which the breast develops. The force of gravity acting on the breast over this relatively fixed structure is responsible for ptosis of the breast. Our objectives included histologically identifying the infrastructure of the inframammary fold, determining the relationship of the fold to the pectoralis muscles, and identifying the level of the fold relative to the costal cartilages and ribs.

Materials and Methods

Eight fresh (unembalmed) female cadavers, two embalmed female cadavers, and two fresh (unembalmed) male cadavers were included in the study. The average age was 77 ± 15 years. The cadavers were obtained from the University of Louisville Fresh Tissue Laboratory.

The anterior chest wall of each cadaver was removed by means of en bloc resection. The incision boundaries included the midsternum medially, the second rib superiorly, the midaxillary line laterally, and the costal margin inferiorly. The pleura and mediastinum were left intact. Once removed, the chest wall segment was attached to a board with a hand drill (Stryker, Santa Clara, Calif.) and Kirschner wires and placed into an orthostatic position (to recreate the natural inframammary fold with gravity) inside a freezer at 0°F. Several days later, a band saw was used to section the frozen specimen into 10 equidistant slices in the sagittal plane perpendicular to the chest wall. The most medial section was designated as slice 1, the second section as slice 2, and so on. Next, a variable-speed hand drill (Stryker) was used to remove the cancellous bone inside the rib, being careful not to disrupt the periosteum. The lateral edges of the slices were then sketched for later reference, and the extraneous tissue (35 mm above and 35 mm below the inframammary fold) was removed.

An initial series (specimens 398 and 399) was set up to determine the appropriate length for fixation and paraffin embedding. Immediately after the sketching was finished, the tissue slices were placed in a neutral buffered formalin for 2 days of tissue fixation. Then, the slices were transferred to a decalcifier (Cal-Ex, Fisher Scientific, Hampton, N.H.) for 12 to 15 weeks of bone decalcification. The use of the large glass basins allowed for maintaining the order of the slices and decalcifying the bone with large volumes of decalcifier. After the ribs were tested for sufficient decalcification with a sharp probe, the tissue was transferred to a series of dehydration and clearing agents: (1) 1 day in 80% ethanol, (2) 1 day in 95% ethanol, (3) 1 day in 100% ethanol, (4) 4 hours in xylene bath 1, (5) 4 hours in xylene bath 2, and (6) overnight in xylene bath 3. To completely remove the xylene from the tissue, several paraffin baths were used before the final embedding medium was incorporated. The tissue was transferred to (1) warm paraffin bath 1 for 4 hours, (2) warm paraffin bath 2 for 4 hours, and (3) warm paraffin bath 3 for 4 hours. Finally, large weighing boats numbered 1 through 10 were used to embed the individual slices in embedding paraffin. Once the paraffin blocks had hardened, a sliding microtome was used to thin-section the individual slice 40 μm thick and 70 mm long. The slides were rehydrated in a series of ethanols and xylenes and immersed in Gomori's trichrome, which stains collagen green, muscle red, and nuclei blue.

Once the initial specimens were sectioned, it was determined that longer fixation time was needed. Similarly, a vacuumed oven was used to facilitate the embedding procedure. Specimens 401 through 418, 527, and 615 followed the improved protocol. Immediately after the sketching was finished, the tissue slices were placed in neutral buffered formalin for 10 days of tissue fixation. Decalcification, dehydration, and clearing procedures were followed as described for the initial series. For paraffin infiltrating, the tissue was transferred to (1) warm paraffin bath 1 for 4 hours, (2) warm vacuum paraffin bath 2 for 4 hours, and (3) warm vacuum paraffin bath 3 overnight. The remainder of the preparation was as described for the initial series.

Because the study was morphologic in nature and did not include significant numbers of comparative groups, there was no quantitative statistical analysis. The study used light and dissecting microscopes to examine the fascial organization of the inframammary fold and the surrounding region. The relationship of the fold to the neighboring costal cartilages and ribs, the thoracic musculature, and the superficial and deep fascias underneath the dermis was recorded and any inconsistencies were noted. The pathways and structural components of the fascial layers in the fold region were also examined and recorded to determine whether a ligament was present. Finally, the structural differences in female and male cadavers were compared.

RESULTS

Anatomic Observations

The inframammary fold and related muscles were identified in each of the sagittal sections of the anterior chest wall. Pectoralis major, rectus abdominis, and serratus anterior muscles were always present between the inframammary fold and the rib cage. Among the 10 female cadavers, there was variability in the fold level relative to the rib cage (Tables I and II). Along its course, the fold was positioned superficial to the anterior arch level of the fifth, sixth, seventh, or eighth ribs. The medial origin of the fold was most often overlying the costal cartilage level of the fifth or sixth rib. The lateral extent was typically related to the seventh or eighth rib near the anterior axillary line. The midclavicular point of the fold, inferior to the areola and nipple, was observed at the sixth or seventh rib level.

Among the 12 cadavers (male and female), there was variability in the borders of the pectoralis major relative to the rib cage. The inferior and lateral borders of the pectoralis major were positioned superficial to the anterior arch of the fourth, fifth, sixth, or seventh ribs. The serratus anterior muscle inserted into the upper eight ribs. No predictable relationships between the inferior and lateral borders of the pectoralis major, the inframammary fold, or a specific rib were observed (Tables I and II).

Histologic Observations

On examination under dissecting and light microscopes, muscle, fascial layers, dermis, adipose tissue, and epidermis were identified in each sagittal section. A deep investing fascial layer was uniformly observed on the anterior surface of the pectoralis major muscle (medial sections 1 through 7) and serratus anterior muscle (lateral sections 8 through 10). Superficial fascia was identified as the connective tissue between the dermis and the deep fascia. It contained variable amounts of adipose tissue. In the 10 female specimens, the adipose tissue and glandular elements of the developed breast divided the superficial fascia into two layers. The more superficial division, also known as Camper's fascia, continued subcutaneously anterior to the glandular tissue, whereas the deeper division, also known as Scarpa's fascia, continued posterior to the glandular tissue. Camper's layer was composed mainly of adipose tissue. Scarpa's fascia, or the deeper membranous layer, in some cases was contiguous with the deep investing fascia.

There was no breast tissue identified in the inframammary fold region. The tissues consisted of epidermis, dermis, superficial fascia, and adipose tissue of varying thickness. The superficial and deep fascial layers were connected to the dermis in the fold region in a variety of configurations. In 2 of the 10 female specimens, the deep fascia fused with the su-

TABLE I

Relationships of the Inframammary Fold to the Pectoralis Major Muscle and the Ribs

Cadaver No.	Inferior Border of Pectoralis Major on Left Breast	Fold Level on Left Breast	Inferior Border of Pectoralis Major on Right Breast	Fold Level on Right Breast
350	No sketches	No sketches	No sketches	No sketches
398	Rib 5	Male, no fold	Ribs 4–5	Male, no fold
399	Ribs 4–5	Ribs 4–5	Rib 4	Ribs 5–6
401	Ribs 4–5	Ribs 5–7	Ribs 4–5	Ribs 5–7
407	Ribs 4–5	Ribs 5–6	Ribs 4–5	Ribs 6–7
409	Ribs 5–6	Ribs 6–7	Ribs 5–7	Rib 7
411	Ribs 5–6	Ribs 7–8	Ribs 5–6	Ribs 7–8
412	Ribs 4–6	Ribs 5–6	Ribs 4–5	Rib 6
416	Ribs 5–6	Ribs 7–8	Rib 6	Ribs 7–8
418	Rib 5	Male, no fold	Ribs 5–6	Male, no fold
527	Ribs 5–7	Small breast	No breast	No breast
615	Rib 5	Rib 6	Ribs 5–6	Rib 6

TABLE II
Origin of the Inframammary Fold in Relation to the Ribs

Cadaver No.	Medial Origin of Fold on Left Breast	Lateral Origin of Fold on Left Breast	Medial Origin of Fold on Right Breast	Lateral Origin of Fold on Right Breast
350	No sketches	No sketches	No sketches	No sketches
398	Male, no fold	Male, no fold	Male, no fold	Male, no fold
399	Rib 5	Rib 8	Rib 6	Rib 6
401	Rib 5	Rib 8	Rib 5	Rib 7
407	Rib 5	Rib 7	Rib 6	Rib 7
409	Rib 7	Rib 7	Rib 7	Rib 7
411	Rib 7	Rib 8	Rib 7	Rib 9
412	Rib 5	Rib 5	Rib 6	Rib 6
416	Rib 7	Rib 8	Rib 7	Rib 8
418	Male, no fold	Male, no fold	Male, no fold	Male, no fold
527	Small breast	Small breast	No breast	No breast
615	No medial origin	No lateral origin	No medial origin	No lateral origin

perficial fascia and dermis at the level of the inframammary fold (Fig. 1). These two cadavers had a thin subdermal adipose layer between the dermis and muscle, approximately 1 to 2 mm in width. In 6 of the 10 female specimens, bundles of collagen fibers arose from the superficial and deep layers of the superficial fascia and obliquely traversed the adipose tissue to insert into the dermis, either at the fold level or slightly inferior to it (Fig. 2). The subdermal adipose layer for these cadavers was much thicker, with an average width of 9.2 mm and a range of 3 to 22 mm. Two of the female cadavers were extremely thin unembalmed specimens. No clear delineation in the tissues was possible. In the two male specimens, colla-

FIG. 1. Fusion of superficial and deep fascia with the dermis at the fold level (specimen 350). Gomori's trichrome stain: collagen = green, muscle = red, nuclei = blue. IF, inframammary fold.

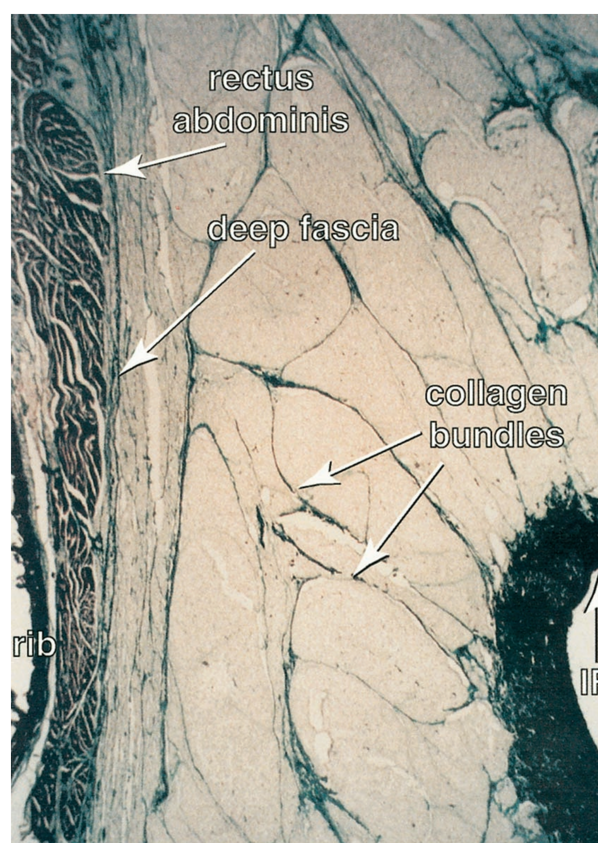

FIG. 2. Collagen fibers from the superficial fascia inserting into the dermis at, or inferior to, the fold level (specimen 416). Gomori's trichrome stain: collagen = green, muscle = red, nuclei = blue. IF, inframammary fold.

gen fibers arose from the superficial fascia, traversed the adipose tissue, and inserted into the dermis. This course was consistent with the vertical septa as described by Lockwood[17] or with retinacula cutis.[18]

The bundles were observed consistently along the fold from midsternum to the midaxillary line. They were separate from the Cooper's suspensory ligaments, which are seen more superiorly in the glandular tissue. No demonstrable ligamentous structure of dense regular connective tissue was identified in the inframammary fold region in any of the 12 specimens.

It should be noted that a few parasagittal sections from each cadaver, those closest to the sternum with a large percentage of bone, either dissolved in the decalcifying solution or shrank in the paraffin baths. They were not included in the study.

Discussion

The inframammary fold is the defining shelf for the inferior pole of the female breast. Embryologically, little is known regarding the specific formation of the connective tissue in the region of the inframammary fold. Breast development begins during the fifth or sixth week of development, when outgrowths from the ectodermal skin layer penetrate into the underlying mesoderm. This outgrowth forms the mammary ridge or milk line. Between the second and fourth months, all of the ectodermal thickenings along the mammary line regress, except for two in the region of the third or fourth ribs. During the fifth month, the ectoderm continues to extend into the underlying mesoderm, and a branching network forms that will eventually become the lactiferous system. The surrounding mesenchyme starts to develop into the supportive connective and adipose tissue of the breast. This development will eventually become the collagen bundles in the fold region. Once puberty begins with proliferation of glandular and adipose tissue, the breast becomes a pendent structure with its inferior border defined by the inframammary fold.[19,20]

By using blunt dissection techniques followed by gross observation, Maillard and Garey[14] described a crescent-shaped ligamentous band stretching from the anterior surface of the pectoralis muscle to the inframammary skin fold. They defined this band as the "prepectoral ligament" and suggested that it was responsible for formation of the fold by supporting the breast as a sling.

Bayati and Seckel[15] also used blunt dissection to identify what was thought to be a ligamentous structure. They believed it arose as a condensation of rectus abdominis fascia medially and fascia of the external oblique and serratus anterior laterally. They named this structure the "inframammary crease ligament" and suggested that this ligament originated medially from the fifth rib periosteum and laterally from the fascia between the fifth and sixth ribs, which inserted into the deep dermis of the inframammary skin fold.

Van Straalen et al.[16] investigated the inframammary fold in a series of female-to-male transsexuals who were undergoing subcutaneous mastectomy. They identified a subcutaneous, dense fibrous strand that followed the margin of the inframammary fold and extended from the sternum to the lateral margin of the pectoralis major muscle at the preaxillary fold, in contrast to Maillard and Garey,[14] who suggested that the ligament was oriented transversely across the chest wall.

The most careful study of the inframammary fold, to date, was that of Garnier et al.,[21] who used gross anatomic, radiographic, ultrasonographic, and histologic approaches. Radiographically, the inframammary fold was found, in the majority of cases, to correspond to the anterior arch of the sixth rib. Their histologic examination showed fibers passing from the prepectoral (deep) fascia to the superficial fascia, but they did not find a connection between the superficial fascia and the dermis of the fold. Thus, they concluded that a ligamentous structure was not present in the fold region, contradicting the previous three studies.

Some of the controversy may arise from the differing interpretations of the structure of a ligament. Several definitions are currently accepted. *Dorland's Illustrated Medical Dictionary*[18] defines a ligament in three manners: (1) a band of fibrous tissue that connects bones or cartilages, serving to support and strengthen joints; (2) a double layer of peritoneum extending from one visceral organ to another; or (3) cordlike remnants of fetal tubular structures that are nonfunctional after birth. The usual histologic description of a ligament is a form of dense regularly arranged fibrous connective tissue. Weiss[22] defined a ligament as "collagen fibers which are thick, long, and grouped into large parallel bundles to provide the required tensile strength."

Gray[23] defined a ligament as bundles "composed mainly of collagenous fibers oriented in parallel, or closely interwoven to present a white shining, iridescent appearance." He commented on the similarities of ligaments, tendons, and aponeuroses but noted that a tendon is connected directly to musculature, whereas an aponeurosis is simply a flattened, tendinous sheet. Fascia was also included in the family of dense fibrous connective tissue, but it had far less organization than the other three. Weibrecht[24] attempted to identify every ligament in the human body by gross dissection. In his review, he defined a ligament as "a part of the body—white, with elastic flexible fibers arranged in parallel bands in a dense structure, solid, and resistant to rupture—that connects two or many parts, which without it could separate, to hold them in their proper relationship." Interestingly, in this comprehensive review, there is no indication of a ligament or ligamentous structure in the inframammary region of the breast.

The findings from our study contrast with those of many previous investigators. These conflicting results may be explained on the basis of investigative techniques. Our study was unable to demonstrate any ligamentous structure of dense regular connective tissue in the inframammary fold region in any of the 12 cadavers. The blunt dissection techniques used by Maillard and Garey[14] and Bayati and Seckel[15] compress the connective tissue connections to the dermis, which the present study identified, into a definable structure. Gross dissection techniques, as used by van Straalen et al.,[16] are an inaccurate method to define microscopic structures such as those demonstrated in this study. Also, the lack of a consistent reporting of a connective tissue structure in the area of the inframammary fold by the many surgeons performing breast procedures casts doubt on the credibility of this observation.[25]

The results of our study are consistent with those of Lockwood[17] and Stuzin et al.[26] Lockwood[17] described the anatomy of the superficial fascial system in a series of embalmed and fresh cadavers and noted several anatomic findings. First, he found that the superficial fascial system consists of one to several horizontal membranous sheets separated by adipose tissue and that these were connected by vertical fibrous septa. There were also fibrous extensions between the superficial fascial system and the dermis. Second, he observed that the anatomy of the superficial fascial system varied with adiposity, and the presence of adipose tissue separated the layers of the superficial fascia until they became indistinct. Finally, he noted the presence of sex differences in areas of fascial attachment.

Our study identified the presence of two horizontal membranous sheets (horizontal in the supine position) in all 10 female specimens, with the most superficial layer proceeding anterior to the breast glandular tissue, and the deeper layer proceeding posterior to the glandular tissue. Adherence between the two horizontal sheets of the superficial fascial system was pronounced in 2 of the 10 female specimens. Because of the sparsity of the subdermal adipose tissue (1 to 2 mm wide), the superficial and deep fascial layers appeared to be fused. By contrast, in those specimens where adipose tissue had accumulated, there was greater thickness of the superficial fascial network (2 to 28 mm wide), and the zone of adherence was less apparent. The male cadavers in this study did not demonstrate the presence of any organized connective tissue structure in the region of the inframammary fold.

Stuzin et al.[26] noted the continuation of fascial layers from the neck into the face, along with the presence of relative areas of adherence that were designated as "retaining ligaments." They believed that the aging process led to an attenuation of some of these retaining ligaments, which results in a relative ptosis of the soft tissues over the more fixed fascial attachments. The findings from this study indicate the continuation of the abdominal fascial layers onto the chest with fascial connections to the dermis. The development of the breast and the progressive effects of gravity on the breast will lead to attenuation of Cooper's ligaments and ptosis of the breast over the inframammary fold, with its fixed attachments to the superficial fascia.

We believe that the results of this study are clinically relevant for several reasons. First is the obvious importance of trying to preserve this very important structure during a mastectomy, which has been emphasized by many other authors.[5–9] Second, the presence of this fold is important in augmentation mammaplasty with regard to development of the double-bubble phenomenon.

Many surgeons have noted the development of the double-bubble phenomenon when attempting transaxillary subpectoral augmentation mammaplasty for the correction of ptosis in patients with tight inframammary folds and when attempting large-volume augmentation.[10,27] What is visualized in these patients is the natural inframammary fold in situ with a new inframammary fold below the existing fold that is created by the implant. In the discussion of this phenomenon, Bayati and Seckel[15] hypothesize that the presence of the double-bubble phenomenon is produced by the disruption of the so-called inframammary crease ligament. On the basis of the results of our study, we would offer an alternative explanation. When dissection is performed from the subpectoral plane by means of a transaxillary approach, even when the pectoralis muscle is divided, the implant will rest under the pectoralis fascia and the normal superficial fascia relationships will remain unchanged. Thus, when a large implant is placed, or when there is ptosis or a tight inframammary fold, the implant below the level of the normal inframammary fold will create the double-bubble. Support for our point of view would seem to come from Barnett,[27] who has used a disruption of the pectoral fascia and sweeping of the attachments of the breast gland to correct breast ptosis in patients undergoing augmentation mammaplasty through the transaxillary subpectoral approach. Brink[28] also advocated a subglandular dissection to release the eccentric attachments of the breast parenchyma from the pectoralis fascia to prevent the double-bubble in patients with a hypoplastic inferior pole of the breast. By performing this maneuver, he is disrupting the inframammary fold and the superficial fascial attachments and allowing the implant to create the new inframammary fold.

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Acknowledgments

The authors thank Norman Cole, M.D., for his careful and thoughtful review of the manuscript. This work was supported by a grant from the Jewish Hospital Foundation.

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