Cellular Reorganization Plays a Vital Role in Acupuncture Analgesia

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ABSTRACT

Background: Acupuncture has a long history of relieving many forms of pain. However, many of acupuncture's mechanisms are still unknown and/or misunderstood.

Objective: This review looks at past research on many different methods and targets of study related to acupuncture. The main focus is upon the importance of connective-tissue planes in and around acupuncture points.

Method: Relevant articles from journals as well as books on the topic were searched manually for information related to the topic.

Results: Various studies offered different (and sometimes interrelated) mechanisms for how acupuncture needling results in analgesia among other effects. Emerging evidence, however, has shown the increasing importance of extracellular matrix rearrangements that result in lower mechanical stress states of surrounding tissues. This leads to lower constant stimulation of regional mechanoreceptors, in turn, reducing chronic pain and discomfort.

Conclusions: The extracellular matrix has emerged as an important area of study on the effects of acupuncture needling.

Keywords: fascia, matrix rearrangement, connective tissue, acupuncture

INTRODUCTION

EVIDENCE, STARTING AT THE END of the last century, suggests that connective tissue could play a role in acupuncture analgesia. While there has been evidence in the past for a possible correlation between acupuncture points and connective tissue, it was not immediately apparent what role connective tissue itself played in acupuncture.¹ One complicating factor in determining acupuncture's effects on connective tissue has been the difficulty in establishing what a "true" acupuncture point, or acupoint, may be. This has led other researchers to cast doubt on possible mechanisms.² Previous theories have been put forth to explain this relationship, and these theories have ranged from changes in brain neural pathways to placebo effects.^{3,4} Concurrent with these theories, emerging evidence supports cellular remodeling within the surrounding needling areas, which then

may result in a lower mechanical stress state in regional mechanoreceptors. Although it remains to be validated, this theory could explain part of acupuncture's effects on pain. This review presents a narrative on how this model fits in with prior acupuncture findings. From this, a better understanding of the various areas of research in the field might be generated, leading to increased understanding of the importance of the properties of connective tissues in the body.

HOW DOES CONNECTIVE TISSUE PLAY A ROLE IN ACUPUNCTURE?

A group of researchers theorized a four-part pathway that began with the winding of connective tissue around the intruding needle, followed by a deformation of the surrounding cellular matrix due to the winding effects on nearby collagen.⁵

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This was then thought to cause a signal transduction into the cells attached to these collagen fibers that, then, finally resulted in a cellular response. This review describes these proposed steps and the supporting evidence behind them, as well as addressing gaps in current knowledge.

To explore the relationship between acupuncture points and connective tissue, one research group examined the location of acupoints on a cadaver and identified the surrounding tissues. Interestingly, 80% of the acupoint locations were found to be within connective tissue planes.⁶ While this finding indicated an association between connective tissue planes, it could not confirm a causal relationship, given that the data were cross-sectional and derived from nonliving tissue. Later analysis of the mechanism for connective tissue winding producing analgesic effects also pointed to connective tissue density being important in acupuncture. In 2001, Langevin et al. showed that the average pullout force required to remove an acupuncture needle increased by 18% at acupuncture points, compared to control points.⁷ Note that this trial must be analyzed with caution, as the treatment and control points were not specified, so it was not confirmed that all traditional acupuncture points match connective tissue planes.

If acupuncture needling involves activation of connective tissue, one might hypothesize that the body locations that produce the largest effect would contain the highest density of connective tissue, such as the junctions between fasciae. Although the term *fascia* can be defined by differing methods, this review defines a fascia as an “uninterrupted viscoelastic tissue which forms a functional-dimensional collagen matrix.”⁸ Fasciae are attractive targets for needling modulation as they are capable of autonomous contraction and relaxation, which can play a role in long-term back stability.⁹ There is also evidence that fasciae are capable of long-distance signaling, which will be further explored later in this review.¹⁰

NEEDLE WRAPPING

The main treatment form of acupuncture involves inserting thin needles into specific locations around the body. During manual stimulation, these needles are then rotated and/or lifted and thrust to engender a characteristic sensation termed *De Qi*. Following that, they are typically left in place for a time before being withdrawn. It is thought that the important part of this is the winding that occurs around the needle, which will eventually deform the extracellular matrix. This is visible when the body appears to “grip” the needle when the practitioner pulls it out. In Traditional Chinese Medicine, this is termed *De Qi* and is both the gripping feeling the practitioner feels as well as a physical sensation the patient feels that is often associated with a dull ache and tingling.¹¹ Previous articles have theorized that this needle grasp is due to muscular contraction around the needle.^{12,13} Further research has shown that this is not the case.

One group found a significant increase in winding connective tissue density at needle sites in deceased rats even when there were no major changes in dermal or muscle tissue density at the same sites.⁵ Another experiment by Langevin et al. showed that increased muscle-tissue penetration did not result in increased pullout force.¹⁴ The researchers also postulated that this connective winding could be involved in signal transduction that takes place outside of the nervous system, which could have effects both in acupuncture and in other related treatments.

THE IMPACT OF NEEDLE MANIPULATION ON TISSUE GRASP

During manual stimulation, acupuncture practitioners elicit the *De Qi* sensation within patients by manipulating the needles following insertion. The needle may be rotated in one or both directions at varying speeds and/or the needle may be partially extracted before being pushed back in. Research has shown these rotations could play an important role in connective tissue winding. In a 2001 article, researchers stated that they found that rotation of acupuncture needles increased the pullout force necessary to remove them.⁷ In addition, the researchers tested pullout force for needles rotated in one direction and needles that were rotated in both directions before being removed. The researchers found that, while both treatments resulted in a higher mean pullout force than stationary needles, needles rotated in one direction required 167% more force as opposed to only 52% for bidirectional treatment. It can be inferred that this means unidirectional treatment results in more connective tissue winding around the needle, possibly due to partial unwinding of collagen fibers when the needle is rotated in the reverse direction. It should be noted that this effect was also noticed at control points, suggesting that needle manipulation could play a bigger role in connective winding than location. These results, however, have yet to be tested in humans undergoing treatment for clinical conditions.

During manual rotation of the needle, there are other aspects of needle manipulation that may engender acupuncture analgesic effects. In 2008, research showed that collagen gels have tearing after significant rotations.¹⁵ The exact number of rotations necessary for tearing is dependent on gel density, the existence of cross-linkage, and the depth of the needle. The researchers found that gels filled with thicker densities of collagen failed at fewer revolutions than thinner gels. Needle depth was also related to gel density, as less-dense gels with greater needle depth failed at fewer rotations. Finally, cross-linked gels failed at fewer rotations and showed less collagen alignment than non-cross-linked treatments. This should be studied further with live patients, as it raises a question regarding whether there are differences in acupuncture winding between elderly and younger patients who have less cross-linking.

There is evidence showing that upward and downward needling motions, in conjunction with rotation, result in more tissue winding, greater pullout force, and greater tissue displacement.¹⁶ Retaining the needle in the body for >5 minutes results in a loosening of the tissue–needle bond. This causes less tissue displacement in conjunction with rotation after the initial waiting period. Overall, the above results suggest that needling is capable of making an impact on underlying connective tissue in the regions surrounding the insertion site. This could help explain the mechanism for long-range manipulation of tissue via needling.

MATRIX DEFORMATION AND SIGNAL TRANSDUCTION

Acupuncture has been shown to make an impact on both short-term and long-term constituents of the surrounding matrix in different ways. Right after acupuncture needling is completed, there is noticeable tissue displacement from 4 cm to 10 cm away from the treatment location.^{16,17} However, there is evidence that certain stress-related connective tissue changes occur only in loose connective tissue. A 2013 article published in the *Journal of Cellular Physiology* focused on unspecialized connective tissue.¹⁸ The study showed that dense connective tissue cells were less capable of cytoskeletal remodeling than areolar connective tissue. It was theorized that these short alterations are an important mechanism of areolar connective tissue, helping to relax the tissue to a lower state of tension.

It is worth mentioning that this effect also occurs in specialized connective tissue.¹⁰ Unlike unspecialized connective tissue, however, matrix changes become more apparent over the course of several days or even weeks after a change in stress. The impact of matrix changes affects the surrounding cells directly, which can also play a role in needling effects.

In a 2005 trial researchers noticed that fibroblast cells become wider and thinner when the tissue was mechanically stretched.¹⁹ However, the cell-surface area increased to greater proportions than expected, leading the researchers to propose an active mechanism by the cells to alter their reaction to the stress. This active mechanism could be useful in further research when examining how mechanical forces result in long distance cellular signaling. These alterations to cellular shape could be caused by needle winding of collagen exerting force on the collagen–cell adhesion junctions.⁵ The winding of the needle pulls on collagen fibers that, in turn, pull on the fibroblasts themselves, which results in the passive cytoskeletal rearrangement.

The direct cause of the active mechanism is still unclear, but there are many possible mechanisms for cellular restructuring, all with early but promising supporting evidence. These mechanisms include cytoskeletal remodeling, hormonal factors, and alterations in gene expression, which can have an impact on not only the surrounding matrix but

also on sensory neuron reception and subsequent transmission of neural activity into the central nervous system (CNS). For instance, both traditional acupuncture and electroacupuncture (EA) seem to change gene expression for *ephrin B-3* at the regional dorsal horn.²⁰ This results in altered pain modulation and thus could play a role in acupuncture analgesia.

CHANGES IN NEUROTRANSMITTER LEVELS AND BRAIN ALTERATIONS

In addition to local changes, it has long been theorized that acupuncture could also affect neurotransmitter levels in the body, leading to the analgesic properties. A 2014 study showed that there was a relationship between EA and increasing levels of norepinephrine and dopamine.²¹ This effect was mediated via the vagal nerve and adrenal glands, as removal of either structure inhibited the increased catecholamine levels. The main effect of this treatment appeared to be caused by specific dopamine receptors, which helped reduce inflammation and improve renal function. While this is an encouraging result, this was only tested with EA, so it is, as of now, unknown how much of a role the needling itself has on this effect.

Additionally, a 2012 study showed that genes associated with neurotransmitter systems are upregulated in rats that have a high response rate to acupuncture.²² This could be a link between needling and individual cellular responses that could play a role in regional analgesia via neurotransmitter release and reception. The particular sensation of De Qi that needling evokes could also indicate slight differences in brain activation and thus analgesia. Several different types of afferent fibers carry the sensations of De Qi ranging from stabbing to soreness, which correlates with the amount of perceived analgesia.¹¹ In this way, acupuncture seems to send differential signaling to the CNS depending on needling technique.

CELLULAR RESPONSE

The deformation of the surrounding matrix causes a mechanical signal to affect nearby cells. These cells must then respond to produce the analgesic effect typically seen in acupuncture. Both the type of cells that induce these effects and the mechanisms for how they do this will require further research. A trial showed a 55% increase in mast-cell density at common acupuncture points; however, it is still not clear what purpose the cells would serve if they were involved in the needling effects.²³ Other researchers have shown that cytoskeletal rearrangement has been linked to cellular migration and protein synthesis.⁵

Furthermore, mechanical stress has been shown to alter gene expression in affected cells. These changes can affect the extracellular matrix, which can allow further signal transduction

throughout the body. Release of adenosine via fibroblast signaling has also been noted, in a 2014 study, in areas treated with needling, including a surprising release further from the treatment site.¹⁷ This is intriguing, as it could explain the noticed effects of analgesia appearing in regions surrounding the treatment site. The researcher put forth multiple explanations for this but noted that it is possible the signal transduction was carried down connective tissue planes, allowing further fibroblasts to signal for extracellular matrix release of adenosine. It is worth noting that the study used mice; thus, distances for signal transduction were limited to much smaller ranges than possible with humans.

Another expected form of response could ultimately come from the brain itself, with connective tissue deformation acting as a signal. Ho and Knight theorized that collagen bonding serves as a form of connective tissue memory.²⁴ This “memory” uses proton conduction pathways to communicate quickly with the brain, which could then respond to the needling stimulation. Further studies would be necessary to prove the importance of proton conduction pathways in tissue–brain communication, but the existence of the tissue layer acting as a memory is consistent with mechanical stress continuously altering the matrix. This could also serve as a path for future research on chronic pain, as altered matrix configurations may play a long-term role in regional pain and sensation.

THE ROLE OF MAST CELLS IN ACUPUNCTURE POINTS

The mast cell—a cell type associated with the immune and neuroimmune systems—could also play a role in acupuncture analgesia. Research suggests that some mast cells might react to acupuncture needling via degranulation—the cellular process of releasing secretory vesicles outside of the cell.²³ Mast cells are known for secreting histamine via this process, among other things. Moreover, when the researchers added disodium chromoglycate—a solution that inhibits the degranulation process of the mast cells—there was a profound decrease in the analgesic effect of acupuncture. Although this study focused on an animal model of acupuncture analgesia (meaning that studies in humans are needed), these results could be seen as showing a possible relationship between mast cells and overall analgesia following acupuncture needling.

In addition to this pathway, other processes also seem to occur during stimulation. Experiments have shown that mechanical stimulation can cause transient receptor potential channels sensitive to valinoid (TRPV) activation, leading to Ca^{2+} influx and further signaling as well as further evidence of mechanical stress causing *HMC1* (a mast-cell line type) degranulation.^{25,26} These data support the notion that TRPV channels could also play a role in signal activation in the mast cells.

A different study found that, in addition to TRPV activation, stretch-activated chloride channels could also play a

role in the degranulation process.²⁷ The stimulation of the tissue surrounding mast cells leads to a Ca^{2+} influx into the cytoplasm of the cell, which could act as a signaling cascade given that Ca^{2+} is an intracellular second-messenger molecule. This signaling cascade could then also cause the release or activation of other signaling factors downstream of mast-cell degranulation.²⁸ Researchers demonstrated the importance of histamine in pain tolerance and possibly acupuncture analgesia by first injecting histamine to show an increase in pain tolerance.²⁶ They then applied a receptor antagonist that blocked the analgesic effects without affecting the degranulation of mast cells during stimulation. These results suggested that histamine release may be an end result of mast-cell degranulation. Mast cells exist in various areas around the body and will react to various stimulation procedures similarly regardless of location.

Interestingly, the density of mast cells has been shown to be significantly higher in areas typically associated with acupuncture points.²³ If true, increased release of histamine from the higher-density mast cells could provoke the analgesic response, explaining acupuncture’s effectiveness. This provides a reasonable explanation for the increased analgesic effects when needling well-known acupuncture meridians.

THE LINK BETWEEN HEALTH AND CONNECTIVE TISSUE

The extracellular matrix for connective tissue plays an important role in structural support and is capable of change over time. For example, there is evidence that lower-back pain is correlated with an increase in connective-tissue density.²⁹ Researchers found that patients suffering from lower-back pain had, on average, a 25% increase in perimuscular tissue density, lending support to tissue makeup potentially affecting overall health. This also raises the question of whether acupuncture needling can provide relief via restructuring of the surrounding tissue without further cellular or matrix responses. As studies have not yet found if this additional tissue density is a cause or an effect of lower-back pain, this will require future research. Fibrosis and diabetes have also been found to increase cross-linking, which may have the effect of reducing fibroblast responsiveness.¹⁸ Age has also been shown to result in an increase in cross-linkages within the connective matrix. The effects of this should be researched further, as acupuncture effectiveness for elderly patients, compared to younger individuals with fewer cross-linkages, is relatively easy to test and could be useful in further research on other cross-linkage disorders.

It is still unclear what connective tissue alterations in different disorders implicate, but the current body of evidence suggests that there is a mechanical link between acupuncture needling and subsequent analgesia. If true, there is a possibility that further findings could be explored through

investigating individuals with differences in their connective tissue matrices.

DISCUSSION

There are important differences to keep in mind when examining results of trials using classic acupuncture techniques, compared to EA. Stimulation of the surrounding tissue due to mechanical acupuncture can differ from EA, as the placement locations in EA are not as precise and needle manipulation is reduced. This can lead to a difference in tissue remodeling and density, as well as adding the confounding variable of the electrical current. For this reason, it is helpful when examining relationships between certain treatments and physiologic effects to see similar studies use both types of acupuncture.

Similar logic applies to laser acupuncture. Analgesic properties have been noticed when using this method. However, laser acupuncture would not be expected to make an impact on the extracellular matrix. For this reason, it is not relevant to this discussion on the proposed reorganization mechanism.

Having addressed that, there have been many observed effects of needling when studies using either method have been carried out. The analgesic properties observed after needling can be a culmination of several of these effects. After close examination of many of these studies, it appears both possible and likely that cellular remodeling exists as a crucial step in the analgesic pathway. Other studies show there might still be importance of needling on the CNS and cellular expression near needle sites, but it is the remodeling itself that plays the largest macro role in reducing inflammation and pain in nearby areas.

Further studies examining tissue density and morphology near needle sites would be beneficial to the field. In particular, the importance of cross-linkage in needling and remodeling is a promising—if unexplored—research topic. There could be a relationship between the density of these cross-links and analgesic outcomes, which would be clinically helpful for treatment as well as academically interesting for altering current conventional thought on how acupuncture works. Morphology and cellular makeup of tissue surrounding acupoints has been examined before but it is the current authors' belief that an increased emphasis on remodeling playing a larger role in acupuncture's impact would allow novel approaches to studies and experiments on this extracellular matrix.

CONCLUSIONS

As with many physiologic processes, there can be multiple mechanisms underlying an observable subjunctive sensation such as pain relief due to needling. Neurotransmitter release

appears to be altered in receptor neurons near the vicinity of needling, as well as surrounding regions, due to the initial matrix deformation. The CNS can also respond independently with descending pathway opioid activation from visual and tactile cues (as well as certain expectations the patient has in the form of a placebo effect). A major reason for more long-lasting alterations in pain responses for patients undergoing acupuncture treatment could be attributed to needling resulting in cellular reorganization, which lowers the mechanical stress state of the tissue's mechanical receptors. Further study will be needed to isolate the exact cause of this cellular reorganization and how it works synergistically with other proposed models of pain relief. In addition, determining a main root cause for acupuncture can help lead future research into more accurate assessments of the short- and long-term benefits of the practice, as well as technique refinements that could improve the industry and patient care.

AUTHOR DISCLOSURE STATEMENT

No competing financial interests exist.

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