

ADVANCES IN GERD

Current Developments in the Management of Acid-Related GI Disorders

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The Pathogenesis of Gastroesophageal Reflux Disease



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G&H Traditionally, what has been the understanding of the pathogenesis of gastroesophageal reflux disease?

RFS The traditional pathogenesis of gastroesophageal reflux disease—the explanation that is taught in medical schools and that has probably been around for 100 years—is that reflux esophagitis develops when gastric juice refluxes from the stomach into the esophagus, and gastric acid inflicts a chemical burn on the esophageal mucosa. A number of studies have shown that reflux hydrogen ions directly damage esophageal epithelial cells. Animal models have also demonstrated that the acidic environment and pepsins attack the tight junctions between the cells that maintain the epithelial barrier, causing the spaces between the cells to dilate. This dilation allows acid and pepsin to obtain access to deeper epithelial layers. With ongoing reflux injury, surface esophageal cells die, which triggers both an inflammatory response (infiltration of neutrophils) and a proliferative response (basal cell and papillary hyperplasia). This pathogenesis has received corresponding confirmation from several human biopsy studies.

G&H What did you and your colleagues find in your study on the pathogenesis of reflux?

RFS We started using a rat model of reflux esophagitis in which we created reflux by surgically attaching the esophagus to the duodenum, which resulted in the free flow of gastric and duodenal contents into the esophagus.

As expected, the rats developed severe reflux esophagitis. However, we were very surprised to learn that it could take weeks to develop esophagitis using this model. If reflux is merely a chemical burn to the surface—as indicated in the traditional understanding of this disease—it did not make sense that it would take weeks for reflux esophagitis to develop. Chemical burns occur extremely quickly. For example, accidentally getting battery acid on your hand would result in a burn immediately; it would not take weeks for damage to develop.

To explore this paradox, we performed a systematic study of the early histologic events in the development of reflux esophagitis in our rat model after performing an esophagoduodenostomy. As discussed above, if reflux esophagitis is caused by the direct, caustic effects of refluxed gastric acid, we would expect injury to start with the death of the surface epithelial cells of the esophagus. This acute chemical burn injury would evoke an acute inflammatory response, with infiltration by neutrophils, which typically respond to acute injuries. The death of the surface cells would eventually cause proliferation of the basal cells as they tried to replace the dead surface cells, and that basal cell proliferation would be manifested by basal cell hyperplasia. Basal cell hyperplasia is a characteristic histologic finding of reflux esophagitis that has always been assumed to be a proliferative response triggered by the death of the surface cells.

What we found in our animal model was exactly the opposite sequence of events. At Day 3 after the esophagoduodenostomy, there was no apparent damage to surface epithelial cells, and the only sign of esophageal

inflammation was the appearance of T lymphocytes in the submucosa. Inflammation, predominantly demonstrated by appearance of T lymphocytes, increased to significantly elevated levels in the lamina propria, under the epithelium, by Week 1, and the T lymphocytes did not reach the epithelial layer until Week 3. Neutrophils were not seen in any layer of the esophagus until Day 7. Furthermore, we found that basal cell hyperplasia was already apparent by Week 1, even though we did not see erosion of surface cells until Week 4.

These findings were exactly the opposite of what we had been taught to expect in the development of reflux esophagitis. An acid burn model of injury would be expected to progress from the surface into the submucosa and to start with infiltration by neutrophils. Instead, what we found in our animal model was that reflux esophagitis started as a lymphocytic infiltration in the submucosa that progressed to the mucosal surface. We did not observe any loss of surface cells until 4 weeks after esophagoduodenostomy, but we saw basal cell hyperplasia within 1 week. Thus, in this animal model, it was not the loss of surface cells that triggered basal cell hyperplasia.

If caustic injury is not the primary mechanism underlying reflux esophagitis, then one alternative hypothesis is that gastroesophageal reflux triggers a cytokine-mediated immune response, and it is that immune response that causes the esophageal injury.

G&H How did you examine this hypothesis?

RFS We performed a number of in vitro experiments to explore this alternative hypothesis. We used normal esophageal squamous epithelial cells developed in our laboratory and exposed them to an acidic, bile salt solution. These cells began to secrete large amounts of inflammatory cytokines, such as interleukin-8, within 2 days. We then took the conditioned media from those cells, which contained the cytokines that they secreted when they were exposed to acid and bile salts, and we found that the conditioned media caused a significant increase in the migration rates of T cells and neutrophils. Thus, our in vitro studies showed that exposing esophageal squamous epithelial cells to acid and bile salts caused those cells to secrete cytokines that can cause inflammatory cells to migrate.

Other investigators have also demonstrated the secretion of proinflammatory cytokines by esophageal epithelial cells in reflux esophagitis, but in most of those studies, it was not clear whether that cytokine secretion was a cause or an effect of the esophagitis. In other words, it was not clear whether inflammatory cells caused the epithelial cells to secrete cytokines. Since our epithelial cell cultures contained no inflammatory

cells, it is clear that acid and bile salts alone can cause epithelial cells to secrete inflammatory cytokines. Thus, our studies support a new understanding of the development of reflux esophagitis, in which the reflux of gastric juice stimulates esophageal squamous epithelial cells to secrete chemokines that attract inflammatory cells, and it is the inflammatory cells, not the acid, that ultimately damage the esophageal mucosa.

G&H What are the limitations of your study?

RFS As with all animal models, it is not clear how faithfully our rat model recapitulates human disease. Nevertheless, we are very excited by these findings, and we feel that this is a fruitful area for future research.

G&H Could the pathogenesis for gastroesophageal reflux disease involve both the traditional understanding as well as your new findings?

RFS We did not directly investigate this issue, so I can only speculate. Our animal model findings suggest that the earliest sign of inflammation in the esophagus is infiltration of the submucosa by lymphocytes, which only later progresses to include neutrophils and involve the epithelial surface. We also found that the initial event appears to be reflux-induced stimulation of esophageal squamous cells resulting in secretion of inflammatory cytokines. We did observe loss of surface cells 4 weeks after the onset of reflux, suggesting that the effects of ongoing caustic injury, as well as cytokine-mediated inflammatory injury, may be occurring. However, this is only speculation and needs to be confirmed by further research.

G&H Have any other studies reported similar findings?

RFS I am not aware that there have been any other systematic evaluations of this issue. Researchers have used animal models before and have reported the development of esophagitis. However, these reports were always given at 6 or 8 weeks. In our model, we reported what happened at 3 days and then every week until 8 weeks. A key feature of our alternative concept for the development of reflux esophagitis is that reflux stimulates esophageal squamous epithelial cells to secrete chemokines that attract inflammatory cells, and it is the inflammatory cells that ultimately damage the esophageal mucosa. In support of this hypothesis, Yamaguchi and colleagues used a rat model of reflux esophagitis and found that the administration of anti-

neutrophil serum could block the development of acute reflux esophagitis, suggesting that an immune-mediated mechanism may underlie the initial development of reflux esophagitis.

G&H What are the next steps for investigating your proposed pathogenesis of reflux?

RFS We are planning studies in human patients to see whether our findings are applicable to human disease.

G&H At this point, does this proposed pathogenesis of reflux have any therapeutic implications for clinicians?

RFS There are no immediate therapeutic implications as of yet. Most gastroenterologists are probably not even aware of our study findings. A novel target in future therapies and, probably, developmental therapies could be a drug that targets cytokine receptors or the downstream target of the receptors. Approximately 40% of gastroesophageal reflux disease patients are refractory to proton pump inhibitor therapy and still experience symptoms. Novel approaches to reflux therapy involving targets of cytokine receptors may finally resolve symptoms in these refractory patients.

G&H Are there any medications currently being used to target cytokine receptors in other diseases that clinicians might be able to use to treat reflux?

RFS Studies have targeted cytokines and their receptors for other diseases, but I think that the risks and toxicities of these therapies outweigh their benefits for use in reflux patients because the majority of gastroesophageal reflux

disease patients do improve with proton pump inhibitor therapy. However, alternative therapies are still in pre-clinical development and are not yet clinically applicable.

G&H If human studies show that reflux is not caused by chemical burns, would reflux patients continue to take proton pump inhibitors, or would other types of medicines likely become first-line therapy?

RFS As mentioned, the majority of patients with gastroesophageal reflux disease respond to proton pump inhibitors, so acid suppression therapy is likely to remain the primary focus of medical therapy for some time to come. Our study suggests that acid is a primary stimulus for esophagitis, even if the mechanism whereby it causes esophagitis is different than previously assumed.

On the other hand, the knowledge that gastroesophageal reflux disease may be a cytokine-mediated disease opens the door to the development of other therapies that may be helpful in controlling gastroesophageal reflux disease symptoms that are not well controlled by acid suppression alone.

Suggested Reading

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