UNIVERSITÀ DEGLI STUDI DELL'AQUILA
DIPARTIMENTO DI SCIENZE CLINICHE APPLICATE E BIOTECNOLOGICHE

Dottorato di Ricerca in Medicina Sperimentale

Curriculum Scienze Cliniche, Mediche e Chirurgiche

XXXII ciclo

Titolo della tesi

Eosinophilic esophagitis in esophageal atresia: tertiary care experience of a “selective” approach for biopsy sampling

SSD MED/38

Dottorando

Dott. Renato Tambucci

Coordinatore del corso

Prof.ssa Mariagrazia Perilli

Tutor

Prof. Alberto Verrotti

A.A. 2018/2019

Eosinophilic esophagitis in esophageal atresia: tertiary care experience of a "selective" approach for biopsy sampling

ABSTRACT

Background: A high prevalence (9.5 to 30%) of eosinophilic esophagitis (EoE) in patients with esophageal atresia (EA) has been reported. The application of the EoE criteria to EA patients might be problematic. To date only studies using a "routine" biopsy approach, even in asymptomatic patients, have been performed. The aim of the study was to establish the prevalence of EoE among symptomatic EA patients (EA/EoE group) without anastomotic stricture (AS) and to compare their characteristics with those of EoE patients from general population (EoE group).

Methods: From 2005 to 2018, we reviewed charts of children with EA and EoE. "Selective" biopsy approach only in EA children without AS and/or endoscopic feature of EoE was performed. Characteristics of EA/EoE and EoE groups were compared.

Results: Among 370 EA and 118 EoE, 15 EA/EoE patients were detected (4.0% of EA patients). Male predominance and a high prevalence of allergy without differences between EA/EoE and EoE groups was observed. EA/EoE children were significantly younger ($p < 0.0001$). PPI-responder patients were significantly more prevalent in EA/EoE group ($p=0.045$).

Conclusion: Our data confirm that EA patients are at high risk for developing EoE. High incidence, early onset and high prevalence of PPI-responders might suggest that esophageal motility disorders interact to increase propensity to EoE in EA patients. However, our study also suggests that overdiagnosis of EoE may occur in EA and that adapted criteria for EoE diagnosis should be developed for EA patients.

Table of Contents

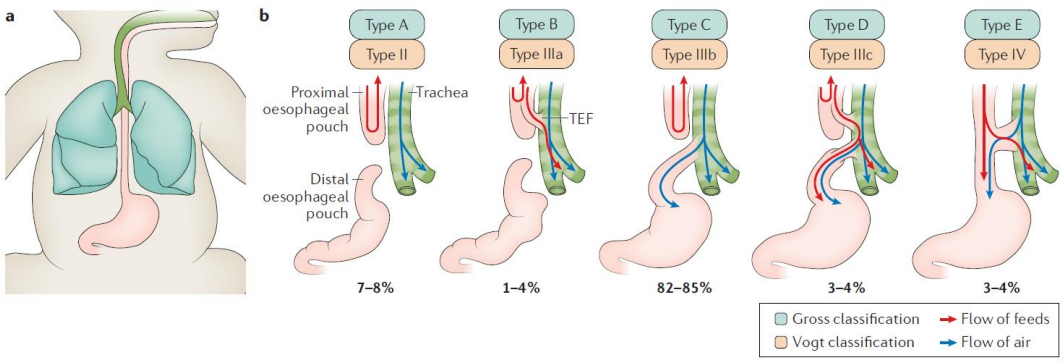
1. ESOPHAGEAL ATRESIA

Esophageal atresia (EA) is the most common congenital abnormality of the esophagus. In ~70–90% of those born with EA, a trachea-esophageal fistula (TEF) co-occurs *(Pedersen 2012; Nassar 2012; Bogs 2018; Spitz 2007)*. The condition is thought to arise as a result of deviations from the normal embryonic development of the foregut. The overall worldwide prevalence of EA as calculated from national and international databases for congenital anomalies is 2.4 per 100,000 births *(Pedersen 2012; Nassar 2012)*.

According to the Gross classification, five subtypes can be defined on the basis of the presence and/or proximity of the TEF (Fig. 1) (*Gross and Ladd 1953*); an overlapping Vogt classification is also available (*Vogt 1929*). Gross type C (EA with a distal TEF) is the most common variant *(Spitz 2007)* (Figure 1).

In most patients born with EA, surgical repair of the atresia and closure of the fistula, if present, should be performed soon after stabilization of the patient and careful preoperative management and assessment of potential comorbidities. Respiratory distress syndrome is a rare indication to perform emergency surgery; trans-pleural ligation of the TEF is required to temporarily improve respiratory status *(Spitz 2007)*. By contrast, delayed surgery is the first option in cases of long-gap atresia (Figure 1) or in those with high-risk severe comorbidities or multiple malformations. The prognosis for infants born with EA has greatly improved with advances in surgical techniques and preoperative and postoperative care. However, esophageal dysfunction occurs in all patients born with EA and is related to primary motility disorders.

Figure 1. Classification of different types of Esophageal Atresia.



a. Normal esophageal anatomy in which the esophagus and trachea are anatomically distinct. **b.** The classification of oesophageal atresia (EA) is as follows: EA without tracheo-esophageal fistula (TEF) (Gross type A , Vogt type II), EA with proximal TEF (Gross type B, Vogt type IIIa), EA with distal TEF (Gross type C, Vogt type IIIb), EA with distal and proximal TEF (Gross type D, Vogt type IIIc) and TEF without EA (Gross type E, Vogt type IV). Values in brackets indicate the frequency of each subtype. Double fistulas are possible but rare. EA can also be defined on the basis of the length of the gap between the proximal and distal oesophageal pouches. ‘Long-gap EA’ is generally considered the most difficult to repair, but its definition differs between studies and cut-offs are in the range 2-3 cm or 2-4 vertebral bodies. In addition, long-gap EA may refer to EA without fistula (Gross type A) or, in the surgical literature, be defined as being difficult to repair by primary anastomosis. The International Network of Esophageal Atresia recommends that long-gap EA should be defined as any EA that has no intra-abdominal air, which, according to the Gross criteria, includes all type A and type B abnormalities, regardless of the exact length of the esophageal gap *(van der Zee 2017; van Lennep 2019)*.

These disorders can be part of the underlying abnormalities (for example, intrinsic abnormalities in myenteric plexus that provides motor innervation to the muscular layer of the gut) or related to operative and postoperative factors (for example, iatrogenic vagal nerve damage, postoperative stricture formation at the anastomosis, peptic esophagitis and/or eosinophilic esophagitis (EoE)) *(Faure 2017)*. EA is also associated with numerous comorbidities that affect the esophagus, such as dysphagia, feeding difficulties, gastro-esophageal reflux disease (GERD) and respiratory problems *(Mousa 2017)*. GERD can lead to further deterioration of esophageal motility and cause chronic inflammation and development of gastric and intestinal metaplasia (Barrett esophagus) and even adenocarcinoma in rare cases *(Pultrum 2005; Alfaro 2005)*. Delayed esophageal clearance, resulting from the abnormal motility, may be one of the factors explaining a possible higher incidence of squamous cell carcinoma identified during screening in those born with EA *(Vergouwe 2018)*.

Therefore, since the first successful primary repair by Cameron Haight in 1941, postoperative outcomes have changed. With the exception of patients experiencing severe concomitant malformations such as congenital heart disease, improvements in operative, and perioperative care issues have shifted the focus from mortality to morbidity and quality-of-life issues *(Kovesi 2004; Rintala 2013; Castilloux 2010)*. EA is no more just a neonatal surgical problem but a lifelong problem.

To standardize the approach for the care of patients with EA, in 2016 joint European Society for Pediatric Gastroenterology Hepatology and Nutrition (ESPGHAN) and North American Society for Pediatric Gastroenterology, Hepatology and Nutrition

(NASPGHAN) consensus guidelines on the management of gastrointestinal and nutritional complications in these patients have been formulated *(Krishnan 2016)*.

1.1 EPIDEMIOLOGY OF EA

The overall worldwide prevalence of EA as calculated from national and international databases for congenital anomalies is 2.4 (range 1.3-4.6) per 100,000 births *(Pedersen 2012; Nassar 2012)*. Although the wide range in prevalence could possibly be due to ethnic, environmental or geographical differences, this has not been shown in the available studies *(Pedersen 2012; Nassar 2012)*.

Among participating countries, only differences in surveillance and reporting procedures can explain the variation in prevalence. The majority of patients born with EA are live born; spontaneous intrauterine death occurs in ~3% of cases and abortion is induced in 3-8% *(Pedersen 2012; Nassar 2012)*. In one study, prenatally detected EA led to induced abortion in 95 out of 351 cases (27%) *(Pedersen 2012)*. The majority (96.8%) of these fetuses had associated anomalies including vertebral, anorectal, cardiac, TEF, renal, radial and/or limb abnormalities (VACTERL association), trisomy 18 or multiple other malformations. Although once considered a fatal anomaly, improved surgical techniques and pediatric care have increased survival to >90% *(Donoso 2016; Lilja 2008; Cassina 2016)*, with reported 1-week survival up to 100% for babies born with EA and without other associated anomalies. Lower rates, up to 87%, are reported for patients born with long-gap EA (Fig. 1), associated cardiac anomalies and very low birthweight (<1,500 g) *(Pedersen 2012; Sulkowski 2014; Powell 2018)*.

Approximately 55% of people born with EA have associated birth defects or other anomalies *(Pedersen 2012)* (Box 1). Approximately 10% of patients have a nonrandom VACTERL association (*Pedersen 2012; Stoll 2017; Solomon 2011)*, although no clear

genetic abnormalities have yet been identified that may underlie this association. In addition, 1% of patients born with EA also have CHARGE syndrome, characterized by coloboma (a malformation of the eye affecting the lens, iris or retina), heart defects (such as tetralogy of Fallot, septal defects (atrial and ventral), aortic coarctation or aberrant subclavian artery, atresia choanae (failed recanalization of the nasal fossae during development leading to blockage), retarded growth and development, genital hypoplasia and/or ear anomalies and/or deafness *(Wyse 1993)*. CHARGE syndrome is caused by an autosomal dominant inherited mutation of the CHD7 gene, encoding chromodomain helicase DNA binding protein 7, which is involved in the organization of chromatin during development *(Sanlaville 2007)*. In addition, of patients born with EA, 6% have a trisomy 18 (Edwards syndrome) and 1-3% of patients have a trisomy 21 (Down syndrome) *(Pedersen 2012; Nassar 2012; Cassina 2016)*.

1.2 COMORBIDITIES

After surgical repair, patients born with EA are at risk of many EA-related problems. According to the ESPGHAN–NASPGHAN guidelines, patients born with EA should ideally be evaluated in a multidisciplinary team consisting of a pediatric surgeon, gastroenterologist, pulmonologist and otolaryngologist *(Krishnan 2016)*. In addition, a clinical geneticist, speech pathologist, physiotherapist and/or dietician should be consulted if needed.

BOX 1. Anomalies associated with EA (*van Lennep 2019*)

Cardiovascular anomalies

- Occur in 29% of patients born with EA
- Tetralogy of Fallot, atrial and ventral septal defects and transposition of the great arteries are screened for using echocardiography and/or electrocardiography
- Vascular malformations are screened for using MRI or CT when dysphagia, dyspnea and/or cyanosis are present

Gastrointestinal anomalies

- Occur in 16% of patients born with EA
- Anorectal malformations are screened for by physical examination and ultrasonography
- Duodenal atresia is screened for using radiography ('double bubble' sign is suggestive)
- Intestinal malrotation (using small intestine follow-through (if needed))
- Heterotopic pancreas and hypertrophic pyloric stenosis are screened for using ultrasonography (if needed)
- Heterotopic gastric mucosa is screened for using gastroscopy (if needed)
- Dumping syndrome is screened for using the oral glucose tolerance test (if needed)

Genitourinary anomalies

- Occur in 16% of patients born with EA
- Renal agenesis, cystic kidneys and ureteral anomalies are screened for using ultrasonography

Musculoskeletal anomalies

- Occur in 13% of patients born with EA
- Vertebral and/or rib anomalies and limb reduction deficiencies are screened for by physical examination and radiography
- Tethered cords are screened for using sacral ultrasonography

Respiratory anomalies

- Laryngotracheomalacia occurs in >17%, laryngeal cleft in <5%, vocal cord paresis in
- 24% (in which 7% have bilateral paresis) and subglottic stenosis in 16% of patients born with EA
- These anomalies are screened for using laryngotracheobronchoscopy

Dermatological anomalies

- Skin anomalies and clinodactyly occur in 21% of patients born with Gross type A (Fig. 1) EA and are screened for by physical examination
- Malformations of the ear are screened for by physical examination

1.2.1 ESOPHAGEAL DYSMOTILITY

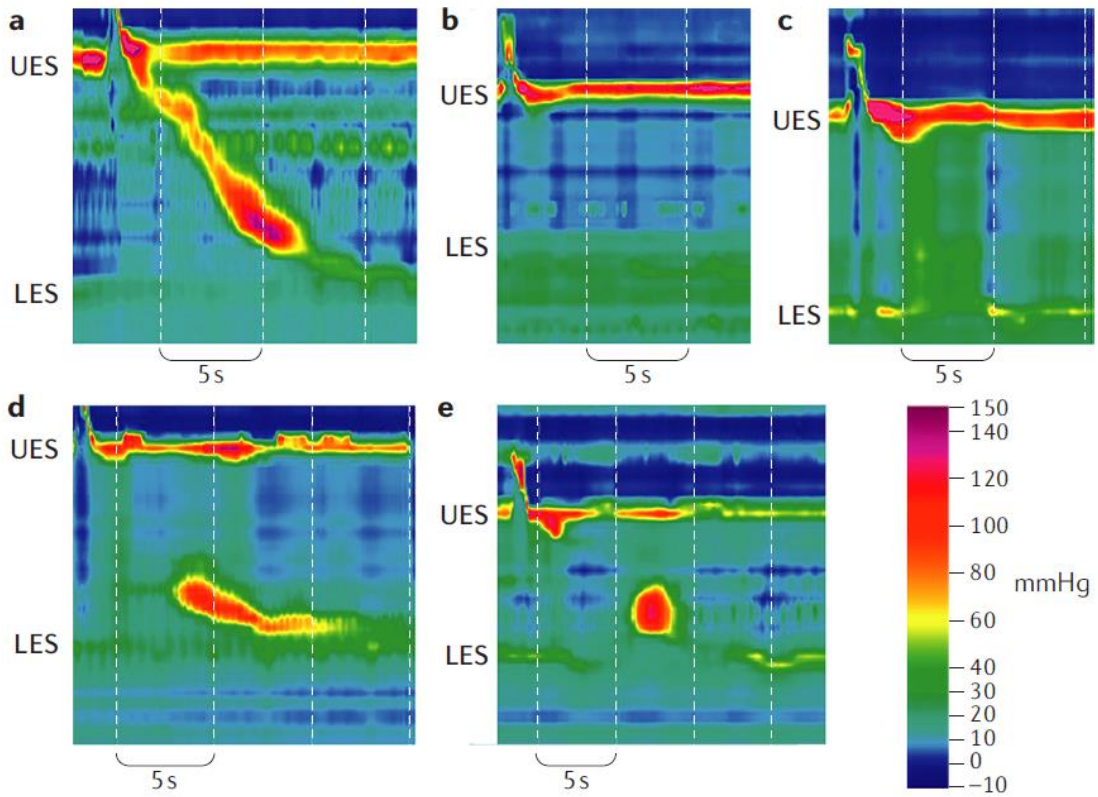
In patients operated for EA, abnormal motility of the esophagus remains the key pathophysiological catalyst leading to digestive and respiratory morbidity throughout life. Indeed, esophageal motility is involved not only in the process of transporting food from the mouth to the stomach but also plays a central role in the defense of the esophagus against gastric reflux. Furthermore, a well-organized swallowing process, from the mouth to the esophagus guarantees an adequate protection of the respiratory tract against aspiration (*Faure 2017*).

Esophageal motility has been assessed in children and adults with EA by esophageal manometry [water perfused *(Sistonen 2010; Orringer 1977; Duranceau 1977; Biller 1987; Dutta 2001; van Wijk 2013; Huynh Trudeau 2015)* or high resolution *(Lemoine 2013; Kawahara 2007; Pedersen 2013; Tovar 1995; Tong 2016; Tambucci 2015)*], esophageal intraluminal impedance (*Tambucci 2015; Di Pace 2011*), or videofluoroscopy (*Montgomery 1998; Dutta 2000*). Studies have reported anomalies at each level of the esophagus including larynx and vocal cords (*Mortellaro 2011; Morini 2011*) and gastric motor function *(van Wijk 2013; Romeo 2000)*.

In particular, abnormal esophageal peristalsis has been reported in almost all patients with EA. It is found in children (*Lemoine 2013; Kawahara 2007; Pedersen 2013; Dutta 2001; van Wijk 2013; Tambucci 2015)* and persist throughout life as demonstrated by adult studies (*Sistonen 2010; Orringer 1977; Duranceau 1977; Biller 1987; van Wijk 2013; Huynh Trudeau 2015)*. Esophageal dysmotility in EA was recently described using high-resolution manometry (HREM) with three types of abnormalities observed: aperistalsis, isolated distal contractions, and pressurization (Figure 2) (*Lemoine 2013; Kawahara 2007; Tambucci 2015)*.

The etiology of the esophageal dysmotility remains controversial. It may be related to (1) factors due to abnormal development of the esophageal smooth muscle and intrinsic innervation and of the vagus nerve or (2) to factors associated with surgical techniques, fibrotic scars, and postoperative complications. Data indicating that the congenital malformative process plays a major role are prominent in the literature, although surgical repair may exacerbate the esophageal dysmotility (*Faure 2017*).

Figure 2. Contraction patterns on HRM in patients born with EA.



High-resolution manometry (HRM) measures esophageal pressure using a transnasally placed catheter with pressure sensors. Pressure patterns can be visualized as a topography color plot with time on the *x*-axis and the position of the catheter on the *y*-axis; red indicates high pressure and blue indicates low pressure. **a.** A normal swallow in a healthy control individual. The red-yellow bar at the top indicates the high-pressure zone at the upper esophageal sphincter (UES). A peristaltic contraction wave is seen throughout the esophagus. At the bottom of the contraction wave, a subtle green band is visible indicating the high-pressure zone at the lower esophageal sphincter (LES). Note the clear relaxation of the UES and LES as part of the coordinated swallow mechanism.

b-e. HRM tracings of patients born with esophageal atresia (EA) are depicted. Three different types of dysmotility have been described in EA: aperistalsis with a complete lack of esophageal pressure change during swallowing (panel **b**); pan-esophageal pressurization, a simultaneous pressure rise in the entire esophagus due to esophageal shortening rather than peristaltic contractions (panel **c**); and several types of distal contraction in the most distal esophagus (panel **d**) and more proximally (panel **e**). (*van Lennep 2019; Lemoine 2013)*

1.2.1.1 Esophageal dysmotility and dysphagia

Dysphagia as a symptom is reported in a majority of patients with EA even though most patients learn to adapt to their unique anatomical and physiological state and do not report any complaints.

Studies have reported that dysphagia occurs in 21–84% of patients with EA at all ages after surgical repair *(Lemoine 2013; Sistonen 2010; Little 2003; Pedersen 2013)*. A prevalence of more than 50% in patients older than 10 years has been reported *(Connor 2015)*, highest prevalence is reported in adults *(Gibreel 2017; Krishnan 2016)*. Symptoms of dysphagia are not specific and vary according to the age of the patient and whether or not solid food has been introduced. Dysphagia should be evoked in patients with EA who present with food aversion, food impaction, difficulty in swallowing, odynophagia, choking, cough, pneumonia, alteration in eating habits, vomiting, and malnutrition *(Krishnan 2016)*. Children may have occasional difficulties with swallowing, are reported as sloweaters or excessive drinkers during meals. Up to three of four of patients with dysphagia report significant changes in their eating habits (need to drink, change in diet, last to finish meal) *(Lemoine 2013)*. Feeding difficulties and food aversion are reported in up to 80% of patients, which in severe cases may result in malnutrition and poor growth. In a study of 75 patients born with EA, malnourishment was common in children <1 year of age, in children with prior fundoplication (a surgical

procedure to treat GERD and hiatal hernia), in those at risk of aspiration and in those who had additional surgery in the first year of life (*Menzies 2017*).

The etiology of the dysphagia may include inflammatory (peptic or eosinophilic esophagitis) and anatomic causes (anastomotic stricture, congenital stenosis, peptic stricture, post-fundoplication obstruction, vascular compression, anastomotic diverticulum, or mucosal bridge), and abnormal esophageal motility *(Krishnan 2016)*.

Dysphagia therefore warrants a systematic workup to rule out all of the abovementioned etiologies. In the absence of one of the previously outlined causes, esophageal dysmotility, which impairs a normal bolus transit, remains the most likely explanation *(Krishnan 2016)*.

1.2.1.2 Esophageal dysmotility and GERD

After EA repair, GERD is highly prevalent from birth to adulthood. Although only a few patients actually report symptoms, either because they cannot report symptoms or because they are asymptomatic. The prevalence of GERD, when objectively measured by endoscopy with biopsies and/or pH measurement, is in the range 30-70% and depends on the diagnostic test used and the EA types included in the studies *(Pedersen 2012; Catalano 2011; Castilloux 2010; Koivusalo 2007; Krug 1999; Taylor 2007; Burjonrappa 2011; Tambucci 2015)*. Although only small studies objectively measuring GERD in patients born with long-gap EA are available, GERD is thought to be present in nearly all these patients *(Lindahl 1995)*. If symptoms are present, infants show irritability, prolonged crying, feeding difficulties, failure to thrive, silent aspiration and brief resolved unexpected events (BRUEs), which are events characterized by brief (<1 minute) and spontaneously resolving cyanosis, pallor, breathing interruptions, hypertonia or hypotonia and/or altered responsiveness *(Krishnan 2016)* 22,37. Older patients (>6 years)

often have typical GERD presentation with symptoms such as regurgitation, heartburn and chest pain *(Gupta 2006)*. EA patients likely develop a severe GER for various reasons including anatomical anomalies (hiatal hernia, abnormal position of the intrathoracic part of esophagus), vagal nerve surgical injury with abnormal gastric emptying and esophageal dysmotility. The latter leads to abnormal esophageal clearance, which increases the duration of mucosal exposure to gastric juice and acid. Several authors have shown in children and in adults that the greater the degree of esophageal dysmotility, the more the GER is complicated by epithelial metaplasia suggesting a correlation between motor disturbances and severity of reflux (*Faure 2017*).

Esophageal motility is disordered in patients born with EA, leading to delayed esophageal clearance. In combination with chronic GER, this may damage the esophageal wall and lead to gastric metaplasia, Barrett esophagus (the histological pre-malignant precursor of esophageal adenocarcinoma) and esophageal adenocarcinoma in rare cases *(Koivusalo 2007; Taylor 2007; Schneider 2017; Vergouwe 2018_2; Sistonen 2010)*. In a prospective study of 151 adults born with EA (mean age 25 years, range 16.8-68.6 years), histologically confirmed esophagitis was present in 23%, gastric metaplasia in 17% and Barrett esophagus was reported in 7% of patients, which is four times higher than the prevalence in the general population *(Vergouwe 2018_2)*. An even higher proportion of confirmed esophagitis (67%) was found in a study in 120 adolescents (aged 15-19 years) born with EA; gastric metaplasia (41%) and intestinal metaplasia (1%) were also identified *(Schneider 2017)*. Interestingly, only 41% of patients in this study reported GERD symptoms and only 28% received anti-reflux medication before endoscopy. Even in young children (median age 10.9 years, range 2.0-17.2 years), intestinal and gastric metaplasia was reported, with 7 out of 542 patients (1.3%) in one study being diagnosed with intestinal metaplasia, of which the youngest was 2 years of age *(Hsieh 2017)*. In

addition, esophageal squamous cell carcinoma in patients born with EA is likely to be caused by delayed esophageal clearance as a result of abnormal motility *(Vergouwe 2018)*.

Esophageal dysmotility and respiratory problems

Respiratory symptoms are frequently reported in patients born with EA, with coughing (in up to 75%), wheezing (in up to 40%) and dyspnea (in up to 37%) being the most common symptoms (*Legrand 2012; Spoel 2012; Porcaro 2017*). Abnormal esophageal motility, thereby hampering an adequate coordination between aerial and digestive tracts, may also foster feeding disorders and aspiration during swallowing, with extraesophageal complications such as recurrent pneumonia, bronchitis, or chronic cough. Once again, many hypotheses such as anastomotic stricture, congenital esophageal stenosis, recurrent or missed fistulae, laryngeal cleft, or developmental issues must be carefully ruled out *(Krishnan 2016)*. If the workup is negative, the motor disturbance of the esophagus remains the explanation. The esophageal dysmotility may involve upper esophageal sphincter (UES) dynamics (*Yalcin 2015; Hörmann 2002; Faure 2017*) and/or abnormal bolus clearance leading to secretions or food retention in the proximal pouch or distal esophagus or an esophageal pooling over a fundoplication.

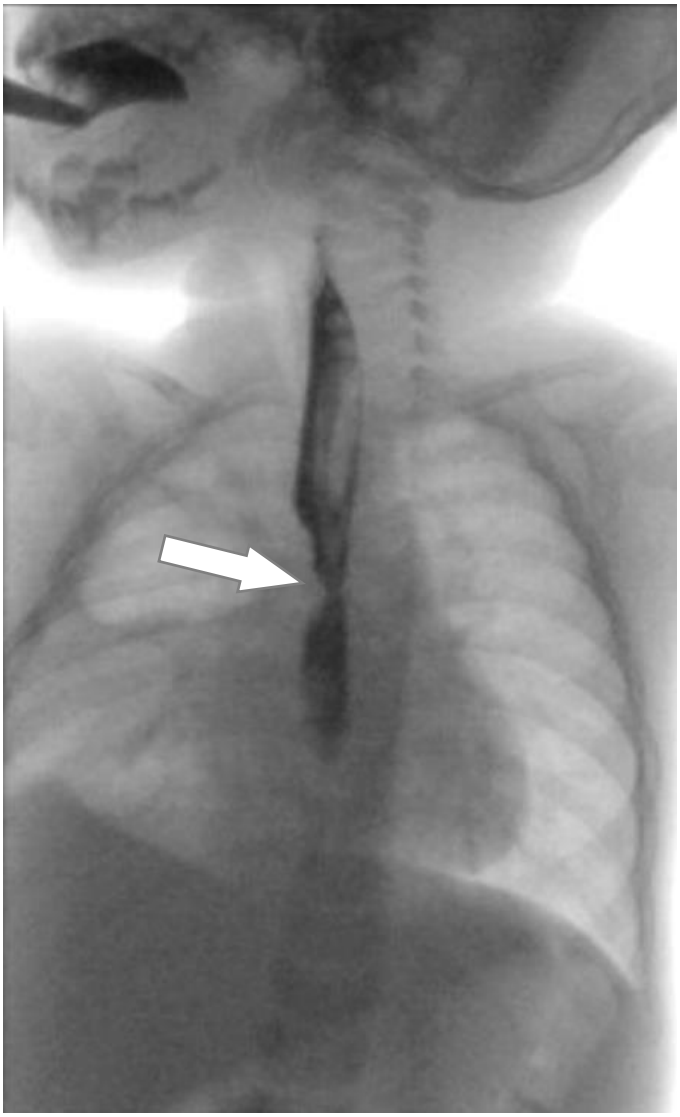
1.2.2 ANASTOMOTIC STRICTURE

Anastomotic strictures (AS) after surgical repair of the anomaly occur in up to two-thirds of patients *(Vergouwe 2019)* and has been reported to be the most frequent post-operative complication in EA, occurring in 18% to 60% of patients *(Allin 2014; Shah 2015; Tambucci 2017)*. AS is defined as a narrowing at the level of the esophageal anastomosis, detected by barium contrast study and/or endoscopy, and associated with significant functional impairment and symptoms *(Krishnan 2016)* (Figure 3).

Gastrointestinal symptoms include feeding and swallowing difficulties, drooling, regurgitation and vomiting, foreign body impaction, and poor weight gain. Respiratory symptoms include cough, oxygen desaturation during feeding, aspiration, and recurrent respiratory infections *(Krishnan 2016; Tambucci 2017; Morini 2018)*. Diagnostic techniques include esophageal contrast X-ray and endoscopy. Radiological images show the esophageal morphology and may detect associated anomalies (i.e., congenital esophageal stenosis) and pulmonary problems, while endoscopy allows combined diagnosis and treatment *(Krishnan 2016; Tambucci 2017)*. There is no consensus about the fluoroscopy or endoscopy definitions for AS in pediatrics. The reduction of luminal diameter must be compared to an age-related normal esophagus (*Tambucci 2017; Gottrand 2016*).

Concerning clinical signs and symptoms, it is worth to underline that in EA children gastrointestinal and respiratory manifestations secondary to AS may overlap with other pathologic conditions, such as esophageal dysmotility, recurrent tracheoesophageal fistula, GERD, eosinophilic esophagitis (EoE), tracheomalacia, laryngeal clefts, and vocal cord dysfunction. Clinicians must be aware that these conditions may coexist and exacerbate AS symptoms *(Baird 2013, Tambucci 2017)*. Moreover, the degree of esophageal narrowing does not correlate with symptoms. Therefore, patients with EA should be evaluated regularly by a multidisciplinary team to rule out the presence of other comorbidities (*Krishnan 2016*). ESPGHAN-NASPGHAN guidelines recommend that AS should be excluded in symptomatic children, and those children who are unable to achieve feeding milestones (Figure 4).

Figure 3. Anastomotic stricture (AS) on esophageal contrast X-ray



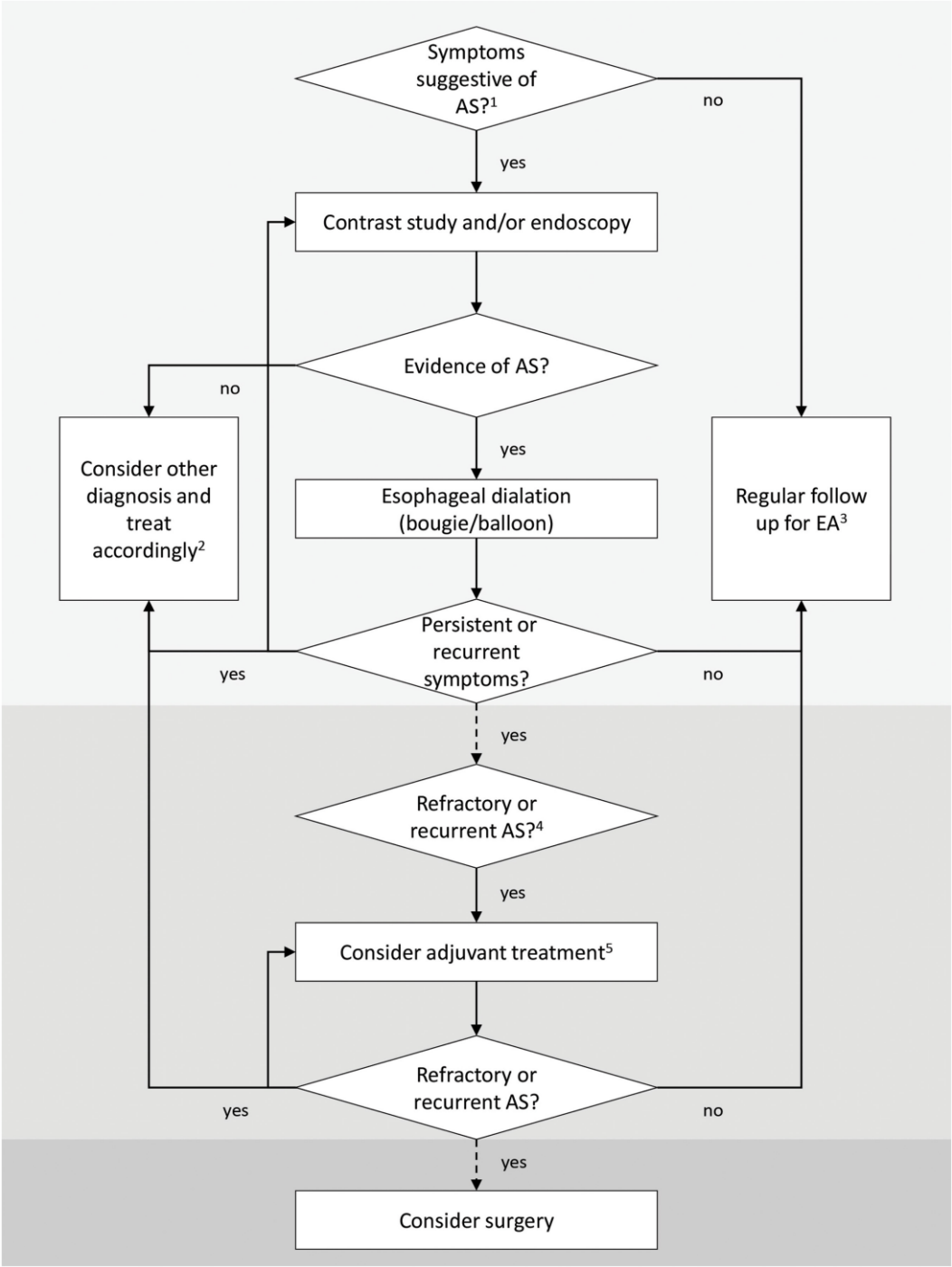
Anastomotic stricture (AS) on contrast esophagram showing intrinsic luminal narrowing (arrow) in a clinically symptomatic EA child

Once an AS has established, the cornerstone of treatment consists on endoscopic dilations that, by exerting expansible forces within the lumen of the stenosis, result in an increased esophageal diameter. The primary goal of esophageal dilation is to achieve symptom relief, permit maintenance of age-appropriate oral nutrition, and reduce the risk of pulmonary aspiration. Endoscopic dilations are the mainstream of the conservative approach and are recommended as a first-line treatment for AS following EA repair (*Krishnan 2016*).

Two main categories of dilators are used in gastrointestinal endoscopy: fixed-diameter push-type dilators (bougie dilators) and radial expanding balloon dilators (*ASGE Technology Committee 2013*). However, due to the lack of strong evidences, the choice between balloon dilation and bougie is only based on the endoscopist’s experience and level of comfort (*Krishnan 2016*).

Despite dilation treatment, some patients may experience symptoms relapse or persistency. The cause of recurrent (≥3 episodes of clinically relevant stricture relapses after dilations or inability to maintain a satisfactory luminal diameter for 4 weeks once the age-appropriate feeding diameter has been achieved) and refractory (inability to successfully remediate the anatomic problem to obtain age-appropriate feeding possibilities after a maximum of five dilation sessions with maximal 4-week intervals) AS is not fully understood (*Tambucci 2017*).

Figure 4. Simplified algorithm for diagnosis and treatment of anastomotic strictures (ASs) after esophageal atresia (EA) repair (*Tambucci 2017*).



[1] Symptoms suggestive of AS depend upon the age of the child and the type of food ingested (liquid or solid) and include feeding and swallowing difficulties, regurgitation and vomiting, mucus or food impaction, cough, drooling, recurrent respiratory infections, foreign body impaction, and poor weight gain. In EA patients, these symptoms may overlap with other pathologic conditions, and none of them alone is sensitive or specific enough to diagnose an AS.

[2] Other diagnosis includes esophageal dysmotility, recurrent tracheoesophageal fistula, gastroesophageal reflux disease, eosinophilic esophagitis, tracheomalacia, laryngeal clefts, and vocal cord dysfunction; these conditions may coexist and exacerbate AS symptoms. Patients with EA should be evaluated regularly by a multidisciplinary team.

[3] EA children in the first 2 years of life (with special attention during the introduction of solid food) and patients with long-gap EA and postoperative anastomotic leak need a closer follow-up.

[4] Recurrent AS: ≥3 episodes of clinically relevant stricture relapses after dilations or inability to maintain a satisfactory luminal diameter for 4 weeks once the age-appropriate feeding diameter has been achieved. Refractory AS: inability to successfully remediate the anatomic problem to obtain age-appropriate feeding possibilities after a maximum of five dilation sessions (refractory) with maximal 4-week intervals (20).

[5] Potential adjuvant treatments may include intralesional and/or systemic steroids, topical application of mitomycin C (MMC), stents, and an endoscopic incisional therapy (14). Temporary stent placement or application of topical MMC following dilation is suggested as a first-line adjunctive treatment in children.

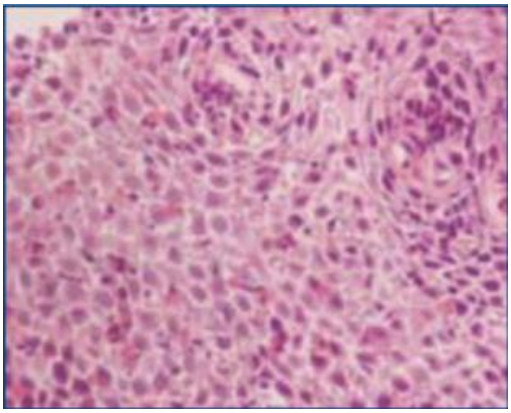
Refractory strictures are reported in 7% of patients after an end-to-end anastomosis, with a median number of 10 (range 5-34) dilations needed *(Vergouwe 2019)*. Patients with refractory strictures have a considerable burden of care. They require frequent hospital admissions for dilations or other therapeutic interventions that involve anesthesia *(Davidson 2016)* may require post-intervention tube feeding and carry the risk of adverse events. Risk factors for refractory strictures are long-gap EA, anastomotic leaks and the occurrence of early postoperative strictures *(Vergouwe 2019)*. Conservative approach for recurrent/refractory should be preferred before considering any surgical treatment. Different non-surgical adjuvant treatments have been used to minimize the risk of stricture reoccurrence, such as intralesional steroids injection, topical application of mitomycin C, esophageal stenting, and endoscopic incisional therapy (*Tambucci 2017*), those children who fail to respond to all conservative strategies require a surgical intervention consisting (stricture resection or esophageal replacement) (Figure 4).

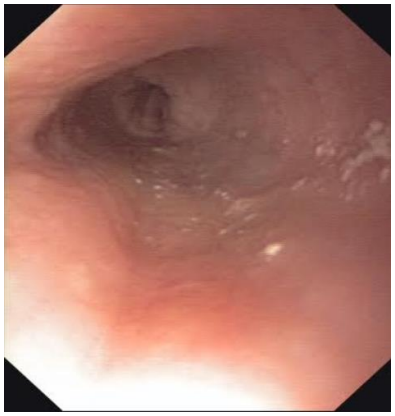
1.2.3 EOSINOPHILIC ESOPHAGITIS.

Currently, EoE is defined as a chronic, local immune-mediated esophageal disease characterized, clinically, by symptoms related to esophageal dysfunction and, histologically, by eosinophil-predominant inflammation *(Lucendo 2017; Papadopoulou 2014)* (Figure 5).

The disease is understood as a clinical-pathological entity, where symptoms and histology must always be considered together, both for the diagnosis and for the follow-up or assessment of the response to treatment (*Dellon 2018*).

Figure 5. Histologic and endoscopic appearance of EoE





Symptoms in EoE are due to esophageal dysfunction. In young children food refusal, difficulties in introducing new foods in the diet, the preference for liquids and soft diets, and a tendency to be “slow eaters” are to be noted, abdominal pain and vomiting are common in pre-school and school ages. In older ages dysphagia for solids and chocking, impaction of food with anxiety at the time of meals, chest pain and symptoms of GERD or non-specific pharyngeal discomfort are the most frequent symptoms *(Lucendo 2017; Papadopoulou 2014; Miehlke 2014)*.

In general, gastrointestinal symptom are nonspecific and can also be attributed to other EA-related conditions including GERD, esophageal strictures and esophageal dysmotility (*van Lennep 2019*). Hence it has been suggested that often either the EoE is mis-diagnosed as refractory GERD or there is a delayed diagnosis of EoE. This delay might not only result in the EA patient with EoE having unnecessary escalating therapeutic interventions for their presumed poorly treated refractory GERD but might also put them at risk for developing complications from their untreated EoE (*Krishnan 2019*).

Although the worldwide prevalence of EoE is 0.03%, prevalence ranging from 9.5 to 30% is reported in children born with EA *(Dhaliwal 2014; Krishnan 2019; Petit 2019; Lardenois 2019; Yasuda 2019; Pesce 2019)*.

1.2.3.1 Etiology and pathogenesis of EoE (with focus on EA)

It is known that EoE has a high degree of heritability, with a majority of the phenotypic variation believed to be genetic in origin as shown by genetic epidemiology studies of twins and families. A study by Alexander et al. showed the EoE relative risk ratio to be increased 10- to 64-fold depending on the family relationship, compared with the general population (*Alexander 2014*).

Since 2010, three GWAS have been published identifying c11orf30, STAT6, ANKRD27, CAPN14 loci which influence risk for EoE in both children and adults (*Sleiman 2015*).

Jensen et al. have also shown a positive association between several early-life factors and EoE, including prenatal (maternal fever; preterm labor), intrapartum (cesarean delivery), and infancy (antibiotic; use of an acid suppressant) factors (*Jensen 2018*). A previous study by the same author had shown an even higher risk (6 times) between antibiotic use in infancy and odds of having EoE (95% CI 1.7–20.8) (*Jensen 2013*). A more recent study by Jensen et al., found an association between genes (CAPN14, 5q, 11q,12q,2p, and LOC283710/KLF13) and early-life environment factors (breast-feeding and NICU admission), which could potentially contribute to EoE susceptibility. They found that breast-feeding in those with the susceptibility gene variant (CAPN14) reduced the risk of EoE (*Jensen 2018*_2).

Apart from Jensen et al., other authors have also postulated that early and prolonged exposure to acid suppression medication may trigger IgE-mediated food allergies in EA patients. Untersmayr et al. found that reduction in gastric acid increased the allergenicity of food proteins (*Untersmayr 2008*). This hypothesis has also been supported in mouse models where PPI use can cause the formation of food-specific immunoglobulin (Ig) E antibodies and trigger food allergy (*Untersmayr 2003*). Orel et al. have also described other possible mechanisms by which PPI exposure might potentially lead to an increased risk of development of EoE in patients, due to their adverse influence on mucosal barrier function, interference with pH-related protein digestion by pepsin, and antigen processing by immune cells. Orel et al. stated that, acid suppressive medications may interfere with peptic food digestion, thereby contributing to an increase in food-specific antigens and may also increase mucosal permeability. These effects together may cause increased allergic reactivity to foods over time, perhaps sensitizing persons and eventually triggering EoE (*Orel 2016*). This is especially of importance in EA patients who are exposed to PPIs from birth for prolonged periods of time.

This early reduction in gastric acid could potentially increase the allergic reactivity to foods by reduced peptic digestion of food proteins and increased mucosal permeability of food proteins and thereby increase the risk for development of EoE.

EA patients are also often premature and delivered by cesarean section especially if the EA is diagnosed antenatally and associated with cardiac and other anomalies. EA cohort are all also exposed to antibiotics post EA repair and for treatment of their recurrent chest infections in early life. EA cohort who are routinely admitted to NICU post EA repair after birth, are often formula fed due to feeding difficulties. As antibiotic use and early exposure to acid suppression cannot be altered in the EA cohort, persisting with

breast feeding in early life and reducing the duration of exposure to acid suppressive medication would potentially be of added benefit in this cohort, although these assumptions would need to be validated in prospective long term studies. Thus, there could be several early life factors in the EA cohort which could potentially increase their risk for subsequent development of EoE. Some of these early life factors are potentially modifiable and hence if confirmed would have implications for improved understanding of EoE pathogenesis and disease prevention, in the EA cohort (*Krishnan 2019*_**2**).

It is now accepted that EoE is the result of a T-helper cell 2-type immune response in which eotaxin 3 and interleukins (IL) 4, 5, and 12 and 13 are upregulated (*Straumann 2001; Bullock 2007*). The gene for eotaxin-3, which is a chemoattractant and activating factor for eosinophils, has been shown to be increased 53-fold above normal levels in patients with EoE (*Blanchard 2007; Bhattacharya 2007*). Both EA and EoE are polygenic conditions, and recently, Gorter et al. postulated a possible genetic association between EA and EoE through mutations in the FOX gene cluster (*Gorter 2012*). In humans, the FOX gene cluster has been shown to be associated with congenital malformations in the esophagus and lung including EA and also with binding sites for FOXF1 were also found in the promoter regions of genes for eotaxin-3 and IL-8 (*Gorter 2012; McLin 2010; Shaw-Smith 2010; Krishnan 2019*).

Recently, the transcriptomes of EA patients with and without EoE, patients with EoE but without EA and healthy controls were compared, showing approximately 25% of EoE signature genes, resulting in abnormal epithelial barrier and type 2 immune-associated gene expression were dysregulated in those born with EA (at baseline) but without EoE compared to healthy controls; these genes were also found to be even more dysregulated in those with EoE but without EA and in EA patients with EoE. The dysregulated genes

included genes related to esophageal epithelial type 2 inflammation (MUC4, a specific mucin in response to TH2 cytokines; SYNPO2, a gene upregulated in EoE mucosa; and FLG, a membranal barrier molecule downregulated in patients with EoE). The presence of this genetic dysregulation in patients born with EA at baseline before the development of EoE might be the reason why there is a higher prevalence of EoE in this population. Prospective longitudinal studies are needed to confirm whether these patients with EA but with dysregulated EoE-predisposing genes at baseline would develop EoE in the future. Large prospective longitudinal studies would also be helpful in determining whether prolonging breast feeding and reducing duration and cumulative dosage of exposure to proton pump inhibitor therapy in EA patients with EoE susceptible genes would reduce their relative risk of developing EoE in the future. Interestingly, EA patients with EoE and EoE patients without EA had similar molecular transcriptomes at time of diagnosis of EoE and in remission after treatment, which is likely to be due to a similar pathogenesis induced by food allergy. In this study there was similarity in the predominance of Caucasian race, male gender and food allergy status of both the EA and non-EA group with EoE, supporting that EoE in the EA cohort is the same disease as the conventional EoE in the general pediatric population. However, children with EA developed EoE at a younger age than those with EoE alone. This underscores the importance of being aware of the risk of EoE and performing endoscopies with sufficient numbers of biopsies at multiple levels, irrespective of the age in symptomatic EA patients. Also EA patients with EoE had a more-severe clinical phenotype, with higher incidence of dysphagia, episodes of food bolus impaction and strictures requiring dilation than in those with EoE alone without EA and EA patients without EoE, highlighting the importance of timely diagnosis and treatment of EoE in EA patients. This study found that 2 genes (ANO1 and CTNNAL1) were expressed more in EA patients with EoE than

in EoE patients without EA. ANO1 is expressed by the interstitial cells of Cajal and is a calcium-activated chloride channel governing gastrointestinal smooth muscle contraction rhythms, which might be associated with dysphagia, food bolus impaction, and stricture development phenotypes observed in patients with EA and EoE. In addition, ANO1, is also an esophageal cancer marker, which is especially important in EA patients who are susceptible to esophageal squamous cell carcinoma development. Thus, ANO1 could potentially be used as a molecular marker for a malignant EA prognosis, EoE symptom monitoring, and esophageal cancer prognosis (*Krishnan 2019; Krishnan 2019_***2***)*.

Apart from a possible genetic, molecular association, and early life factors, other hypotheses have also been postulated explaining the increased incidence of EoE in EA patients. In EA after restoration of the esophageal interruption by surgery in the neonatal period, the esophageal dysmotility and predisposition to severe GERD persists. Hence as EoE an allergic insult on mucosal epithelium by food antigens or aeroallergens plays a role (*Sherrill 2011*).

Acid peptic mucosal injury due to severe GERD may impair the mucosal barrier function in EA patients and thereby increase the risk of sensitization to food and aeroallergens thereby increasing the risk of developing EoE. It has been shown that the normally impermeable esophageal mucosa when exposed to acid becomes permeable for peptides up to 20 KD thereby allowing food allergens to enter the sub-epithelial layer and induce eosinophilic inflammation (*Allen 2011*).

Esophageal dysmotility and prolonged bolus clearance time in EA patients could also result in increased exposure to potential allergens in the esophageal mucosa which is already inflamed due to acid reflux. This sustained exposure to potential food and aero

allergens resulting in sensitization could result in a T-helper cell 2 immune response (*Gorter 2012*).

2. RESEARCH STUDY

2.1 INTRODUCTION

Esophageal atresia (EA) with or without tracheoesophageal fistula is a developmental defect of the upper gastrointestinal tract, representing the most common congenital anomaly of the esophagus. The overall incidence ranges from one in every 2,400 to 4,500 live births worldwide (*Shaw-Smith 2006; Pedersen 2012; Nassar 2012*). Since the first successful surgical repair in 1941, a significant improvement in survival has been reached. Advances in neonatal intensive care, neonatal anesthesia, and surgical techniques have profoundly changed the natural history of EA, limiting mortality to cases with coexistent severe life-threatening anomalies such as congenital heart disease (*Ijsselstijn 2013*). Therefore, long-term morbidity and quality-of-life issues have now become priority targets in managing EA patients (*Krishnan 2016*). Anastomotic stricture (AS) formation, esophageal dysmotility-related conditions, such as gastroesophageal reflux disease (GERD) and dysphagia, as well as respiratory problems are the most common complications encountered in EA survivors (*Krishnan 2016*). Furthermore, emerging data suggest that EA patients are more likely to develop eosinophilic esophagitis (EoE) compared to general population (*Krishnan 2016; Krishnan 2019_2*).

EoE is currently defined as a chronic, immune-mediated or antigen-mediated esophageal disease characterized by symptoms related to esophageal dysfunction and eosinophil-predominant inflammation that is limited to the esophagus (*Papadopoulou 2014*). Recognition of the disease has been increasingly over the last 15 years. Current estimated annual incidence is approximately 10/100,000 cases, while prevalence ranges from 10 to 57 cases per 100,000 persons (*Moawad 2018*).

Recently, the joint ESPGHAN (European Society for Pediatric Gastroenterology Hepatology and Nutrition) and NASPGHAN (North American Society for Pediatric Gastroenterology, Hepatology and Nutrition) guidelines on long-term management of EA suggest excluding EoE in all symptomatic EA patients, especially before proceeding to anti-reflux surgery (*Krishnan 2016*).

Despite recent attention to the coexistence of EoE in patients with EA, current literature remains limited, as less than 100 cases have been detailed in the literature, with few of those cases from Europe and none from Italy.

The purpose of the present retrospective study was to further examine the relationship between EA and EoE. The primary aim was to establish the prevalence of EoE among surviving patients with EA from Italy and describe their demographic and disease characteristics. The secondary aim was to compare features of EoE in patients with EA (EA/EoE group) with those of patients from general population (EoE group).

2.2 METHODS

We conducted a retrospective chart review of children with EA and EoE, from January 2005 to October 2018. Baseline demographics, disease history and outcome data were analyzed. The study has been notified the Ethic Committee (according to the National Guidelines for Observational Study of the Italian Drug Agency, “Agenzia Italiana del Farmaco” (AIFA), retrospective studies do not need formal approval by the Ethic Committee and can be initiated by the proposer after notification, using the procedure of silence/consent). Personal data of patients have been collected totally unidentified and patients’ confidentiality was protected.

2.2.1 EA patients

EA patients had surgery at our institution or in other centers. The type of EA was classified according to the Gross classification (A-E classification) (*Gross 1953*) (Figure 1). The presence of associated anomalies, such as cardiac anomaly or VACTERL (Vertebra, Anorectal, Cardiac, Tracheo-Esophageal, Renal, Limb) association was noted. Long-gap EA (LGEA) was defined as an anatomic distance of ≥3 vertebral bodies between the proximal and distal esophageal segments and was determined according to the hospital protocol (*Bagolan 2013*). AS was defined as a luminal narrowing at the level of the esophageal anastomosis leading to a functional esophageal impairment and related symptoms (*Krishnan 2016*). Recurrent and refractory AS were defined according to the ESPGHAN and ESGE (European Society of Gastrointestinal Endoscopy) guidelines on pediatric endoscopy (*Thomson 2017*). As per ESPGHAN-NASPHAN guidelines, EA children with symptoms of esophageal dysfunction, such as dysphagia and feeding difficulties, regurgitation and vomiting, food impaction, cough or drooling, first underwent testing to rule out AS. In case of no evidence of AS, other diagnoses were considered including esophageal dysmotility, GERD, recurrent tracheoesophageal fistula, tracheomalacia, laryngeal clefts, and EoE (*Tambucci 2017*) (Figure 4). In patients with AS who underwent endoscopic esophageal dilation, biopsy specimens were not routinely collected, unless endoscopic features suggestive of EoE were present (as detailed below). Conversely, routine esophageal biopsy sampling was carried out to rule out EoE in symptomatic patients without AS. EoE diagnosis and treatment were based on current ESPGHAN guidelines (as detailed below) (*Papadopoulou 2014*).

2.2.2 EoE patients

EoE patients received diagnosis at our Institution or elsewhere. All EoE patients were followed-up in the dedicated multidisciplinary eosinophilic gastrointestinal disease clinic, providing a comprehensive evaluation from a highly experienced team of pediatric gastroenterologists, allergists and dietitians. According to ESPGHAN guidelines on EoE, diagnosis was made with both clinical and histological features (*Papadopoulou 2014*). Symptoms included dysphagia, vomiting, feeding difficulties, abdominal/chest pain, other symptoms suggestive of GERD and food impaction. Multiple biopsies were obtained from distal, mid, and proximal esophagus. At least 15 eosinophils in at least 1 high-power microscopy field (EOS/HPF) were needed for EoE diagnosis. Endoscopic typical features were noted, such as multiple esophageal rings, linear furrows, white plaques, and crêpe-paper mucosa. As per current recommendations, all patients received a trial of 8 weeks of proton pump inhibitors (PPIs) (esomeprazole or lansoprazole 2 mg/kg/day) followed by endoscopic and histological reassessment (*Papadopoulou 2014*). Patients showing clinico-histological response to PPIs were labeled as having PPI-responsive esophageal eosinophilia (PPI-REE). PPI-nonresponsive children underwent dietary treatment (amino acid-based formula or empiric elimination diet) and/or swallowed topical corticosteroids (fluticasone propionate or oral viscous budesonide) (*Papadopoulou 2014*). Data on personal and family history of allergic disorders were collected, including bronchospasm, allergic rhinitis, eczema, and food allergy. All patients underwent skin prick testing and specific IgE evaluation for both food and inhalant allergens.

2.2.3 Upper gastrointestinal endoscopy

In all patients, any acid suppression medication was discontinued at least 4 weeks before UGIE. UGIEs were performed under general anesthesia with a pediatric videoendoscope (Olympus GIF N180, XP190N, XP 160, H180J, H190, Q165; Olympus Medical, Tokyo, Japan).

2.2.4 Statistical Analysis

Collected data are presented as count and proportions (categorical data) or mean, median, standard deviation and interquartile range (continuous data). Comparisons between EoE patients from general population (EoE group) and children with EA and EoE (EA/EoE group) were performed through Fisher exact test for categorical data; Mann Whitney Wilcoxon test were applied for continuous data. Statistical tests used for comparison data analysis are specified in table 2.

A p-value of ≤ 0.05 was considered significant. Statistical analyses were performed using Prism version 6.0 (GraphPad Software, Inc., San Diego, CA, USA).

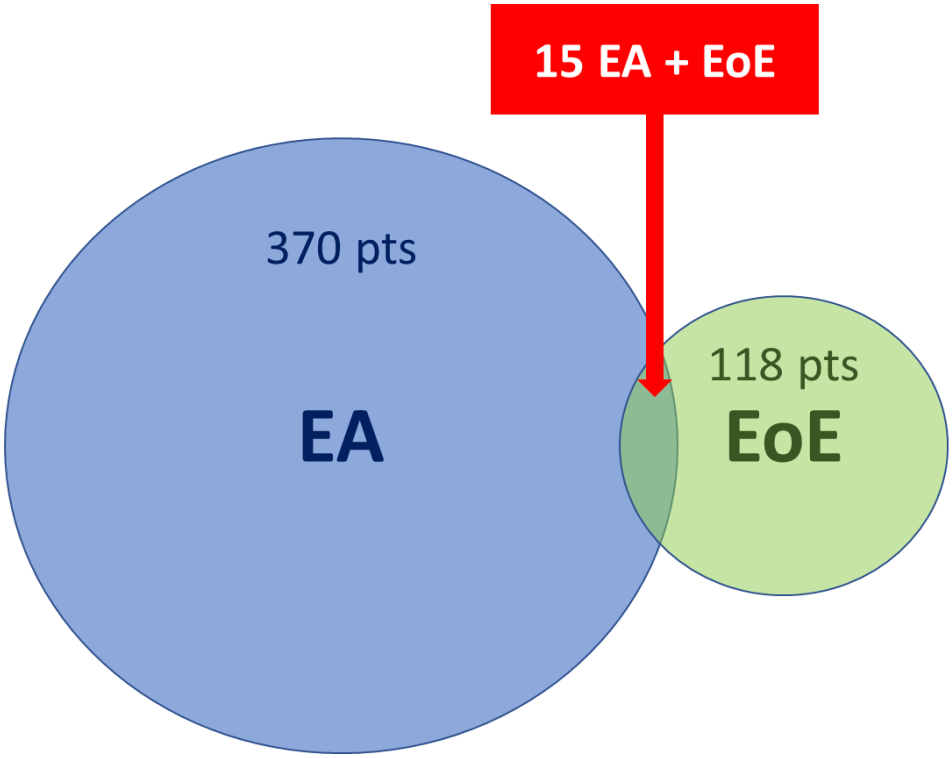
2.3 RESULTS

During the study period, clinical charts of 370 patients with EA and 118 patients with EoE were reviewed. Among these, 15 children with both EA and EoE (EA/EoE group) were identified; therefore, 4.0% of EA patients developed EoE and 12.7% of EoE children had a previous history of EA repair (Figure 6).

All 15 children of the EA/EoE group were in follow-up since birth. The median age at last visit was 9 years (range 4.8-18) and a male predominance (66.6%) was detected. The type C was the most dominant subtype of EA (86.6%). Six children (40.0%) had

LGEA (all underwent esophago-esophageal primary anastomosis). Associated congenital defects were reported in 11 patients (73.3%). History of AS was observed in 12 (80%) patients, 5 of them (33.3%) had recurrent/refractory AS. Five (33.3%) children previously underwent antireflux procedures. Demographics and disease characteristics of EA/EoE children are summarized in Table 1.

Figure 6. Graphical representation of the study population



EA: esopgaheal atresia; EoE: eosinophilic esophagitis

Table.1 Demographics and EA characteristics in children with EA and EoE (EA/EoE group)

Number of patients		15
Age at last visit [years; median (range)]		9.0 (4.8-18)
male/female		10/5
Type of EA [a] [number (percentage)]		
	Type A	1 (6.6)
	Type B	1 (6.6)
	Type C	13 (86.6)
	Type D	0 (0.0)
	Type E	0 (0.0)
Long gap EA [b] [number (percentage)]		6 (40.0)
Associated anomalies [number (percentage)]		11 (73.3)
	VACTERL association	4 (26.6)
	Heart defect	4 (26.6)
	CNS abnormalities	3 (20.0)
	Anorectal malformation	2 (13.3)
	Pulmonary defect	1 (6.6)
	Genitourinary defect	1 (6.6)
History of AS [number (percentage)]		12 (80)
	Recurrent/refractory AS [c]	5 (33.3)
Previous antireflux surgery [number (percentage)]		5 (33.3)
	Toupet fundoplication	3 (20.0)
	Nissen fundoplication	2 (13.3)

[a] according to the Gross classification [7]; [b] ≥3 vertebral bodies between the proximal and distal esophageal segments [8]; [c] according to the ESPGHAN and ESGE definition [9].

Abbreviation: EA, esophageal atresia; EoE, eosinophilic esophagitis; VACTERL, Vertebra-Anorectal-Cardiac-Tracheoesophageal-Renal-Limb; CNS, central nervous system; AS, anastomotic stricture

Comparison of clinical characteristics between EA/EoE group and EoE patients from general population (EoE group) is illustrated in Table 2.

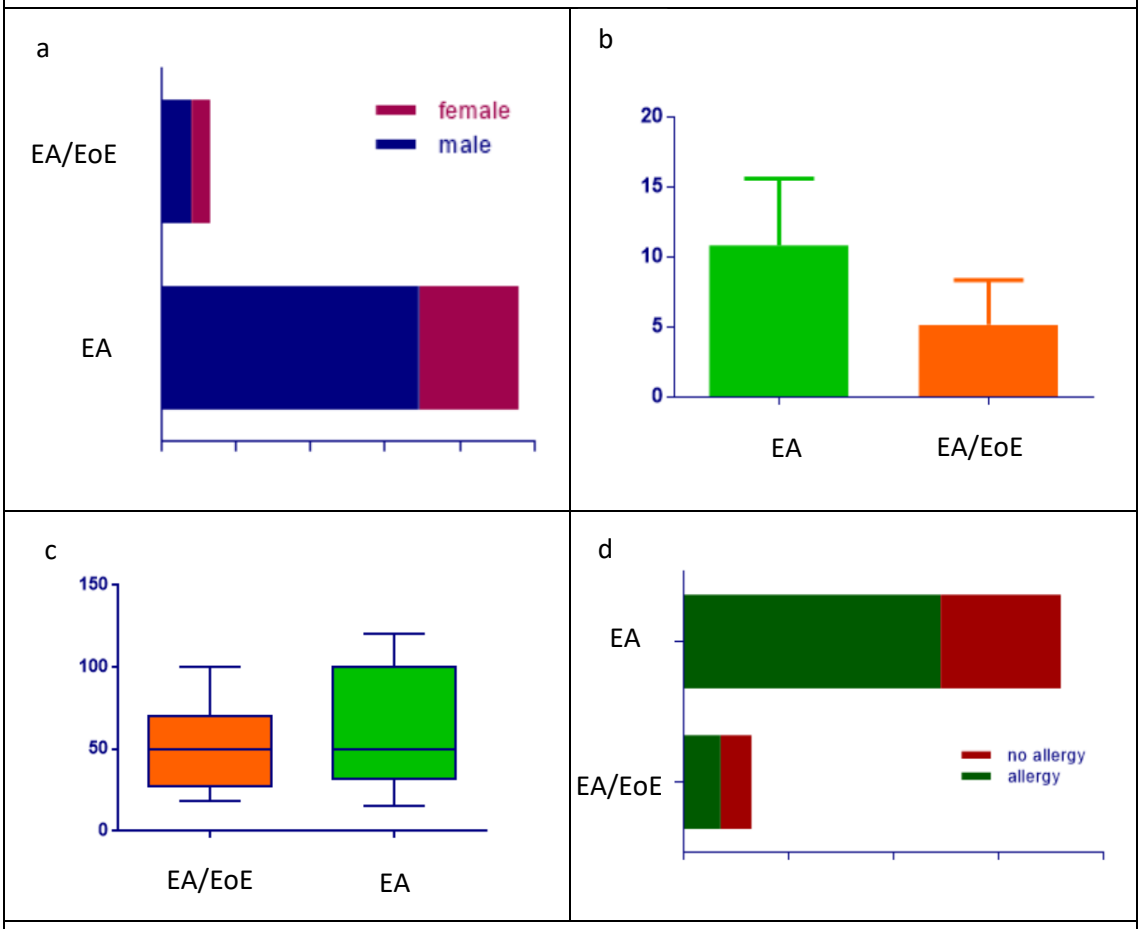
Table 2. Clinical characteristics of EA/EoE and EoE patients

	EA/EoE	EoE	p
Number of patients	15	103	
Male/female (ratio)	10/5 (2.0)	75/28 (2.6)	0.75[a]
Age at EoE diagnosis [years; median (range)]	4 (1.1-12.5)	10.9 (1.7- 23.5)	**<0.0001**[b]
Peak EOS/HPF at diagnosis [number; mean±SD]	50.1±26	59.8±29	0.24[b]
History of allergy [number (percentage)]	8 (53.3)	70 (67.9%)	0.38[a]
PPI-REE [number (percentage)]	10 (66.6)	37 (35.9)	**0.045**[a]

[a] Fisher's exact test; [b]Mann Whitney test; bold text indicates a statistically significant difference
Abbreviation: EA, esophageal atresia; EoE, eosinophilic esophagitis; EOS/HPF, eosinophils per high-power field; PPI-REE, proton pump inhibitor - responsive esophageal eosinophilia; SD, standard deviation.

Overall, among all 118 EoE children, male-to-female prevalence ratio was 2.5 with no difference between groups (2.0 vs 2.6 for EA/EoE and EoE group, respectively; p=0.75). At the time of EoE diagnosis, EA/EoE children were significantly younger than EoE patients from general population (median: 4 vs 10.9 years; p< 0.0001). Peak EOS/HPF did not differ between groups (mean ± standard deviation: 50.1±26 vs 59.8±29 EOS/HPF; p=0.24). Overall, 66.1% of children had allergies with no difference between groups (53.3 vs 67.9%; p=0.38) (Figure 7). PPI-responder patients were significantly more prevalent in EA/EoE group that in EoE group (66.6% vs 35.9%; p=0.045). Among the 5 EA/EoE patients who were non-PPI-responders, 2 achieved clinical and histological remission while on dietary treatment (milk free diet) and 3 on swallowed topical corticosteroid.

Figure 7. Graphical representation of demographic differences between EA and EA/EoE groups



a. Overall, among all 118 EoE children, male-to-female prevalence ratio was 2.5 with no difference between groups (2.0 vs 2.6 for EA/EoE and EoE group, respectively; p=0.75).
b. At the time of EoE diagnosis, EA/EoE children were significantly younger than EoE patients from general population (median: 4 vs 10.9 years; p< 0.0001).
c. Peak EOS/HPF did not differ between groups (mean ± standard deviation: 50.1±26 vs 59.8±29 EOS/HPF; p=0.24).
d. Overall, 66.1% of children had allergies with no difference between groups (53.3 vs 67.9%; p=0.38).

Abbreviation: EA, esophageal atresia; EoE, eosinophilic esophagitis; EOS/HPF, eosinophils per high-power field

2.4 DISCUSSION

This retrospective series is the first from an Italian cohort to evaluate the coexistence of EA and EOE and confirms previous reports from other populations that EA patients are at high risk for developing EoE.

The prevalence of EoE in our cohort of EA survivors (4.0%) was much greater than the 0.1-0.57% estimated in general population (*Papadopoulou 2014*), but lower than previously reported in other populations, ranging from 9.5% to 30% *(Dhaliwal 2014; Krishnan 2019; Petit 2019; Lardenois 2019; Yasuda 2019; Pesce 2019)*. Characteristics and main results of previous series are summarized in Table 3.

Table 3. Details of studies reporting EoE prevalence in EA children

	Study period	Institution (Country)	Total Nr of EA patients	EA/EoE	EoE prevalence	Study design	Patients included
Dhaliwal 2014	1999-2012	SCH (Australia)	103	18	17%	retrospective	All surviving patients who had surgery for EA
Krishnan 2019	2000-2014	SCH (Australia)	110	20	18%	retrospective	#
Petit 2019	2005-2014	CHU Sainte-Justine (Canada)	73	15	21%	prospective	Children born with EA-TEF were prospectively included
Lardenois 2019	2007-2015	University Hospitals of Lille and Strasbourg (France)	63	6	9.5%	prospective	All patients aged 15–20 years with medical history of EA
Yasuda 2019	2016-2018	Boston Children’s Hospital (United States)	310	47	15%*	retrospective	Patients with EA who underwent at least

							one upper endoscopy with biopsies
Pesce 2019	2015-2017	GOSH (United Kingdom) SCH (Australia)	63	19	30%	retrospective	All children with EA referred consecutively either for refractory upper GI symptoms or as part of surveillance program

non clearly detailed, conceivably as the study by Dhaliwal et al. (similar study periods were analyzed)
* Patients who met histologic criteria of > 15 eosinophils/ high powered field
Abbreviation: EA, esophageal atresia; EoE, eosinophilic esophagitis; GOSH, Great Ormond Street Hospital; SCH, Sydney Children’s Hospital

While regional differences between our cohort and others may contribute to some of the observed difference from previous reports, there are some methodological differences between our study and previously reported cohorts. The main factor accounting for this difference is the approach to esophageal biopsy sampling: “routine” versus “selective”. Indeed, we collected biopsies only in symptomatic EA patients without AS and/or with typical endoscopic features of EoE (*Tambucci 2017*) (Figure 4), while other authors performed routine biopsies in all patients, even asymptomatic. It is still not clear whether all EA patients should undergo routine esophageal biopsies to rule out esophageal eosinophilia (EE). Current guidelines on EA recommend, with a low level of evidence, excluding EoE in symptomatic EA patients, especially before anti-reflux surgery (*Krishnan 2016*).

EE does not always mean EoE. By definition, EoE is a clinicopathologic entity, therefore high mucosal eosinophil count must be associated with symptoms related to esophageal dysfunction (*Papadopoulou 2014*). Recently published international consensus on EoE criteria points out that the presence of EE on histologic examination without further consideration of the clinical presentation is not diagnostic of EoE. Authors also highlight that EoE is ultimately diagnosed after excluding other contributing factors for symptoms and EE (*Dellon 2018*). However, the application of the EoE clinical criteria to EA patients is problematic, since esophageal symptoms in EA patients might arise from many different underlying conditions (*Krishnan 2016*). Furthermore, the exclusion of other contributing factors to both symptomatology and histopathology is often impossible (*Yasuda 2019*).

Presumably, esophageal dysmotility plays an important role in this context. Virtually all EA survivors have an impaired esophageal motility (Figure 2), which is the key pathophysiological factor leading to long-term digestive and respiratory morbidity (*Lemoine 2013; Tambucci 2015*). It is conceivable that esophageal dysmotility in EA patients might play a pivotal causative role also in EoE development (*Stave Salgado 2018*). Indeed, esophageal dysmotility increases the risk of severe GERD, but also produces stasis of food and saliva into the esophageal lumen. Prolonged mucosal acid exposure time and retained material into the esophagus, whether it be swallowed food and saliva or refluxed gastric content, might cause itself mucosal injury and esophageal eosinophilic-predominant inflammation (*Little 1986; Kim 2018*). Moreover, esophageal stasis may also result in prolonged exposure to allergens (both aero and food allergens) which facilitates the inflammatory eosinophilic cascade in susceptible patients (*Stave Salgado 2018*). Obviously, the presence of AS and food impaction, by worsening esophageal stasis and clearance, exacerbated these mechanisms.

The topic of AS and its relation to EE and EoE deserves a specific point of discussion. The presence of AS may be a predisposing factor for mucosal eosinophil recruitment, and the presence of eosinophilic inflammation may exacerbate the fibrous remodeling of the anastomosis. AS is the most frequent post-operative complication of EA and has to be first excluded in all symptomatic patients (*Krishnan 2016*). AS may contribute to eosinophil inflammation due to the mechanism described above, such as stasis and retained bolus. Therefore, AS treatment by esophageal dilation may interrupt the chain of events leading to mucosal inflammation, by improving both anterograde and retrograde flow through the esophagus. A recently published case report describing the resolution of EE in a patient with achalasia, who underwent pneumatic dilation of the cardia, supports this speculation (*Frieling 2019*).

On the other hand, patients with EA and EE were described to be at increased risk of recurrent/refractory AS formation (*Krishnan 2015*). The occurrence of recurrent and refractory AS remains a major challenge in the postoperative management of EA patients and exposes patients to several invasive treatments, including surgical procedures (*Tambucci 2017*). Although the pathogenesis of recurrent/refractory AS is not fully understood, it is likely that in a subset of patients the presence of EE, whether it be EoE or not, might have an important role in stricture formation. The use of topical corticosteroids by reversing the subepithelial fibrotic process, might prevent AS recurrence. Therefore, even though our retrospective series does not allow us to draw any definitive conclusion, we do agree with previous reports suggesting that any EA patient with recurrent/refractory AS should undergo esophageal biopsies to rule out EE (*Dhaliwal 2014*). Indeed, treating EE with topical corticosteroids may possibly result in a reduction of AS recurrence, and subsequent need further dilatations and other invasive procedures (*Krishnan 2015*).

In the present study we also aimed to compare demographic and disease characteristics between EA/EoE children and a large group of EoE patients from general population. Consistent with literature data on EoE (*Markowitz 2018*), we found a strong male predominance and a high prevalence of allergy without differences between groups. Moreover, no difference in tissue eosinophilia levels (peak EOS/HPF) was observed.

Similarities in gender distribution, atopic background and histopathological findings suggest that common genetic susceptibility factors might underlie EoE development in EA patients. This hypothesis is corroborated by the study of Krishnan et al. demonstrating a similar gene expression pattern between EoE patients with and without EA (*Krishnan 2019*).

On the other hand, we observed that EoE in EA patients occurred at a younger age than EoE in children from general population. High incidence and early onset of EoE in EA children might be related to multiple factors (*Stave Salgado 2018*). Early endoscopic surveillance in EA children might enable early diagnosis of EoE. Furthermore, mutations in the Forkhead box (FOX) gene might constitute a possible genetic link between EA and EoE, as suggested by Gorter et (*Gorter 2012*). However, the above-mentioned motility impairment is likely to be primarily implicated in enhancing the propensity to EoE in EA patients, as supported by the high percentage of LGEA in our EA/EoE population (40%).

Interestingly, in our cohort of EA/EoE children we observed a significantly higher percentage of PPI-responders than children with EoE from general population. Different mechanisms have been proposed to explain the PPI response in EoE (*Dellon 2018; Gutiérrez-Junquera 2018*). PPIs mainly exert their effect by reducing the gastric acid secretion. Therefore, in patients with EE secondary to GERD, by restoring the acid reflux-induced impairment of mucosal integrity, PPIs correct the abnormal mucosal

permeability and prevent antigen penetration (*van Rhijn 2014*). Gastric acid-inhibiting effect is likely to have a significant role in resolving EE in EA children because of their considerably higher risk of developing severe GERD. In support of this, Yasuda et al. point out that only 6 out of 31 EA patients with EE received swallowed corticosteroid or dietary elimination while the others responded to interventions maximizing reflux treatment (*Yasuda 2019*). Moreover, the study by Pesce et al. showed that EA children with EE exhibited a significantly prolonged esophageal acid exposure time as measured by pH-impedance monitoring, suggesting that inhibition of acid secretion plays an important role in this population (*Pesce 2019*). Beside antisecretory action, PPIs have been shown to also exert anti-inflammatory effects (*Dellon 2018*) by inhibiting the production of pro-inflammatory cytokines and adhesion molecules that act as ligands on the eosinophil cell surface (*Kedika 2009*). In EoE, PPIs reduce eosinophil recruitment by inhibiting T-helper 2 cytokine-induced eotaxin-3 expression in esophageal epithelial cells (*Cheng 2013; Cheng 2016*). Of course, as for EoE from general population, these anti-inflammatory properties of PPIs can also explain PPI-REE observed in EA patients without evidence of GERD.

This study has some limitations to be considered. First, our findings may be somewhat limited by the retrospective nature. On the other hand, our study analyzed the largest cohort ever reported of patients with EA (370) and EoE (118). The “selective” approach in performing esophageal biopsies likely underestimates the prevalence of EE in EA patients. In fact, according to previously reported findings from studies using the “routine” approach we have possibly missed some EE diagnosis. On the other hand, as point out by Yasuda et al. “eosinophil count alone in EA patients to diagnose EoE as previously reported is likely insufficient” (*Yasuda 2019*). Indeed, EA patients labeled as having EoE in previous “routine” studies might not meet the clinical criteria or may not

have received evaluation to exclude other possible contributing causes for symptoms and EE, and therefore may overestimate the prevalence of EoE in EA. Perhaps our cohort more accurately reflects the risk compared to the considerably higher risks detailed in previous reports. In our view, the “*a posteriori*” analysis over a long period (13 years) of symptomatic EA children without AS, could represent the most reliable picture of the relationship between EA and “true” EoE. In particular, the possible presence of EE in patients showing sustained symptom relief after AS resolution would not meet the clinical criteria to diagnose EoE, because symptoms of esophageal dysfunction resolved after dilation.

In summary, our study confirms in our Italian cohort that EA patients are more prone to develop EoE than general population, but notably estimates a lower risk than what has been seen in previous case series. Established risk factors for EoE, such as male gender and history of atopy, may contribute to EoE development in EA patients as with the general population. However, our study suggests that adapted criteria for EoE diagnosis should be developed for EA patients. Indeed, while underdiagnosis of EoE may occur if routine biopsies are not obtained in all patients with EA (*Krishnan 2016*), overdiagnosis may also occur if eosinophilia is present, but symptoms of esophageal dysfunction relate to complications of EA rather than EoE. It should be kept in mind that EoE is a specific chronic condition carrying a significant burden of disease, as it requires intensive monitoring and long-term medications or dietary restrictions (*Dellon 2018*_**2**). Moreover, the high prevalence of PPI-REE in our EA/EoE population re-emphasizes the importance of a PPI-trial that has been recently removed from the diagnostic algorithm for EoE (*Dellon 2018*). In fact, our results suggest that PPIs should at least be considered as initial treatment in any EA patient showing EE.

Growing evidence indicates that EoE is an umbrella term for conditions that are unified by EE but that different disease subgroups with various inflammatory esophageal patterns and/or different clinical features exist (*Mudde 2016*). Our study supports the concept that EoE in EA represent a specific subtype of EoE and strongly sustains the vision toward tailored treatment strategies according to different EoE phenotypes (*Mudde 2016*). Future research should devote more attention about the role of EE, whether it be EoE or not, in EA, especially in patients experiencing recurrent/refractory AS.

3. REFERENCES

Alexander ES, Martin LJ, Collins MH, Kottyan LC, Sucharew H, He H et al. Twin and family studies reveal strong environmental and weaker genetic cues explaining heritability of eosinophilic esophagitis. J Allergy Clin Immunol. 2014;134:1084-1092.e1.

Alfaro L, Bermas H, Fenoglio M, Parker R, Janik JS. Are patients who have had a tracheoesophageal fistula repair during infancy at risk for esophageal adenocarcinoma during adulthood? J Pediatr Surg. 2005;40:719-20.

Allin B, Knight M, Johnson P, Burge D; BAPS-CASS. Outcomes at one-year post anastomosis from a national cohort of infants with oesophageal atresia. PLoS One. 2014;9:e106149.

ASGE Technology Committee, Siddiqui UD, Banerjee S, Barth B, Chauhan SS, Gottlieb KT et al. Tools for endoscopic stricture dilation. Gastrointest Endosc. 2013;78:391-404.

Bagolan P, Valfrè L, Morini F, Conforti A. Long-gap esophageal atresia: traction-growth and anastomosis—before and beyond. Dis Esophagus. 2013;26:372-9.

Baird R, Laberge JM, Lévesque D. Anastomotic stricture after esophageal atresia repair: a critical review of recent literature. Eur J Pediatr Surg. 2013;23:204-13.

Biller JA, Allen JL, Schuster SR, Treves ST, Winter HS. Long-term evaluation of esophageal and pulmonary function in patients with repaired esophageal atresia and tracheoesophageal fistula. Dig Dis Sci. 1987;32:985-90.

Bogs T, Zwink N, Chonitzki V, Hölscher A, Boemers TM, Münsterer O et al. Esophageal Atresia with or without Tracheoesophageal Fistula (EA/TEF): Association of Different EA/TEF Subtypes with Specific Co-occurring Congenital Anomalies and Implications for Diagnostic Workup. Eur J Pediatr Surg. 2018;28:176-182.

Burjonrappa SC, Youssef S, St-Vil D. What is the incidence of Barrett's and gastric metaplasia in esophageal atresia/tracheoesophageal fistula (EA/TEF) patients? Eur J Pediatr Surg. 2011;21:25-9.

Cassina M, Ruol M, Pertile R, Midrio P, Piffer S, Vicenzi V et al. Prevalence, characteristics, and survival of children with esophageal atresia: A 32-year population-based study including 1,417,724 consecutive newborns. Birth Defects Res A Clin Mol Teratol. 2016;106:542-8.

Castilloux J, Bouron-Dal Soglio D, Faure C. Endoscopic assessment of children with esophageal atresia: Lack of relationship of esophagitis and esophageal metaplasia to symptomatology. Can J Gastroenterol. 2010;24:312-6.

Castilloux J, Noble AJ, Faure C. Risk factors for short- and long-term morbidity in children with esophageal atresia. J Pediatr 2010;156:755–60.

Catalano P, Di Pace MR, Caruso AM, Casuccio A, De Grazia E. Gastroesophageal reflux in young children treated for esophageal atresia: evaluation with pH-multichannel intraluminal impedance. J Pediatr Gastroenterol Nutr. 2011;52:686-90.

Cheng E, Zhang X, Huo X, Yu C, Zhang Q, Wang DH, et al. Omeprazole blocks eotaxin-3 expression by oesophageal squamous cells from patients with eosinophilic oesophagitis and GORD. Gut. 2013;62:824-32.

Cheng E, Zhang X, Wilson KS, Wang DH, Park JY, Huo X, et al. JAKSTAT6 pathway inhibitors block eotaxin-3 secretion by epithelial cells and fibroblasts from esophageal eosinophilia patients: promising agents to improve inflammation and prevent fibrosis in EoE. PLoS One. 2016;11:e0157376.

Connor MJ, Springford LR, Kapetanakis VV, Giuliani S. Esophageal atresia and transitional care--step 1: a systematic review and meta-analysis of the literature to define the prevalence of chronic long-term problems. Am J Surg. 2015;209:747-59.

Davidson AJ, Disma N, de Graaff JC, Withington DE, Dorris L, Bell G et al. Neurodevelopmental outcome at 2 years of age after general anaesthesia and awake-regional anaesthesia in infancy (GAS): an international multicentre, randomised controlled trial. Lancet. 2016;387:239-50.

Dellon ES, Hirano I. Epidemiology and Natural History of Eosinophilic Esophagitis. Gastroenterology. 2018;154:319-32.e3.**_2**

Dellon ES, Liacouras CA, Molina-Infante J, Furuta GT, Spergel JM, Zevit N, et al. Updated International Consensus Diagnostic Criteria for Eosinophilic Esophagitis: Proceedings of the AGREE Conference. Gastroenterology. 2018;155:1022-33.e10.

Dhaliwal J, Tobias V, Sugo E, Varjavandi V, Lemberg D, Day A, et al. Eosinophilic esophagitis in children with esophageal atresia. Dis Esophagus. 2014;27:340-7.

Di Pace MR, Caruso AM, Catalano P, Casuccio A, Cimador M, De Grazia E. Evaluation of esophageal motility and reflux in children treated for esophageal atresia with the use of combined multichannel intraluminal impedance and pH monitoring. J Pediatr Surg. 2011;46:443-51.

Donoso F, Kassa AM, Gustafson E, Meurling S, Lilja HE. Outcome and management in infants with esophageal atresia - A single centre observational study. J Pediatr Surg. 2016;51:1421-5.

Duranceau A, Fisher SR, Flye M, Jones RS, Postlethwait RW, Sealy WC. Motor function of the esophagus after repair of esophageal atresia and tracheoesophageal fistula. Surgery. 1977;82:116-23.

Dutta HK, Grover VP, Dwivedi SN, Bhatnagar V. Manometric evaluation of postoperative patients of esophageal atresia and tracheo-esophageal fistula. Eur J Pediatr Surg. 2001;11:371-6.

Dutta HK, Rajani M, Bhatnagar V. Cineradiographic evaluation of postoperative patients with esophageal atresia and tracheoesophageal fistula. Pediatr Surg Int. 2000;16:322-5.

Faure C, Righini Grunder F. Dysmotility in Esophageal Atresia: Pathophysiology, Characterization, and Treatment. Front Pediatr. 2017;5:130.

Frieling T, Heise J, Kreysel C, Blank M, Hemmerlein B, Beccu L, et al. Eosinophilic esophagitis and achalasia - just a coincidence? Z Gastroenterol. 2019;57:151-5.

Gibreel W, Zendejas B, Antiel RM, Fasen G, Moir CR, Zarroug AE. Swallowing Dysfunction and Quality of Life in Adults With Surgically Corrected Esophageal Atresia/Tracheoesophageal Fistula as Infants: Forty Years of Follow-up. Ann Surg. 2017;266:305-310.

Gorter RR, Heij HA, van der Voorn JP, Kneepkens CM. Eosinophilic esophagitis after esophageal atresia: is there an association? Case presentation and literature review. J Pediatr Surg. 2012;47: e9-e13.

Gottrand M, Michaud L, Sfeir R, Gottrand F. Motility, digestive and nutritional problems in Esophageal Atresia. Paediatr Respir Rev. 2016;19:28-33.

Gross RE. The Surgery of Infancy and Childhood: Its Principles and Techniques. Philadelphia: Saunders Company; 1953.

Gupta SK, Hassall E, Chiu YL, Amer F, Heyman MB. Presenting symptoms of nonerosive and erosive esophagitis in pediatric patients. Dig Dis Sci. 2006;51:858-63.

Gutiérrez-Junquera C, Fernández-Fernández S, Cilleruelo ML, Rayo A, Román E. The Role of Proton Pump Inhibitors in the Management of Pediatric Eosinophilic Esophagitis. Front Pediatr. 2018;6:119.

Hörmann M, Pokieser P, Scharitzer M, Pumberger W, Memarsadeghi M, Partik B et al. Videofluoroscopy of deglutition in children after repair of esophageal atresia. Acta Radiol. 2002;43:507-10.

Hsieh H, Frenette A, Michaud L, Krishnan U, Dal-Soglio DB, Gottrand F et al. Intestinal Metaplasia of the Esophagus in Children With Esophageal Atresia. J Pediatr Gastroenterol Nutr. 2017;65:e1-e4.

Huynh Trudeau V, Maynard S, Terzic T, Soucy G, Bouin M. Dysphagia among adult patients who underwent surgery for esophageal atresia at birth. Can J Gastroenterol Hepatol. 2015;29:91-4.

Ijsselstijn H, van Beelen NW, Wijnen RM. Esophageal atresia: long-term morbidities in adolescence and adulthood. Dis Esophagus. 2013;26:417-21.

Kawahara H, Kubota A, Hasegawa T, Okuyama H, Ueno T, Watanabe T et al. Lack of distal esophageal contractions is a key determinant of gastroesophageal reflux disease after repair of esophageal atresia. J Pediatr Surg. 2007;42:2017-21.

Kawahara H1, Kubota A, Hasegawa T, Okuyama H, Ueno T, Watanabe T et al. Lack of distal esophageal contractions is a key determinant of gastroesophageal reflux disease after repair of esophageal atresia. J Pediatr Surg. 2007;42:2017-21.

Kedika RR, Souza RF, Spechler SJ. Potential anti-inflammatory effects of proton pump inhibitors: a review and discussion of the clinical implications. Dig Dis Sci 2009;54:2312-7.

Kim H, Park H, Choi H, Shin Y, Park H, Youn YH, et al. Retention Esophagitis as a Significant Clinical Predictor of Progression to Esophageal Cancer in Achalasia. Clin Endosc. 2018;51:161-6.

Koivusalo A, Pakarinen MP, Rintala RJ. The cumulative incidence of significant gastrooesophageal reflux in patients with oesophageal atresia with a distal fistula--a systematic clinical, pH-metric, and endoscopic follow-up study. J Pediatr Surg. 2007;42:370-4.

Kovesi T, Rubin S. Long-term complications of congenital esophageal atresia and/or tracheoesophageal fistula. Chest 2004;126:915-25.

Krishnan U, Lijuan C, Andrew GJ, Rothenberg ME, Wen T. Analysis of eosinophilic esophagitis in children with repaired congenital esophageal atresia. J Allergy Clin Immunol 2019;143:1455-64.e2.

Krishnan U, Mousa H, Dall'Oglio L, Homaira N, Rosen R, Faure C et al. ESPGHAN-NASPGHAN Guidelines for the Evaluation and Treatment of Gastrointestinal and Nutritional Complications in Children With Esophageal Atresia-Tracheoesophageal Fistula. J Pediatr Gastroenterol Nutr. 2016;63:550-570.

Krishnan U. Eosinophilic Esophagitis in Children with Esophageal Atresia. Eur J Pediatr Surg. 2015;25:336-44.

Krishnan U. Eosinophilic Esophagitis in Esophageal Atresia. Front Pediatr. 2019 29;7:497. **_2**

Krug E, Bergmeijer JH, Dees J, de Krijger R, Mooi WJ, Hazebroek FW. Gastroesophageal reflux and Barrett's esophagus in adults born with esophageal atresia. Am J Gastroenterol. 1999;94:2825-8.

Lardenois E, Michaud L, Schneider A, Onea M, Rebeuh J, Gottrand-Aumar M, et al. Prevalence of Eosinophilic Esophagitis in Adolescents with Esophageal Atresia. J Pediatr Gastroenterol Nutr. 2019;69:52-56.

Legrand C, Michaud L, Salleron J, Neut D, Sfeir R, Thumerelle C et al. Long-term outcome of children with oesophageal atresia type III. Arch Dis Child. 2012;97:808-11.

Lemoine C, Aspirot A, Le Henaff G, Piloquet H, Lévesque D, Faure C. Characterization of esophageal motility following esophageal atresia repair using high-resolution esophageal manometry. J Pediatr Gastroenterol Nutr. 2013;56:609-14.

Lilja HE, Wester T. Outcome in neonates with esophageal atresia treated over the last 20 years. Pediatr Surg Int. 2008;24:531-6.

Lindahl H, Rintala R. Long-term complications in cases of isolated esophageal atresia treated with esophageal anastomosis. J Pediatr Surg. 1995;30:1222-3.

Little AG, Correnti FS, Calleja IJ, Montag AG, Chow YC, Ferguson MK, et al. Effect of incomplete obstruction on feline esophageal function with a clinical correlation. Surgery 1986;100:430-6.

Little DC, Rescorla FJ, Grosfeld JL, West KW, Scherer LR, Engum SA. Long-term analysis of children with esophageal atresia and tracheoesophageal fistula. J Pediatr Surg. 2003;38:852-6.

Lucendo AJ, Molina-Infante J, Arias Á, von Arnim U, Bredenoord AJ, Bussmann C et al. Guidelines on eosinophilic esophagitis: evidence-based statements and recommendations for diagnosis and management in children and adults. United European Gastroenterol J. 2017;5:335-358.

Markowitz JE, Clayton SB. Eosinophilic Esophagitis in Children and Adults. Gastrointest Endosc Clin N Am. 2018;28:59-75.

Menzies J, Hughes J, Leach S, Belessis Y, Krishnan U. Prevalence of malnutrition and feeding difficulties in children with esophageal atresia. J Pediatr Gastroenterol Nutr. 2017;64:e100-e105.

Miehlke S. Clinical features of eosinophilic esophagitis. Dig Dis. 2014;32:61-7.

Moawad FJ. Eosinophilic Esophagitis: Incidence and Prevalence. Gastrointest Endosc Clin N Am. 2018;28:15-25.

Montgomery M, Witt H, Kuylenstierna R, Frenckner B. Swallowing disorders after esophageal atresia evaluated with videomanometry. J Pediatr Surg. 1998;33:1219-23.

Morini F, Conforti A, Bagolan P. Perioperative Complications of Esophageal Atresia. Eur J Pediatr Surg. 2018;28:133-140.

Morini F, Iacobelli BD, Crocoli A, Bottero S, Trozzi M, Conforti A, Bagolan P. Symptomatic vocal cord paresis/paralysis in infants operated on for esophageal atresia and/or tracheo-esophageal fistula. J Pediatr. 2011;158:973-6.

Mortellaro VE, Pettiford JN, St Peter SD, Fraser JD, Ho B, Wei J. Incidence, diagnosis, and outcomes of vocal fold immobility after esophageal atresia (EA) and/or tracheoesophageal fistula (TEF) repair. Eur J Pediatr Surg. 2011;21:386-8.

Mousa H, Krishnan U, Hassan M, Dall'Oglio L, Rosen R, Gottrand F et al. How to Care for Patients with EA-TEF: The Known and the Unknown. Curr Gastroenterol Rep. 2017;19:65.

Mudde AC, Lexmond WS, Blumberg RS, Nurko S, Fiebiger E. Eosinophilic esophagitis: published evidences for disease subtypes, indications for patient subpopulations, and how to translate patient observations to murine experimental models. World Allergy Organ J. 2016;9:23.

Nassar N, Leoncini E, Amar E, Arteaga-Vázquez J, Bakker MK, Bower C et al. Prevalence of esophageal atresia among 18 international birth defects surveillance programs. Birth Defects Res A Clin Mol Teratol. 2012;94:893-9.

Orringer MB, Kirsh MM, Sloan H. Long-term esophageal function following repair of esophageal atresia. Ann Surg. 1977;186:436-43.

Papadopoulou A, Koletzko S, Heuschkel R, Dias JA, Allen KJ, Murch SH et al. Management guidelines of eosinophilic esophagitis in childhood. J Pediatr Gastroenterol Nutr. 2014;58:107-18.

Pedersen RN, Calzolari E, Husby S, Garne E; EUROCAT Working group. Oesophageal atresia: prevalence, prenatal diagnosis and associated anomalies in 23 European regions. Arch Dis Child. 2012;97:227-32.

Pedersen RN, Markøw S, Kruse-Andersen S, Qvist N, Hansen TP, Gerke O et al. Esophageal atresia: gastroesophageal functional follow-up in 5-15 year old children. J Pediatr Surg. 2013;48:2487-95.

Pesce M, Krishnan U, Saliakellis E, Lopez R, Lindley KJ, Thapar N, et al. Is There a Role for pH Impedance Monitoring in Identifying Eosinophilic Esophagitis in Children with Esophageal Atresia? J Pediatr. 2019;210:134-140.

Petit LM, Righini F, Ezri J, Jantchou P, Aspirot A, Soglio DD, et al Prevalence and Predictive Factors of Histopathological Complications in Children with Esophageal Atresia. Eur J Pediatr Surg. 2019;29:510-515.

Porcaro F, Valfré L, Aufiero LR, Dall'Oglio L, De Angelis P, Villani A et al. Respiratory problems in children with esophageal atresia and tracheoesophageal fistula. Ital J Pediatr. 2017;43:77.

Powell L, Frawley J, Crameri J, Teague WJ, Frawley GP et al. Oesophageal atresia: Are "long gap" patients at greater anesthetic risk? Paediatr Anaesth. 2018;28:249-256.

Pultrum BB, Bijleveld CM, de Langen ZJ, Plukker JT. Development of an adenocarcinoma of the esophagus 22 years after primary repair of a congenital atresia. J Pediatr Surg. 2005;40:e1-4.

Rintala RJ, Pakarinen MP. Long-term outcome of esophageal anastomosis. Eur J Pediatr Surg 2013;23:219-25.

Romeo C, Bonanno N, Baldari S, Centorrino A, Scalfari G, Antonuccio P et al. Gastric motility disorders in patients operated on for esophageal atresia and tracheoesophageal fistula: long-term evaluation. J Pediatr Surg. 2000;35:740-4.

Sanlaville D, Verloes A. CHARGE syndrome: an update. Eur J Hum Genet. 2007;15:389-99.

Schneider A, Michaud L, Gottrand F. Prevalence of Barrett Esophagus in Adolescents and Young Adults With Esophageal Atresia. Ann Surg. 2017;266:e96.

Shah R, Varjavandi V, Krishnan U. Predictive factors for complications in children with esophageal atresia and tracheoesophageal fistula. Dis Esophagus. 2015;28:216-23.

Shaw-Smith C. Oesophageal atresia, tracheo-oesophageal fistula, and the VACTERL association: review of genetics and epidemiology. J Med Genet. 2006;43:545-54.

Sistonen SJ, Koivusalo A, Nieminen U, Lindahl H, Lohi J, Kero M et al. Esophageal morbidity and function in adults with repaired esophageal atresia with tracheoesophageal fistula: a population-based long-term follow-up. Ann Surg. 2010;251:1167-73.

Solomon BD. VACTERL/VATER Association. Orphanet J Rare Dis. 2011;6:56.

Spitz L. Esophageal atresia. Lessons I have learned in a 40-year experience. J Pediatr Surg. 2006;41:1635-40.

Spitz L. Oesophageal atresia. Orphanet J Rare Dis. 2007;2:24.

Spoel M, Meeussen CJ, Gischler SJ, Hop WC, Bax NM, Wijnen RM et al. Respiratory morbidity and growth after open thoracotomy or thoracoscopic repair of esophageal atresia. J Pediatr Surg. 2012;47:1975-83.

Stave Salgado KV, Rocca AM. Eosinophilic esophagitis and esophageal atresia: coincidence or causality? Arch Argent Pediatr. 2018;116:e61-e69.

Stoll C, Alembik Y, Dott B, Roth MP. Associated anomalies in cases with esophageal atresia. Am J Med Genet A. 2017;173:2139-2157.

Sulkowski JP, Cooper JN, Lopez JJ, Jadcherla Y, Cuenot A, Mattei P et al. Morbidity and mortality in patients with esophageal atresia. Surgery. 2014;156:483-91.

Tambucci R, Angelino G, De Angelis P, Torroni F, Caldaro T, Balassone V et al. Anastomotic Strictures after Esophageal Atresia Repair: Incidence, Investigations, and Management, Including Treatment of Refractory and Recurrent Strictures. Front Pediatr. 2017;5:120.

Tambucci R, Thapar N, Saliakellis E, Pescarin M, Quitadamo P, Cristofori F et al. Clinical relevance of esophageal baseline impedance measurement: just an innocent bystander. J Pediatr Gastroenterol Nutr. 2015;60:776-82.

Taylor AC, Breen KJ, Auldist A, Catto-Smith A, Clarnette T, Crameri J et al. Gastroesophageal reflux and related pathology in adults who were born with esophageal atresia: a long-term follow-up study. Clin Gastroenterol Hepatol. 2007;5:702-6.

Thomson M, Tringali A, Dumonceau JM, Tavares M, Tabbers MM, Furlano R, et al. Paediatric gastrointestinal endoscopy: European Society for Paediatric Gastroenterology Hepatology and Nutrition and European Society of Gastrointestinal Endoscopy Guidelines. J Pediatr Gastroenterol Nutr. 2017;64:133-53.

Tong S, Mallitt KA, Krishnan U. Evaluation of Gastroesophageal Reflux by Combined Multichannel Intraluminal Impedance and pH Monitoring and Esophageal Motility Patterns in Children with Esophageal Atresia. Eur J Pediatr Surg. 2016;26:322-31.

Tovar JA, Diez Pardo JA, Murcia J, Prieto G, Molina M, Polanco I. Ambulatory 24-hour manometric and pH metric evidence of permanent impairment of clearance capacity in patients with esophageal atresia. J Pediatr Surg. 1995;30:1224-31.

van der Zee DC, Bagolan P, Faure C, Gottrand F, Jennings R, Laberge JM et al. Position Paper of INoEA Working Group on Long-Gap Esophageal Atresia: For Better Care. Front Pediatr. 2017;5:63.

van Lennep M, Singendonk MMJ, Dall'Oglio L, Gottrand F, Krishnan U, Terheggen-Lagro SWJ et al. Oesophageal atresia. Nat Rev Dis Primers. 2019;5(1):26.

van Rhijn BD, Weijenborg PW, Verheij J, van den Bergh Weerman MA, Verseijden C, van den Wijngaard RM, et al. Proton pump inhibitors partially restore mucosal integrity in patients with proton pump inhibitor-responsive esophageal eosinophilia but not eosinophilic esophagitis. Clin Gastroenterol Hepatol 2014;12:1815-23.e2.

van Wijk M, Knüppe F, Omari T, de Jong J, Benninga M. Evaluation of gastroesophageal function and mechanisms underlying gastroesophageal reflux in infants and adults born with esophageal atresia. J Pediatr Surg. 2013;48:2496-505.

Vergouwe FW, Gottrand M, Wijnhoven BP, IJsselstijn H, Piessen G, Bruno MJ et al. Four cancer cases after esophageal atresia repair: Time to start screening the upper gastrointestinal tract. World J Gastroenterol. 2018;24:1056-1062.

Vergouwe FWT, IJsselstijn H, Biermann K, Erler NS, Wijnen RMH, Bruno MJ et al. High Prevalence of Barrett's Esophagus and Esophageal Squamous Cell Carcinoma After Repair of Esophageal Atresia. Clin Gastroenterol Hepatol. 2018;16:513-521.e6. **_2**

Vergouwe FWT, Vlot J, IJsselstijn H, Spaander MCW, van Rosmalen J, Oomen MWN et al. Risk factors for refractory anastomotic strictures after oesophageal atresia repair: a multicentre study. Arch Dis Child. 2019;104:152-157.

Vogt EC. Congenital esophageal atresia. Am J Roentgenol. 1929; 22: 463-465.

Wyse RK, al-Mahdawi S, Burn J, Blake K. Congenital heart disease in CHARGE association. Pediatr Cardiol. 1993;14:75-81.

Yalcin S, Demir N, Serel S, Soyer T, Tanyel FC. The evaluation of deglutition with videofluoroscopy after repair of esophageal atresia and/or tracheoesophageal fistula. J Pediatr Surg. 2015;50:1823-7.

Yasuda JL, Clark SJ, Staffa SJ, Blansky B, Ngo PD, Hamilton TE, et al. Esophagitis in Pediatric Esophageal Atresia: Acid May Not Always Be the Issue. J Pediatr Gastroenterol Nutr. 2019;69:163-170.