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Acid and Weakly Acidic Gastroesophageal Reflux and Pepsin Isoforms (A and C) in Tracheal Secretions of Critically Ill Children

Cristiane Hallal, PhD; Veridiana S. Chaves, MD; Gilberto C. Borges, MD; Isabel C. Werlang, PhD; Fernanda U. Fontella, PhD; Ursula Matte, PhD; Marcelo Z. Goldani, PhD; Paulo R. Carvalho, PhD; Eliana A. Trotta, PhD; Jefferson P. Piva, PhD; Sergio G. S. Barros, PhD; and Helena A. S. Goldani, MD, PhD

BACKGROUND: Gastroesophageal reflux (GER) and pulmonary aspiration are frequent in patients in the ICU. The presence of pepsin in airways seems to be the link between them. However, pepsin isoforms A (gastric specific) and C (pneumocyte potentially derived) need to be distinguished. This study aimed to evaluate GER patterns and to determine the presence of pepsin A and C in tracheal secretions of critically ill children receiving mechanical ventilation.

METHODS: All patients underwent combined multichannel intraluminal impedance-pH (MII-pH) monitoring. Tracheal secretion samples were collected to determine the presence of pepsin. Pepsin A and C were evaluated by Western blot. MII-pH parameters analyzed were number of total GER episodes (NGER); acid, weakly acidic, and weakly alkaline GER episodes; and proximal and distal GER episodes.

RESULTS: Thirty-four patients (median age, 4 months; range, 1-174 months) were included. MII-pH monitoring detected 2,172 GER episodes (77.0% were weakly acidic; 71.7% were proximal). The median NGER episodes per patient was 59.5 (25th-75th percentile, 20.3-85.3). Weakly acidic GER episodes per patient were significantly more frequent than acid GER episodes per patient (median [25th-75th percentile], 43.5 [20.3-68.3] vs 1.0 [0-13.8], respectively; $P < .001$). Only three patients had an altered acid reflux index (44.9%, 12.7%, and 13.6%) while not taking antacid drugs. Pepsin A was found in 100% of samples and pepsin C in 76.5%.

CONCLUSIONS: The majority of GER episodes of children in the ICU were proximal and weakly acidic. All patients had aspiration of gastric contents as detected by pepsin A in tracheal fluid. A specific pepsin assay should be performed to establish gastropulmonary aspiration because pepsin C was found in > 70% of samples. CHEST 2015; 148(2):333-339

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ABBREVIATIONS: ARI = acid reflux index; GER = gastroesophageal reflux; MII-pH = multichannel intraesophageal impedance-pH; PA = pulmonary aspiration

AFFILIATIONS: From the Post-Graduate Program Sciences in Gastroenterology and Hepatology (Drs Hallal, Barros, and H. A. S. Goldani); Pediatric Gastroenterology Unit (Drs Hallal, Borges, and H. A. S. Goldani), Pediatric Intensive Care Unit (Drs Chaves, Carvalho, Trotta, and Piva) and Laboratory of Translational Pediatrics (Drs Werlang, Fontella, M. Z. Goldani, and H. A. S. Goldani), Hospital de Clínicas de Porto Alegre; and Post-Graduate Program in Child and Adolescent Health (Drs Matte, M. Z. Goldani, Carvalho, and H. A. S. Goldani), Faculdade de Medicina, Universidade Federal do Rio Grande do Sul, Porto Alegre-RS, Brazil.

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CORRESPONDENCE TO: Cristiane Hallal, PhD, Rua Ramiro Barcelos 2350, CEP 90035-903, Porto Alegre-RS, Brazil; e-mail: cris.hallal@yahoo.com

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Patients in the ICU experience several disturbances that may cause GI motility abnormality and increase the risk of gastroesophageal reflux (GER) and pulmonary aspiration (PA). Swallowing dysfunction, impaired esophageal clearance, reduced activity of lower esophageal sphincter function, slow gastric emptying, and impairment of small intestinal motility are some of these abnormalities.[1,2] In addition, several factors may contribute to the high risk of GER and PA, such as the presence of a nasogastric tube, supine positioning, lower esophageal sphincter relaxing medication, sedation, and mechanical ventilation.[3,4]

GER is a common problem in patients in the ICU and may lead to erosive esophagitis and PA.[4,5] Although the mechanisms that cause GER in critically ill patients receiving mechanical ventilation are well studied,[6] the patterns regarding pH (acid, weakly acidic, weakly alkaline) and the height reached by the refluxate (proximal, distal) in the esophagus have not yet been described.

PA of gastric contents may result in a range of lung injuries. Large-volume PA may cause mechanical obstruction and life-threatening pneumonia. Microaspiration may be silent and asymptomatic, but if sustained, it may lead to lung injury as a consequence of chemical damage and release of proinflammatory mediators.[7,8] Ventilator-associated pneumonia is the major consequence of PA and one of the most frequent causes of hospital infection in patients in the ICU, increasing the length of hospital stay and use of medical resources.[7,9] Early diagnosis of GER and PA is essential to establish treatment and prevent complications.

Pepsin has been regarded as an important biomarker of extraesophageal reflux and gastric content aspiration.[10,11] Its presence has been shown in the trachea, lungs, larynx, middle ear, and saliva.[12-16] Five types of pepsin are known. Pepsinogen A, a precursor of pepsin A, is found exclusively in the stomach. Pepsinogen C, a precursor of pepsin C or gastricsin, is also found in sites distant from the gut, such as lungs, pancreas, and seminal vesicles.[17]

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PA and GER are common problems in patients in the ICU and may worsen clinical outcome.[7,12,18] Pepsin seems to be the link between these events, but this association has not been clearly demonstrated. Previous reports did not show a relationship between the presence of pepsin in the airways of critically ill children and the presence of GER. The prevalence of PA in critically ill patients has been assessed by a nonspecific pepsin assay that does not distinguish between pepsin A and pepsin C. The present study aimed to (1) evaluate the GER patterns (acid, weakly acidic, and weakly alkaline; proximal and distal) assessed by multichannel intraesophageal impedance-pH (MII-pH) monitoring in critically ill children receiving mechanical ventilation and (2) to determine the presence of pepsin A and pepsin C in the tracheal secretions of these patients.

Materials and Methods

This study was approved by the Ethics and Research Committee of the Hospital de Clínicas de Porto Alegre (protocol no. 09/631). Informed and written consent were obtained from all parents or legal guardians.

A prospective observational study was performed in patients requiring mechanical ventilation admitted to the pediatric ICU between January 2011 and December 2012. All patients were under sedation and on full enteral intermittent feeding every 3 h. The majority of patients were on an antacid regimen (proton pump inhibitor or H_2 blocker) as prophylaxis for GI bleeding. Enteral feeding was given by gastric or postpyloric tube. Patients who had contraindications for the insertion of a nasogastric tube (orofacial problems, GI bleeding, or severe coagulopathy) or who had previous surgery at the gastroesophageal junction were excluded.

Study Protocol

MII-pH Monitoring: All patients underwent MII-pH monitoring during a period of at least 20 h. All procedures were performed by the same examiner, with age-appropriate probes with seven impedance sensors 1.5 or 2.0 cm apart and one distal pH sensor (Sandhill Scientific). The pH electrode was calibrated in buffer pH 4 and 7 at the beginning of each study. The catheter was inserted transnasally, approximately positioned according to modified Strobel formula,[19] and set at two vertebral bodies above the diaphragmatic angle, as recommended for infants and children.[20] The pH probe location was confirmed by chest radiography and adjusted if necessary. The probe was connected to an ambulatory device containing the amplifiers that record data (Sleuth; Sandhill Scientific). An external reference electrode was attached to the patient's abdomen wall. The recording was uploaded onto a personal computer, and data were manually analyzed independently by two trained examiners (C. H. and H. A. S. G.) using BioVIEW Analysis version 5.6 software (Sandhill Scientific). The MII-pH monitoring data were analyzed following criteria described elsewhere.[20] The agreement between examiners was analyzed, and disagreement was resolved by consensus. MII-pH parameters analyzed were number of total GER episodes, height of refluxate (proximal or distal), acid reflux index (ARI) as the percentage of time when pH $<$ 4, and reflux content (acid when pH $<$ 4; weakly acidic when pH 4-7; weakly alkaline when pH $>$ 7).[21] Distal reflux was considered as that which reached the two most distal impedance channels (channels 1 and 2), and proximal reflux was defined as that started in channels 1 and 2 and reaching the most proximal channels (channels 3 and up). ARI was considered altered when $>$ 10% in children aged $<$ 1 year and $>$ 5% in children aged $>$ 1 year.[20]

Tracheal Secretion Samples: The tracheal secretion samples were collected from each patient by using a 6F or 8F suction catheter throughout the prolonged MII-pH monitoring period. The catheter was passed into the orotracheal tube down to the end of the endotracheal tube without previous saline solution according to local routine standards. After that, the tube was immediately flushed with 1 mL normal saline to collect the residual sample in the catheter. The samples were centrifuged at 4°C for 10 min at 3,000 rpm.[18] The supernatant was