Potential Benefits of pH 8.8 Alkaline Drinking Water as an Adjunct in the Treatment of Reflux Disease

Article *in* The Annals of otology, rhinology, and laryngology · July 2012

DOI: 10.1177/000348941212100702 · Source: PubMed

2 authors, including:

Nikki Johnston
Medical College of Wisconsin
93 PUBLICATIONS **2,066** CITATIONS

SEE PROFILE

1242-042-D

Annals of Otology, Rhinology & Laryngology 121(?):??-??.

Potential Benefits of pH 8.8 Alkaline Drinking Water as an Adjunct in the Treatment of Reflux Disease

Jamie A. Koufman, MD; Nikki Johnston, PhD

Objectives: At the cellular level, tissue-bound pepsin is fundamental to the pathophysiologic mechanism of reflux disease, and although the thresholds for laryngeal damage in laryngopharyngeal reflux and for esophageal damage in gastroesophageal reflux disease differ, both forms of damage are due to pepsin, which requires acid for its activation. In addition, human pepsin remains stable at pH 7.4 and may be reactivated by hydrogen ions from any source. Thus, most tap and bottled waters (typically pH 6.7 to 7.4) would not be expected to affect pepsin stability. The purposes of these in vitro studies were to investigate whether artesian well water containing natural bicarbonate (pH 8.8) might irreversibly denature (inactivate) human pepsin, and to establish its potential acid-buffering capacity.

Methods: Laboratory studies were performed to determine whether human pepsin was inactivated by pH 8.8 alkaline water. In addition, the buffering capacity of the alkaline water was measured and compared to that of the two most popular commercially available bottled waters.

Results: The pH 8.8 alkaline water irreversibly inactivated human pepsin (in vitro), and its hydrochloric acid–buffering capacity far exceeded that of the conventional-pH waters.

Conclusions: Unlike conventional drinking water, pH 8.8 alkaline water instantly denatures pepsin, rendering it permanently inactive. In addition, it has excellent acid-buffering capacity. Thus, the consumption of alkaline water may have therapeutic benefits for patients with reflux disease.

Key Words: acid reflux, alkaline water, Barrett's esophagus, gastroesophageal reflux disease, laryngopharyngeal reflux, pepsin.

INTRODUCTION

Gastric reflux, popularly termed acid reflux, is an expensive, high-prevalence disease.[1-7] Esophageal reflux and airway reflux — gastroesophageal reflux disease and laryngopharyngeal reflux (LPR), respectively — are epidemic.[1,2] In 2007, an analysis of 17 prevalence studies[2] revealed that reflux disease had increased an average of 4% per year since 1976. At present, 40% of Americans have reflux — 22% with gastroesophageal reflux disease and another 18% with LPR[1] (also unpublished observations).

During this same period of time (ie, since the 1970s), the incidence of esophageal adenocarcinoma has increased 850%, according to US National Cancer Institute data,[1,7] and this is now the fastest growing cancer in the United States.[1,7-9] In addition, in the past 25 years, esophageal cancers have trended toward more poorly differentiated pathologic features.[8,9] Finally, the prevalence of Barrett's esophagus has been found to be equal in patients with LPR, who have symptoms such as hoarseness and chronic cough, and in patients with GERD, who have symptoms of heartburn and indigestion.[10]

The reflux epidemic, and the related epidemic in esophageal cancer, are usually attributed to the obesity epidemic; however, patients with airway reflux are typically not obese.[11] Other factors, such as *Helicobacter pylori* and extensive proton pump inhibitor use, have been suggested as possible explanations, as well; however, there are no significant data to support these factors as causative.

Koufman et al[1,12] were the first to suggest a relationship between the reflux/cancer epidemic and excessive acid in the American diet. Since 1973, all bottled and canned foods and beverages (except water) have been required by law to have a pH of lower than 4.6, and most are well below pH 4.0.[12-14] In addition, it has been reported that a low-acid diet is of therapeutic benefit to patients with recalcitrant LPR (resistant to proton pump inhibitors).[1]

The cell biology of LPR supports the rationale for recommending a low-acid diet for patients with LPR, because virtually all patients with LPR dem-

From the Voice Institute of New York (Koufman) and the Department of Otolaryngology, New York Eye and Ear Infirmary, New York Medical College (Koufman), New York, New York, and the Department of Otolaryngology, Medical College of Wisconsin, Milwaukee, Wisconsin (Johnston).

Correspondence: Jamie A. Koufman, MD, Voice Institute of New York, 200 W 57th St, Suite 1203, New York, NY 10019.

onstrate tissue-bound pepsin 3b on laryngeal biopsy.[15-21] In addition, because pepsin 3b, which comprises more than 90% of human pepsin, remains stable to pH 6.5,[5,21] it may be reactivated by hydrogen ions from any source, including dietary sources.[1,2] The biology of reflux suggests that alkalinization of the diet of reflux sufferers may be an effective therapeutic strategy.[1,12-14]

All studies on the mechanisms of laryngeal and esophageal tissue injury concur that it is a peptic, not acid, tissue injury.[1,5,6,14-27] The confusion in the medical literature and the lay press about acid reflux may exist because pepsin requires acid for activation.[5,21] Johnston et al[17] reported that 19 of 20 patients with pH-documented LPR, and only 1 of 20 controls, were found to have tissue-bound pepsin by Western analysis. Once pepsin is tissue-bound, it is associated with depletion of key cellular protective proteins, including carbonic anhydrase, E-cadherin, and stress proteins.[15-18,22] In addition, Johnston et al[23] recently reported that pepsin produces up-regulation of the genetic markers associated with laryngeal cancer.

Important in understanding LPR is consideration of the stability and spectrum of activity of human pepsin.[21,24-27] In the past, it was believed that pepsin was inactive at pH 4.[25] Those early experiments were done with porcine pepsin, not human pepsin. (Pig pepsin is inactive above pH 4.[5,25]) Human pepsin 3b, however, retains proteolytic activity up to pH 6.5; for example, it can still break down collagen at pH 6.5.[21] Peak (100%) pepsin 3b activity occurs at pH 2.0, and laryngeal damage in patients with LPR occurs at pH 5.0 or less.[16,21] Pepsin is remarkably stable at in vivo pH levels, and it is *not* inactivated at pH 7.4.[21] After reacidification of pepsin from pH 7.4 to pH 3.0, 72% of peptic activity remains.[21] In the human aerodigestive tract, a pH above 7.4 is virtually never seen.[5]

The aims of antireflux therapy are to restore normal pharyngoesophageal physiology and to help remove ("wash out") tissue-bound pepsin from tissue.[1] For years, the senior author (J.A.K.) has recommended a low-acid (and low-fat) diet for reflux patients.[1,12] It now appears that this is a beneficial adjunct in the treatment of LPR,[1] because although the benefits of a low-acid diet have not yet been conclusively proven, the diet is easy to recommend to patients because of the potential therapeutic benefits and lack of associated risks. The newest idea for adjunctive therapy is alkaline drinking water. If alkaline water denatures pepsin in vitro, it might similarly denature pepsin in the aerodigestive tract of human reflux sufferers.

MATERIALS AND METHODS

There were two parts to these laboratory studies: 1) pepsin stability assays for assessment of the effects of alkaline water on the stability of pepsin 3b (compared to normal-pH water); and 2) determinations of the buffering capacity of pH 8.8 alkaline water. The alkaline water tested was Evamor natural artesian water (pH 8.8; Covington, Louisiana). For the comparison, the two conventional-pH waters tested were the best-selling bottled water brands in the United States: Dasani (pH 6.9; Coca-Cola Company, Atlanta, Georgia) and Aquafina (pH 7.1; PepsiCo Inc, Purchase, New York).

Human pepsin 3b was diluted to 1.5 μg/mL in phosphate-buffered saline solution pH 8.8, Evamor (pH 8.8), phosphate-buffered saline solution pH 7.0, tap water (pH 7.0), and 0.1 mol/L glycine (pH 3.0). After incubation, the pepsin was then diluted in an equal volume of 0.2 mol/L glycine pH 3.0. Synthetic pepsin substrate containing paranitrophenylalanine (PNP) chromophore at the site of pepsin cleavage (Lys-Pro-Ala-Glu-Phe-PNP-Arg-Leu-COOH) was reconstituted to 20 mmol/L in methanol and diluted to 140 μmol/L in 0.2 mol/L glycine pH 3.0. The synthetic pepsin substrate was added to the pepsin solution, and the A300 was recorded every 12 seconds for 15 minutes.

The control study without pepsin was performed in substrate/glycine solution. A graph of the mean hydrolysis rate (n = 5) from the initial reaction period was plotted. Statistical significance was determined by *t*-test (2-tailed; 2-sample; unequal variance). The pH levels of Evamor, Dasani, and Aquafina bottled waters (25 mL volume) were recorded after sequential addition of 1 mol/L hydrochloric acid (HCl; n = 1).

RESULTS

Pepsin was irreversibly inactivated in Evamor water, but not in pH 7.0 water ($p < 0.001$); this effect was instantaneous. The peptic activity at pH 3.0 following dilution in Evamor or in phosphate-buffered saline solution of pH 8.8 was not significantly different from that of the control with no pepsin ($p > 0.05$). Pepsin diluted in pH 7.0 phosphate-buffered saline solution or in tap water retained near-maximal activity when the pH was reduced to 3.0 in glycine (Fig 1).

More than 8 mmol/L of HCl is required to reduce the pH of Evamor water to that at which pepsin is maximally active (pH 2.0).[21,27] Approximately 4 mmol/L of HCl is needed to reduce the pH of Evamor water to that at which pepsin has 50% activity (pH 4.5). Dasani and Aquafina, America's most

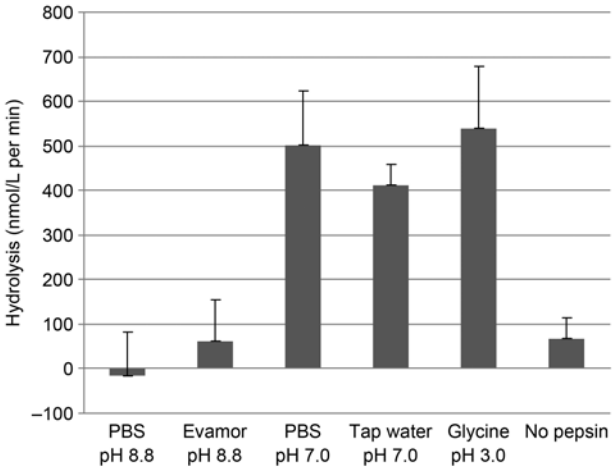

Fig 1. Hydrolysis of synthetic peptide substrate (70 μmol/L) shows stability of pepsin in phosphate-buffered saline solution (PBS) pH 8.8, Evamor (pH 8.8), PBS pH 7.0, tap water (pH 7.0), and 0.1 mol/L glycine (pH 3.0).

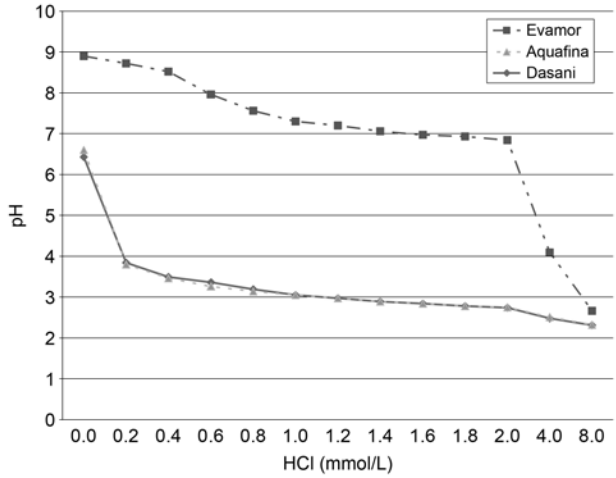

Fig 2. Results of buffering capacity experiments.

popular bottled water brands, have virtually no buffering capacity against HCl (Fig 2).

DISCUSSION

Alkaline water (pH 8.8; Evamor natural artesian water) rapidly and irreversibly denatures human pepsin 3b in vitro and demonstrates moderate HCl buffering capacity. We have previously shown that pepsin is not denatured at pH 7.4.[21] Because almost all tap and bottled waters in the United States are pH 6.7 to 7.4 and do not contain bicarbonate, one would not expect conventional waters to have any therapeutic benefits for reflux.

The authors recognize that the therapeutic benefits of alkaline water and of a low-acid diet in the treatment of reflux disease must be systematically studied, and we believe that randomized clinical trials should assess the impact of a low-acid diet (with and without alkaline water) on treatment outcomes. Nevertheless, as of this writing, the preliminary clinical data from our patients with LPR suggest that alkaline drinking water does indeed have therapeutic benefits.

We have initiated three alkaline water–related research protocols: 1) the impact of alkaline water on therapeutic outcomes (symptoms and findings) in patients with LPR; 2) the impact of alkaline water consumption on real-time reflux episodes assessed during pH monitoring studies; and 3) a longitudinal study of the impact of acid suppression with a low-acid diet and pH 8.8 alkaline water on 20 patients with biopsy-proven Barrett's esophagus.

CONCLUSIONS

Naturally alkaline artesian water at pH 8.8 instantaneously and permanently denatures (inactivates) human pepsin 3b, and it also appears to effectively buffer acid (HCl). These in vitro data suggest that alkaline water may be a useful, risk-free adjunctive treatment for reflux disease.

REFERENCES

1. Koufman JA. Low-acid diet for recalcitrant laryngopharyngeal reflux: therapeutic benefits and their implications. Ann Otol Rhinol Laryngol 2011;120:281-7.

2. El-Serag HB. Time trends of gastroesophageal reflux disease: a systematic review. Clin Gastroenterol Hepatol 2007;5: 17-26.

3. El-Serag H, Becher A, Jones R. Systematic review: persistent reflux symptoms on proton pump inhibitor therapy in primary care and community studies. Aliment Pharmacol Ther 2010;32:720-37.

4. Altman KW, Stephens RM, Lyttle CS, Weiss KB. Changing impact of gastroesophageal reflux in medical and otolaryngology practice. Laryngoscope 2005;115:1145-53.

5. Koufman JA. The otolaryngologic manifestations of gastroesophageal reflux disease (GERD): a clinical investigation of 225 patients using ambulatory 24-hour pH monitoring and an experimental investigation of the role of acid and pepsin in the development of laryngeal injury. Laryngoscope 1991;101(suppl 53):1-78.

6. Koufman JA. Commentary. In: Branski R, Sulica L. Classics in voice and laryngology. San Diego, Calif: Plural Publishing, 2009:179-88.

7. Pohl H, Welch HG. The role of overdiagnosis and reclassification in the marked increase of esophageal adenocarcinoma incidence. J Natl Cancer Inst 2005;97:142-6.

8. Lund O, Hasenkam JM, Aagaard MT, Kimose HH. Time-related changes in characteristics of prognostic significance in carcinomas of the oesophagus and cardia. Br J Surg 1989;76:1301-7.

9. Conio M, Blanchi S, Lapertosa G, et al. Long-term endoscopic surveillance of patients with Barrett's esophagus. Incidence of dysplasia and adenocarcinoma: a prospective study.

Am J Gastroenterol 2003;98:1931-9.

10. Reavis KM, Morris CD, Gopal DV, Hunter JG, Jobe BA. Laryngopharyngeal reflux symptoms better predict the presence of esophageal adenocarcinoma than typical gastroesophageal reflux symptoms. Ann Surg 2004;239:849-58.

11. Halum SL, Postma GN, Johnston C, Belafsky PC, Koufman JA. Patients with isolated laryngopharyngeal reflux are not obese. Laryngoscope 2005;115:1042-5.

12. Koufman JA, Stern M, Bauer M. Dropping acid: the reflux diet cookbook and cure. Minneapolis, Minn: Brio Publishing, 2010.

13. Acidified foods. Code of Federal Regulations—Title 21—Food and Drugs; Chapter I, Department of Health and Human Services; Subchapter B—Food for Human Consumption Part 114. United States Food and Drug Administration. Arlington, Va: Washington Business Information, 2010.

14. Generally recognized as safe food additives: FDA Database of Selected GRAS Substances. United States Food and Drug Administration. Springfield, Va: National Technical Information Service, 2009.

15. Axford SE, Sharp N, Ross PE, et al. Cell biology of laryngeal epithelial defenses in health and disease: preliminary studies. Ann Otol Rhinol Laryngol 2001;110:1099-108.

16. Johnston N, Bulmer D, Gill GA, et al. Cell biology of laryngeal epithelial defenses in health and disease: further studies. Ann Otol Rhinol Laryngol 2003;112:481-91.

17. Johnston N, Knight J, Dettmar PW, Lively MO, Koufman J. Pepsin and carbonic anhydrase isoenzyme III as diagnostic markers for laryngopharyngeal reflux disease. Laryngoscope 2004;114:2129-34.

18. Gill GA, Johnston N, Buda A, et al. Laryngeal epithelial defenses against laryngopharyngeal reflux: investigations of E-cadherin, carbonic anhydrase III, and pepsin. Ann Otol Rhinol Laryngol 2005;114:913-21.

19. Samuels TL, Johnston N. Pepsin as a marker of extraesophageal reflux. Ann Otol Rhinol Laryngol 2010;119:203-8.

20. Knight J, Lively MO, Johnston N, Dettmar PW, Koufman JA. Sensitive pepsin immunoassay for detection of laryngopharyngeal reflux. Laryngoscope 2005:115:1473-8.

21. Johnston N, Dettmar PW, Bishwokarma B, Lively MO, Koufman JA. Activity/stability of human pepsin: implications for reflux attributed laryngeal disease. Laryngoscope 2007;117:1036-9.

22. Johnston N, Dettmar PW, Lively MO, et al. Effect of pepsin on laryngeal stress protein (Sep70, Sep53, and Hsp70) response: role in laryngopharyngeal reflux disease. Ann Otol Rhinol Laryngol 2006;115:47-58.

23. Johnston N, Yan JC, Hoekzema CR, et al. Pepsin promotes proliferation of laryngeal and pharyngeal epithelial cells. Laryngoscope (in press).

24. Goldberg HI, Dodds WJ, Gee S, Montgomery C, Zboralske FF. Role of acid and pepsin in acute experimental esophagitis. Gastroenterology 1969;56:223-30.

25. Piper DW, Fenton BH. pH stability and activity curves of pepsin with special reference to their clinical importance. Gut 1965;6:506-8.

26. Lillemoe KD, Johnson LF, Harmon JW. Role of the components of the gastroduodenal contents in experimental acid esophagitis. Surgery 1982;92:276-84.

27. Johnson LF, Harmon JW. Experimental esophagitis in a rabbit model. Clinical relevance. J Clin Gastroenterol 1986;8 (suppl 1):26-44.