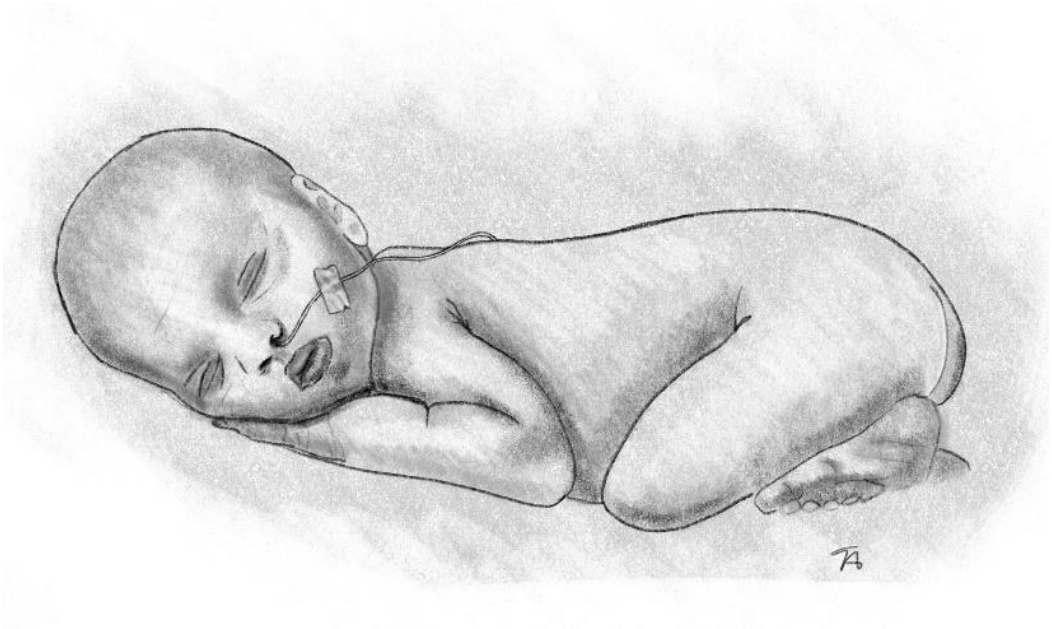
Prenatal diagnosis of esophageal atresia

Literature study

Arntzen T.



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Abbreviations

Abbreviation	*EXPLANATION*
AFI	Amniotic fluid index
AFP	Alpha Feto protein
AMP	Aminopeptidase
Ab	Abstract
CHARGE	Syndrome characterized by: Coloboma of the eye, Heart defects, Atresia of the choana, Growth restriction and Ear abnormalities
EA/OA	Esophageal/oesophageal atresia
F	False positives
GA	Gestational age
GGTP	Gamma-glutamyl transpeptidase
GERD	Gastroesophageal reflux disease
iALP	Intestinal alkaline phosfatase
Kw	Keyword
MRI	Magnetic resonance imaging
MSAFP	Maternal Serum alpha fetoprotein
NIPT	Non-invasive prenatal test
NPV	Negative predictive value
PPV	Positive predictive value
Se	Sensitivity
Sp	Specificity
TEF	Tracheoesophageal fistula
Ti	Title
VACTERL syndrome	Syndrome characterized by anomalies in the following body partsVertebrae, Anorectal, Cardiac, Tracheal, Esophageal, Renal and Limbs

Summary

Background:

Esophageal atresia (EA) is a congenital malformation affecting the esophagus and trachea. Prenatal diagnosis is a diagnosis set prior to birth. Ultrasound, MRI and biochemical analyses are the chosen modalities used in the prenatal diagnosis of EA. The benefits of prenatal diagnosis include parental information and counselling, scheduled delivery, and early treatment. In this study, we aimed to describe the trends in the research field, the prevalence of prenatal diagnosis in EA patients, and the modalities and signs used to diagnose EA prenatally.

Method:

We searched for literature in PubMed and Cochrane databases published between 1990 and 2020. We screened the papers according to our predefined criteria. We analyzed the data with a focus on trends, prevalence, modalities and prenatal signs.

Results

Of the 376 papers screened, only 33 were eligible. The 30 years of publications yielded an average of one publication per year. One third of the papers were published in 2014 and 2015. One third of the papers originated in France, which is the country that produced most publications. Ultrasound is the most commonly used modality, as reported in 90% of the papers. Prenatal EA detection has a sensitivity (Se) of 10-40%, which has not changed throughout the search period. In patients with type A the sensitivity is 85-100%.

Using ultrasound, polyhydramnios is a sign with a sensitivity of 56% and specificity (Sp) of 61%. Small or absent stomach is a sign with Se of 67% and Sp of 50%. Both signs are indicators that lead to further examinations. The pouch sign (dilated esophageal pouch) has a Se of 40-62% and Sp of 97-100%.

The signs included in an MRI diagnosis are small or absent stomach, the pouch sign and an incomplete visualization of the esophagus. MRI diagnosis has a Se of 50-100% and Sp of 82-100%.

The biochemical analyses are performed in maternal serum and/or amniotic fluid samples. Alpha fetoprotein measured in maternal serum has a Se of 19% for detecting EA. Amniotic fluid is analyzed using the EA index (AFP*GGTP). The EA index has a Se of 88-98% and Sp of 60-100%.

The combination of ultrasound, MRI and amniocentesis, with the requirement of at least 2 out of 3 present, has a Se of 80% and Sp of 100%.

Conclusion:

Esophageal atresia is challenging to diagnose prenatally. Ultrasound is the main screening tool, and the combination of ultrasound, MRI and amniocentesis improve the sensitivity and specificity of the diagnosis.

Preface

Esophageal atresia is a congenital malformation that is difficult to diagnose prenatally. Throughout time, different methods have been applied to detect the malformation prenatally. Prenatal diagnostics is an evolving field with ethical difficulties. As technology and diagnostic methods evolve, new ethical problems arise. This is what first drew me towards research on prenatal diagnostics of esophageal atresia.

I would like to thank my supervisors Audun Mikkelsen, Guttorm Haugen and Ragnhild Emblem. Their support and advices are invaluable. I would also like to thank my aunt, Noelle, for helping me to improve my writing.

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1 Introduction

1.1 Esophageal atresia

1.1.1 What is Esophageal Atresia?

Esophageal atresia (EA) is a congenital malformation affecting the esophagus. A child born with EA has a discontinuous esophagus and/or a tracheoesophageal fistula (TEF). In a paper by Spitz from 2007, the definition of EA is *"Oesophageal atresia encompasses a group of congenital anomalies comprising an interruption of the continuity of the oesophagus combined with or without a persistent communication with the trachea."* (1). In patients with EA, the communication between the esophagus and the stomach is interrupted. The discontinuity of the esophagus results in difficulties to swallow both food and liquids, including saliva. When the patient cannot swallow the saliva, it will eventually enter the airways and the patient may suffocate. EA is a serious malformation if not treated.

1.1.2 Prevalence of Esophageal Atresia

Studies report a prevalence of 2-4 cases per 10.000 births (2-4). In Norway 55.000 babies are born each year which equals an expected annual birth rate of 11-22 patients with EA (5).

1.1.3 Etiology of Esophageal Atresia

EA is a congenital malformation of the esophagus. The etiology of EA is still predominantly unknown (6). Some studies have aimed to identify factors that influence the likelihood of having a child born with EA, such as smoking, obesity and socioeconomic status. So far, none seems to be significant (7).

1.1.4 Classification of Esophageal atresia

Five different types of EA are described by Ladd and Gross (8, 9). The most common classification currently used in Europe is Gross classification, as seen in figure 1.1.4. Type A is often referred to as a pure EA since there is no TEF. Type B has an upper TEF, type C has a lower TEF and type D has both an upper and lower TEF. Type E has no atresia, but a TEF. For type A and B, the distance between the two esophageal ends tend to be longer due to the absence of a lower fistula connected to the stomach. If there is a longer gap between the two esophageal ends, the malformation is referred to as Long gap EA. Type C is the most frequent type of EA, identified in approximately 85% of the patients with EA (2, 10, 11).

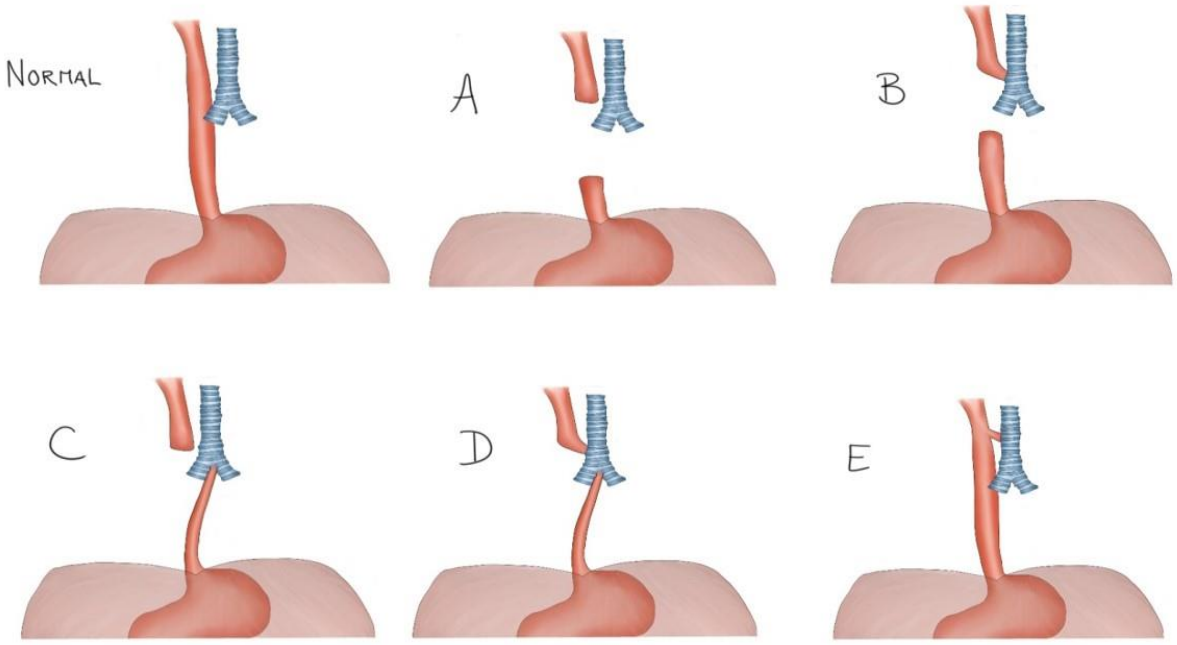

Figure 1.1.4: *Gross classification of Esophageal Atresia*

1.1.5 Postnatal signs and diagnosis of Esophageal Atresia

After birth there may be several signs of EA. Due to the discontinuity of the esophagus, the saliva will build up in the upper esophageal pouch. In neonates with EA, this results in excess saliva in the mouth, often observed as frothing. The saliva enters the airways resulting in breathing difficulties and cyanotic spells. When the child is fed, it is likely to vomit or aspirate. The main concern for the newborn EA patient is aspiration of saliva and gastric juices resulting in aspiration pneumonia, breathing difficulties, and in worst case, respiratory failure resulting in death.

When a neonate has a foamy mouth and vomits every mouthful of milk/formula fed to him/her, most doctors will try to insert a nasogastric tube. In patients with EA the nasogastric tube typically stops and coils up in the upper esophageal pouch. With chest X-ray you may identify the nasogastric tube and find that it is coiled up in the upper esophageal pouch. Another important observation on the X-ray is the detection of gas in the stomach and small intestines or not. Gas will only appear in EA patients with a lower tracheoesophageal fistula. In patients without a lower fistula, the abdomen is gasless. Both findings support the EA diagnosis.

1.1.6 Associated anomalies

Half of the patients with EA have additional anomalies and comorbidities (1, 12). One of the most common associated syndrome is VACTERL syndrome. The definition of VACTERL is the presence of three or more anomalies in the following body parts: Vertebrae, Anorectal, Cardiac, Tracheal, Esophageal, Renal and Limbs. Other common associated syndromes include trisomy 21, trisomy 18 and CHARGE syndrome (1). CHARGE syndrome is characterized by Coloboma of the eye, Heart defects, Atresia of the choana, Growth restriction and Ear abnormalities (13).

1.1.7 Treatment of Esophageal Atresia

EA is treated surgically. Preoperative treatment of EA patients includes continuous suction of the upper esophageal pouch and, if necessary, respiratory support. Surgical treatment is most often performed within the first days of life. The surgical principles are the closure of any fistula and connecting the esophageal ends by anastomosis. If the esophageal ends are located too far apart for primary anastomosis, the approach is different. There are several surgical options, such as; the Foker-process (14), colonal interposition (15), gastric pull up (16) or delayed primary anastomosis (17). The Foker-process is a surgical approach that induces growth of the esophageal ends with applying traction (18). Postoperative monitoring includes controlling for anastomotic leakage, bleeding, infections, and additional complications. During the first few days after surgery, the baby is sedated to allow the anastomosis to heal. Feeding during the first days or weeks is primarily through a nasogastric tube. Once the baby is able to swallow milk/formula, breastfeeding may be attempted. To ensure proper growth weight is measured daily.

1.1.8 Prognosis and sequalae

Treatment of EA has improved since 1941 when Mr. R. H. Franklin wat the first to attempt surgical repair of EA. The same year Dr. Swenson adopted Franklins technique and the first patient survived surgery (9). Today, EA is characterized by morbidity, not by mortality. Mortality has fallen from 100% to 5-10% (2, 19). However, patients born with EA are likely to have long-term complaints with comorbidities and sequalae. These include feeding difficulties, esophageal strictures, dysphagia, esophageal stenosis, gastro-esophageal reflux disease (GERD), tracheomalacia and Barrett's esophagus (metaplasia), musculoskeletal disorders and neurodevelopmental disabilities (20, 21).

1.2 Prenatal diagnosis

1.2.1 What is prenatal diagnosis?

A prenatal diagnosis is a diagnosis made before birth. Ultrasound is the most used examination modality. Routine ultrasound examinations are most often performed by general practitioners, gynaecologists, or midwives, whereas most of the supplementary examinations are performed at a fetal medical centres. Supplementary examinations are amniocentesis, MRI, NIPT (non-invasive prenatal test) and chorionic villus sampling.

1.2.2 Ultrasound examination

Since 1981, pregnant women in Norway have been offered a routine ultrasound examination during gestational weeks 17-19 (22). The aim of the ultrasound examination is to:

1) Estimate gestational age of the fetus and the due date
2) Determine the number of fetuses
3) Localization of the placenta
4) An overview of the fetal anatomy and development

Some pregnancies have higher risks of malformations and complications and are therefore offered an earlier ultrasound examination during gestational week 11-14 (23).

1.2.3 Benefits of a prenatal diagnosis

There may be several benefits of a prenatal diagnosis.

1) **Parental information and counselling**

- If we can diagnose a malformation prenatally, we can inform the parents and they can prepare for the expected baby. The parents may acquire further information regarding the syndrome or malformation (24).

- It may be helpful to know if they are expecting a child that might need more resources than they are able to give. If they consider termination of pregnancy as an alternative, they can make an informed decision at this point (24).

2) **Scheduled delivery**

- With rare malformations that might require urgent surgery shortly after birth, the number of highly specialized hospitals are limited. If the malformation is known before birth, we can plan the mode and place of the delivery with the best possible outcome.

3) **Early treatment**

Following prenatal diagnosis, the treatment can start shortly after birth. As an example, a suction catheter may be inserted shortly after birth to prevent aspiration of saliva. We may perform relevant pre-operative diagnostic tests to confirm the prenatal diagnosis, and surgical treatment might be carried out sooner.

1.2.4 Concerns with prenatal diagnosis

Several studies on parental reactions in the perinatal period also describe concerns with prenatal diagnosis.

1) **Parental distress**

A study from Sweden showed that parents of children with a prenatal diagnosis of a congenital heart defect had less coherence compared to parents of children with a postnatal diagnosis (25). Another study suggests that the parents receiving a prenatal diagnosis often

experience two emotional traumas: the first upon receiving a prenatal diagnosis and the second at delivery (26). Mothers of newborns with congenital malformations have a higher stress level when receiving a prenatal diagnosis compared to receiving a postnatal diagnosis (27).

2) **Uncertainty related to a prenatal diagnosis**

The malformation and the potential presence of additional malformations may be difficult to assess following a prenatal diagnosis. A prenatal finding might be unspecific and not related to a specific diagnosis or type of malformation. This makes it difficult for parents to prepare mentally. The parents often prepare for worst-case scenarios and become concerned and nervous (26). A prenatal diagnosis generates not only uncertainties including logistics around delivery and early treatment, but also regarding the severity of the malformation and/or presence of additional malformations (28). The uncertainty can even generate more stress than generated by the severity of the malformation (29).

1.3 Prenatal signs of Esophageal Atresia

1.3.1 Prenatal findings on ultrasound and MRI

The prenatal findings on ultrasound and MRI are similar. There are three signs that derive from having a discontinuous esophagus.

1) **Polyhydramnios:**

Polyhydramnios means that there is excess amniotic fluid surrounding the fetus. The prevalence of hydramnios is estimated to be between 0.7% and 2% of all pregnancies. Polyhydramnios is idiopathic in approximately 50% of the cases, and the severity of polyhydramnios correlates with the likelihood of an underlying cause. The underlying causes may be congenital malformations (10-40%), maternal diabetes mellitus (10-40%), multiple pregnancies (10%) and fetal anaemia (1%). Among the congenital malformations, gastrointestinal tract malformations and esophageal atresia are the most frequent (30-32).

The Amniotic Fluid Index (AFI) or the depth of the largest pocket of amniotic fluid defines polyhydramnios. AFI is calculated by adding the deepest pockets in the four quadrants of the uterus of the mother. The definition of polyhydramnios is an AFI > 25 cm. The deepest

amniotic fluid pocket should be >8 cm (33). The reason most pregnancies with EA present prenatally with polyhydramnios is because the fetus is unable to swallow amniotic fluid. The amniotic fluid level in the uterus increases, eventually causing polyhydramnios.

2) **Small or absent stomach**:

If the fetus is unable to swallow amniotic fluid, the fetal stomach is small or absent on ultrasound and MRI. If there is a lower TEF, making an indirect connection between the mouth and the stomach, some fluid may be detected in the stomach. These signs may be difficult to interpret, especially when presented as a small stomach rather than an absent (empty) stomach.

3) **The pouch sign**: The upper esophageal pouch in the EA fetus is poorly drained. The fetus will attempt to swallow amniotic fluid. The accumulation of fluids results in a esophageal pouch, which is visible on ultrasound and MRI. The pouch sign was introduced by Zemlyn in 1981 (34).

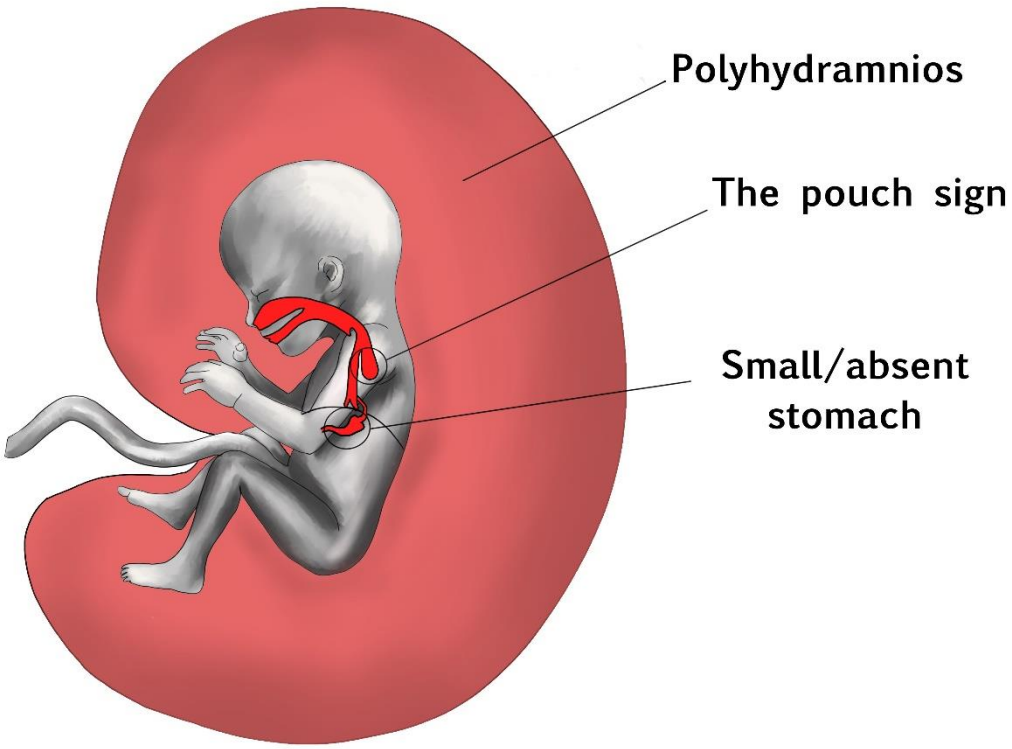

Figure 1.3.1: *An illustration of the common prenatal signs of EA*

Prenatal diagnosis of EA involves uncertainties and must be confirmed after birth. Because of the uncertainty of a prenatal diagnosis of EA, we refer to a prenatal suspicion, especially when informing the parents. Compared to many other malformations, EA is difficult to diagnose prenatally, because the signs are mostly indirect and ambiguous. Thus, the prenatal signs only raise a suspicion of EA and do not make a definitive diagnosis. The European Union defines a prenatal diagnosis on their webpage: “Prenatal Diagnosis is defined as a diagnosis suspected/made in a live fetus at any gestation” (35). In this study, we will use the term prenatal diagnosis as defined by the European Union.

1.3.2 Biochemical markers and amniocentesis

Biochemical markers can be detected in a maternal blood sample and in the amniotic fluid. Maternal serum alpha-fetoprotein (MSAFP) is a marker used to prenatally diagnose congenital malformations, such as neural tube defects, omphalocele and gastroschisis (36). Other biochemical markers, such as gamma-glutamyl transpeptidase (GGTP), L-leucine-aminopeptidase (AMP) and intestinal alkaline phosphatase (iALP) are digestive enzymes found in the amniotic fluid (37). These digestive enzymes are produced at different levels in

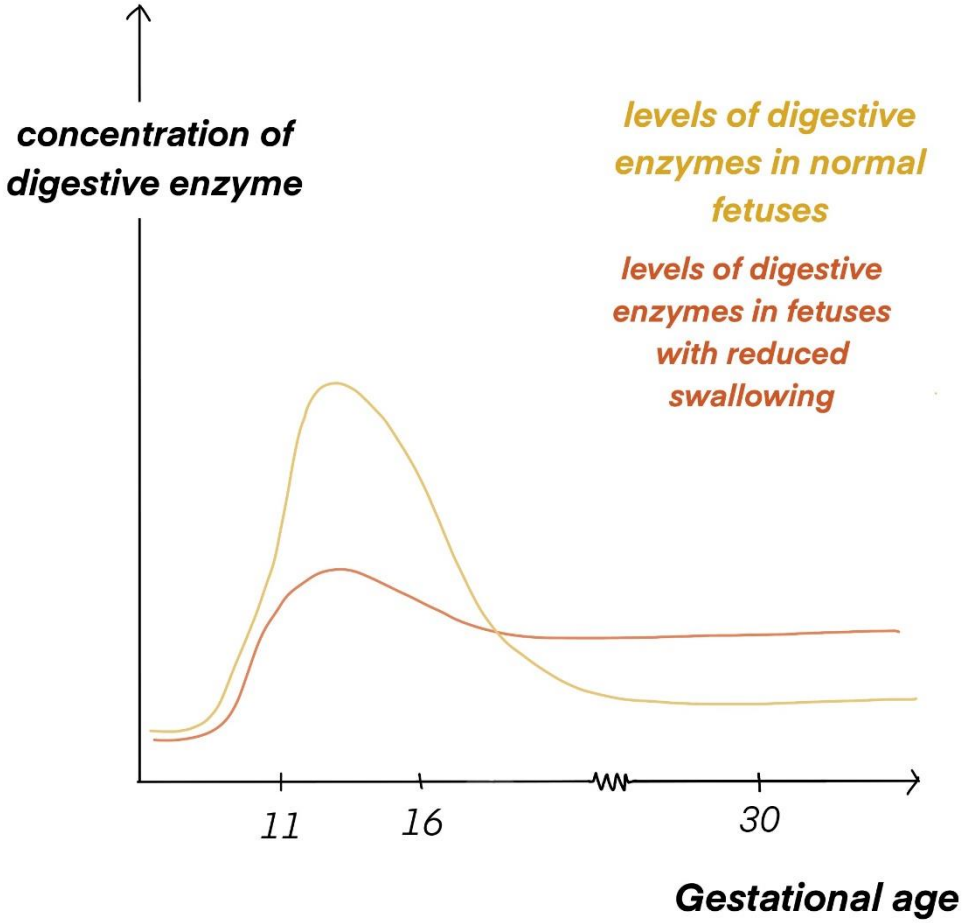

Figure 1.3.2 The concentration of digestive enzymes throughout pregnancy in normal fetuses and fetuses with reduced swallowing (33). From Gestational week 11, digestive enzymes are excreted per anus into the amniotic fluid and swallowed per os. The excretion and swallowing reach a plateau. Between GA-week 16 and 18, the anal sphincter is formed. The anal sphincter reduces the excretion but not the swallowing, and thus reduces the concentration of digestive enzymes in the amniotic fluid and stores it in the bowel as part of the meconium.

In fetuses with reduced swallowing, such as fetuses with EA, the plateau is reached at a much lower concentration due to reduced fluid and enzymes in the GI-tractus because of the reduced swallowing. During the formation of the anal sphincter, the fetus is still not swallowing and thus the digestive enzymes accumulate in the amniotic fluid.

the gastrointestinal tract and can therefore be of diagnostic value in the prenatal diagnosis of gastrointestinal malformations. To understand how these biochemical markers are useful, we need to know how and when they end up in the amniotic fluid. At gestational week 11-12 the fetus starts to swallow amniotic fluid. Fluids and digestive enzymes accumulate inside the gastrointestinal tract of the fetus and are then excreted per anus into the amniotic fluid. The swallowing and excretion plateau at week 16-18 when the anal sphincter forms and the excretion decreases. (37)

1.4 Aim of the study

We aimed to:

- search for and collect all the available **literature** on prenatal diagnosis of EA.
- describe the **trends** in publication in this field over the period 1990-2020.
- find the **prevalence** of prenatal diagnosis in EA patients.
- identify the different examination **modalities** and accuracy of prenatal diagnosis of EA.
- identify the **prenatal signs** of EA described in the literature.

2 Method

2.1 Search strategy

We performed a literature search and wrote a systematic review that encapsulates the current knowledge on prenatal diagnosis of EA. The literature search was performed with predefined search terms in PubMed (1990-2020) and Cochrane. We did not search in Medline since it is included in PubMed. We completed the search on January 16th, 2020. We searched in PubMed and the following search terms were used: (Esophageal atresia*[mesh terms] OR Tracheoesophageal fistula*[mesh terms]) AND (Prenatal diagnosis*[mesh terms]), (Esophageal atresia*[Title/abstract] OR EA*[Title/abstract] OR Oesophageal atresia*[Title/abstract] OR OA*[Title/abstract] OR Tracheoesophageal fistula*[Title/abstract] OR Tracheoesophageal fistulae*[Title/abstract] OR TEF*[Title/abstract]) AND (Prenatal diagnosis*[Title/abstract] OR Antenatal diagnosis*[Title/abstract] OR Prenatal suspicion*[Title/abstract]). The search in PubMed provided 371 papers. The same search strategy was used for Cochrane by replacing "[Title/abstract]" with ": ti, ab, kw". The search in Cochrane yielded 5 papers.

T. Arntzen screened the results of the searches using inclusion and exclusion criteria.

2.2 Assessment of study eligibility

The papers were evaluated by their title, abstract and full text. All the papers from the search were included and screened. The assessment of study eligibility was based on the inclusion and exclusion criteria. The papers were stored in an endnote library.

We included all the papers from the literature search. The inclusion criteria were applied in the search engine. The inclusion criteria were as following:

1) The paper is in English

2) The paper is published after 1990

After the inclusion of the papers from PubMed and Cochrane we applied the exclusion criteria.

The assessment of study eligibility was performed in three stages:

1) Categorical exclusion of duplicates

   The exclusion of duplicates was performed using the “Search for duplicates” function in Endnote.

2) Title and abstract screening was performed with these exclusion criteria
   - The paper is not available in full text
   - The paper is in the wrong language
     - The paper is not in English
   - The content is wrong
     - The paper is not about esophageal atresia
   - The study design is incorrect
     - The study includes less than 5 patients with EA
     - The study is not an original study
3) Full text screening was performed with this criterion
   - The paper does not focus on prenatal diagnosis of esophageal atresia

2.3 Analysis

The papers were read and analysed based on our aims. The trends in the papers were evaluated based on the modalities used, the country and journal that published the paper, and the distribution across the time period. The prevalence of prenatal diagnosis of EA was referred directly or calculated from the numbers if provided. The modalities and signs, if included, were analysed based on the sensitivity, specificity, positive predictive value, and negative predictive value.

3 Results

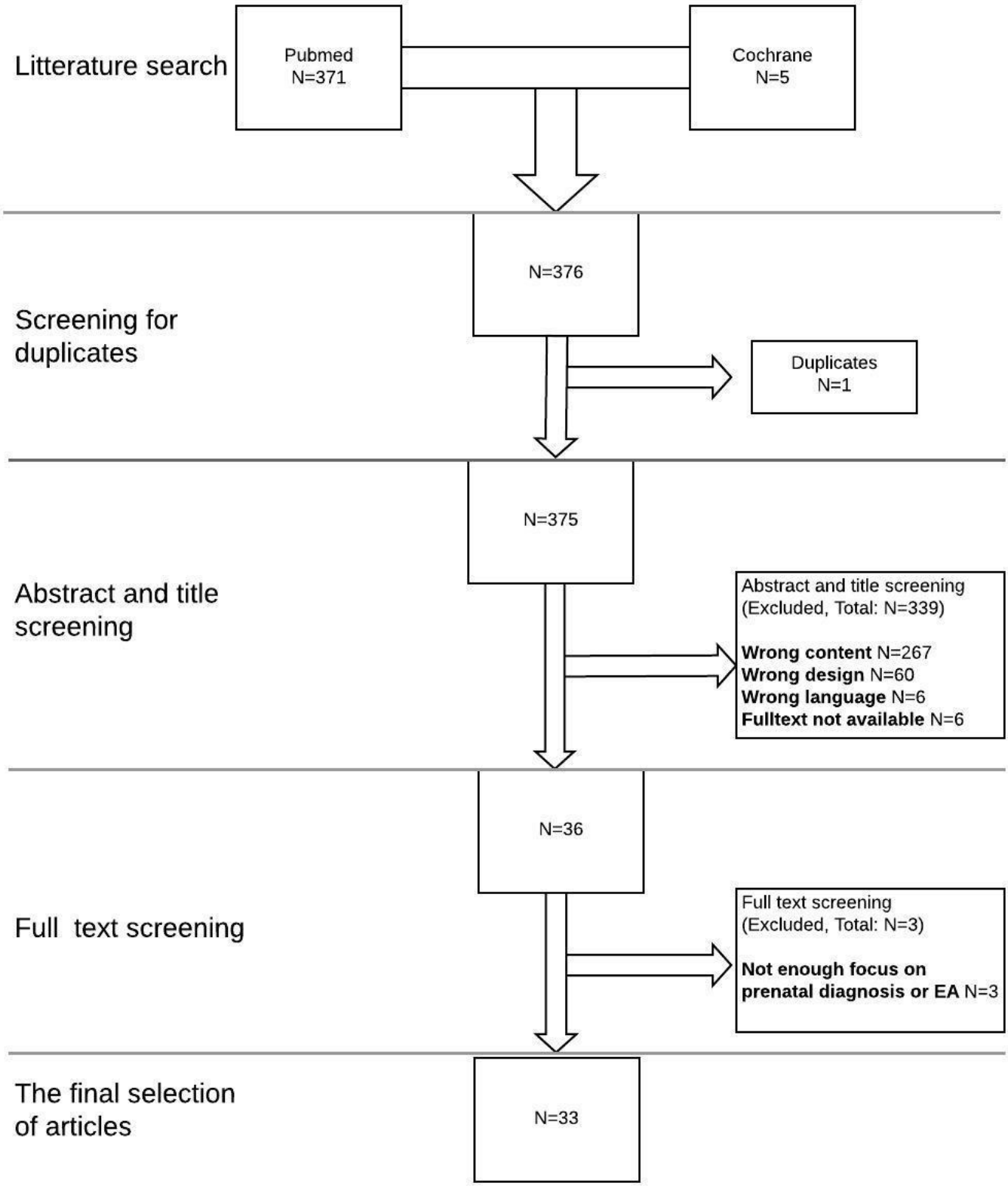

Figure 3.1.1: *Flowchart of the assessment of study eligibility.*

3.1 The search results

The literature search resulted in 376 hits, mainly from PubMed. Figure 3.1.1 is a flowchart of the process of screening the papers and eliminating those that were ineligible. The original search was wide and included not only the name of the malformation, esophageal atresia, esophageal atresia and tracheoesophageal fistula, but also the abbreviations such as EA.

During the full text screening, three papers had too little focus on EA and prenatal diagnosis and were therefore excluded. In the final selection, all papers were from PubMed and none from Cochrane. After the assessment of study eligibility, 33 papers remained.

We collected information regarding country of origin, journal and year of publication, as well as the diagnostic modalities used. This information provided us with information regarding the trends in our selection of papers.

We analyzed the prevalence of prenatal diagnosis. Some of the papers did not present prevalence and were excluded from the analysis on prevalence.

To analyze the modalities, we selected the papers that focused on either ultrasound, MRI, biochemical analysis, or a combination of two or more. The sensitivity, specificity, negative predictive value and positive predictive value were included when noted in the papers.

When calculated in the given paper, the prenatal signs were analyzed with positive and negative predictive values, sensitivity and specificity. We evaluated how the different prenatal signs performed alone and in combination with other signs and other examination modalities.

3.2 Trends in the literature on prenatal diagnosis of Esophageal atresia research

We found 33 papers that fulfilled our criteria, and the majority were published during the last decade. Most of the papers originated from France. The dominating diagnostic modality was ultrasound. The selection of papers represents eleven countries. Table 3.2.1 presents year of publication, country of origin, number of patients, number of EA cases and diagnostic modality.

Most of the papers [18/33(55%)] were published in journals of pediatric surgery. The remaining papers were published in journals of prenatal diagnostics (21%), gynaecology and obstetrics (12%), radiology (3%), and in a specific journal concerning the esophagus (3%), epidemiology (3%) and general medicine (3%).

3.2.1 Trends in diagnostic examinations

Trends in the entire selection

Research on prenatal diagnosis of esophageal atresia was more frequent in the last decade compared to the previous two decades combined. The years with the most frequent publications were 2014 and 2015 with five publications each. Figure 3.2.1 shows the distribution of the publications during the period.

Trends in studies using ultrasound in diagnosing Esophageal atresia

Thirty of the 33 papers used ultrasound in diagnosing EA. Ultrasound was used throughout the entire period, with the first publication in 1994 and the last in 2018. On average, one paper was published each year, and almost 1/3 of the papers were published in 2014 and 2015.

Trends in studies using MRI and biochemistry in diagnosing Esophageal atresia

Ten studies investigate MRI as a diagnostic modality for EA. The first study was published in 2001 and the last in 2018. On average there was one paper published every second year from 2001 to 2018. Two out of three papers were published in 2014 and 2015.

Eleven papers describe using amniocentesis as a diagnostic method. These papers were published from 1994 to 2018. On average, one paper was published every two or three years.

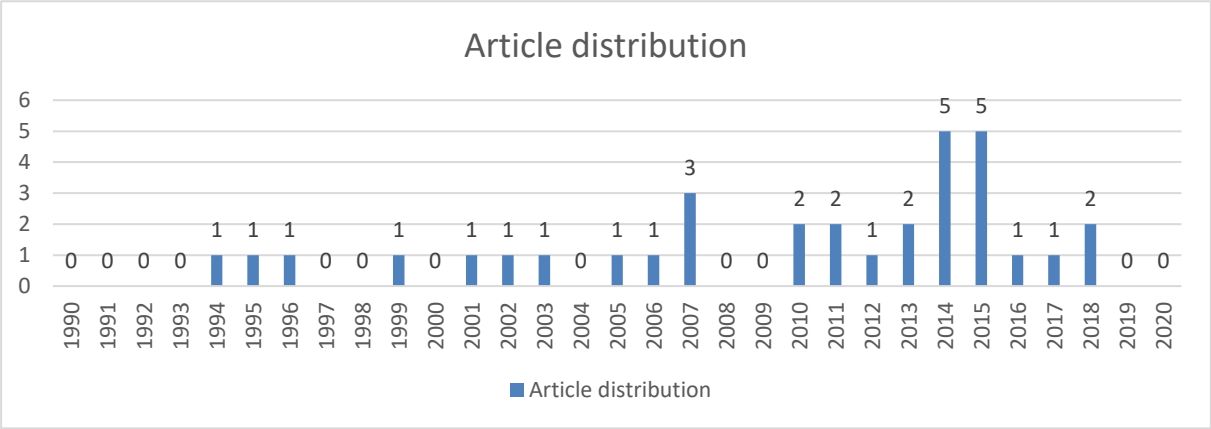

Figure 3.2.1 Distribution of the papers throughout the time period.

Author	Year	Country	Total number of patients	Cases of EA	Diagnostic examination
Borsellino (38)	2006	Italy	157	8	US
Bradshaw (12)	2016	UK	58	58	US
Brantberg (39)	2007	Norway	48	48	US
Chodirker (40)	1994	Canada	16	16	US, Amniocentesis
Choudhry (41)	2007	UK	62	62	US
Czerkiewicz (42)	2011	France	44	44	US, amniocentesis
de Jong (6)	2010	Netherlands	79	79	US
Ethun (43)	2014	USA	33	15	US, MRI
Fallon (44)	2014	USA	91	91	US, MRI
Garabedian (45)	2018	France	1118	1118	US, MRI, Amniocentesis
Garabedian (46)	2014	France	15	10	US, MRI, Amniocentesis
Garabedian (47)	2015	France	469	469	US, MRI, Amniocentesis
Garne (48)	2007	UK	1480	376	US, MRI, Amniocentesis
Hochart (49)	2015	France	18	11	MRI
Khorshid (50)	2003	Saudi Arabia	78	78	US
Kunisaki (51)	2014	USA	22	11	US
Lal (52)	2017	USA	396	396	US
Langer (53)	2001	Canada	10	5	US, MRI
Leoncini (54)	2015	Australia	260	260	US
Muller (37)	2013	France	252	31	US, Amniocentesis
Pedersen (55)	2012	Eurocat	1222	1222	US
Pini prato (56)	2015	Italy	146	146	US
Quarello (57)	2011	France	7	7	US, Amniocentesis
Séguier-Lipszyc (58)	2005	France	10	10	US
Juhee Seo (59)	2010	South Korea	81	81	Amniocentesis
Sfeir (60)	2013	France	307	307	Not described
Shulman (61)	2002	Israel	25	6	US
Spaggiari (11)	2015	France	122	122	US, MRI, Amniocentesis
Sparey (62)	1999	UK	176	158	US
Stol (63)	1996	France	129	33	US
Stringer (64)	1995	USA	87	15	US
Takahashi (65)	2014	Japan	74	74	US, Amniocentesis
Tracy (66)	2018	USA	75	39	US, MRI

Table 3.2.1: *The selection of papers included in our literature study.*

3.3 Prevalence of prenatal diagnosis of Esophageal Atresia

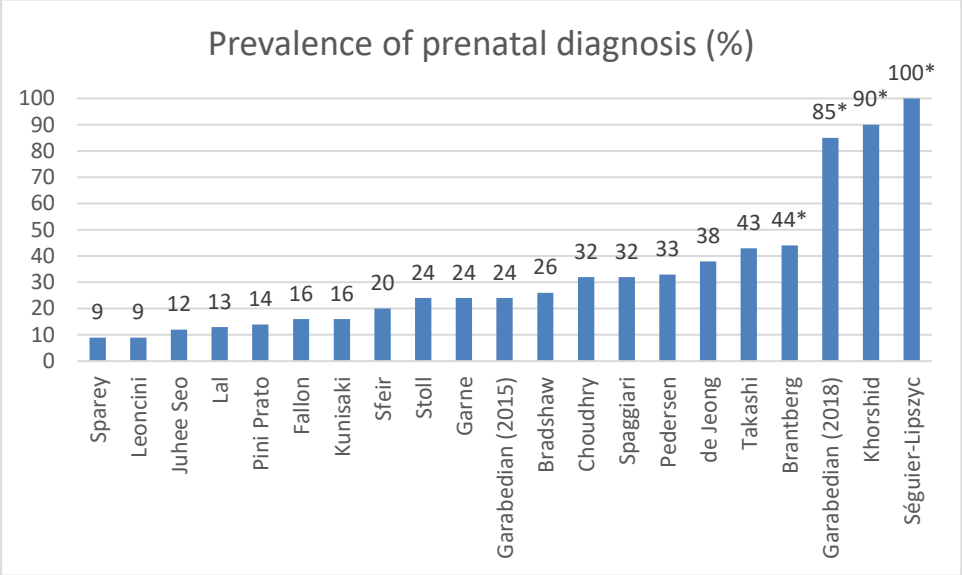

Figure 3.3.1 Prevalence of prenatal diagnosis of EA

The range of prenatal diagnosis was 9-100%. Most papers report prenatal diagnosis with a prevalence of 10-40%. Three papers present a prevalence above 60%. These are marked with an "*".

*Séguier-Lipszyc et al. only included patients with long gap esophageal atresia (LGEA) (58). *Garabedian et al. only included patients with EA type A (45). Type A EA is more likely to be diagnosed prenatally since there is no fistula connecting the mouth and the stomach. Therefore, the stomach is even more likely to be small or absent (45, 58).

*Khorshid et al. found that 90% of their patients had a prenatal diagnosis. They collected data on all patients born between 1994-1998 at Maternity and Children's Hospital, Riyadh in Saudi Arabia and found 78 patients with EA. A prenatal diagnosis including polyhydramnios and/or small/absent stomach was detected by ultrasound. Ninety % of the patients had polyhydramnios and received a prenatal diagnosis. The authors do not mention if there were any false positives in their population (50).

In Figure 3.3.1 the papers are arranged by publication year. We excluded the three outliers from the figure (45, 50, 58).

The prevalence of prenatal diagnosis did not improve during the 30 years, as presented in figure 3.3.2.

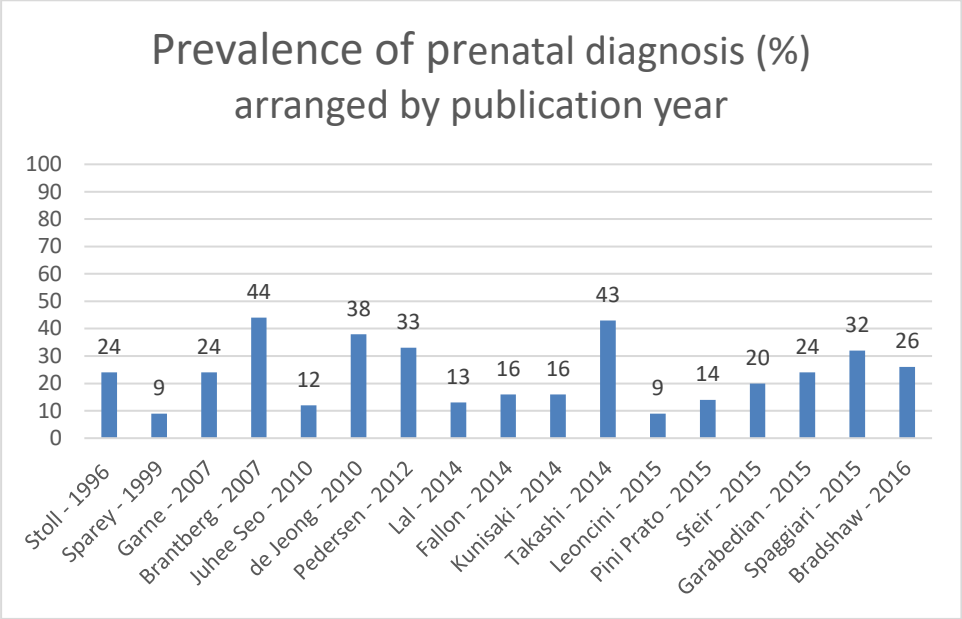

Figure 3.3.2 Prevalence of prenatal diagnosis arranged by year.

3.4 Methods to diagnose Esophageal Atresia

From our literature search 33 papers met our study criteria. The modalities used to diagnose EA prenatally were ultrasound, MRI and biochemical analysis. In most of the papers, the course of the diagnostic investigations described for the individual pregnant woman is the same.

It starts with phase one, which is a prenatal ultrasound examination with an abnormal finding. As a result, the patient is referred to a specialist. The specialist may be located at a general hospital or a tertiary center. In phase two, the specialist examines the patient. The specialist makes a prenatal diagnosis or not depending on the ultrasound examination and possibly other examinations according to their hospitals guidelines. Phase three takes place after birth when the baby is examined, and a postnatal diagnosis is made. The phases are illustrated in figure 3.4.1 using Bradshaw et al. as an example. Bradshaw et al. included all patients that had a routine ultrasound examination during pregnancy in a tertiary centre during a 10-year period, regardless of prenatal suspicion of EA (12).

Ultrasound is inevitable in the prenatal diagnosis of EA. After abnormal findings on ultrasound, the patient may be examined more thoroughly with additional ultrasound examinations, MRI or biochemical analyses. In the next chapters, we will discuss the prenatal findings leading to a diagnosis.

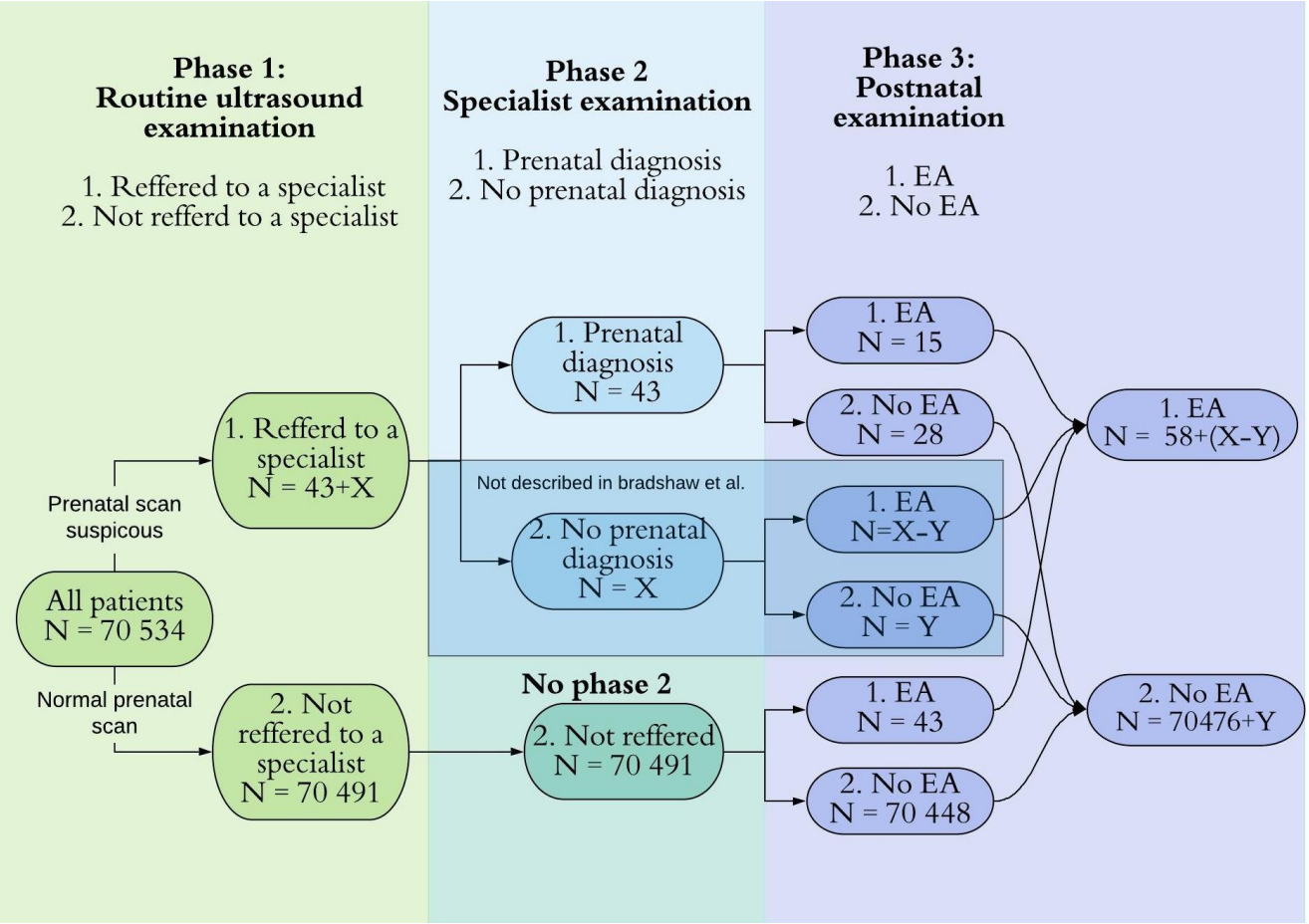

Figure 3.4.1: The three phases in diagnosing esophageal atresia with the numbers from Bradshaw et al. used as an example (12).

3.5 Prenatal findings on ultrasound

Seven papers describe ultrasound examinations with a predefined set of findings leading to a prenatal diagnosis. Six of the papers describe patients referred to a fetal diagnostic centre. The six studies are displayed in table 3.5.1.

The papers varied in what diagnostic markers that led to a referral to a specialist and what ultrasonic findings regarded as necessary for a prenatal EA diagnosis.

Choudhry et al. did a retrospective study in the hospital records of John Radcliffe Hospital in Oxford, UK. During a 10-year period, 62 patients had an EA diagnosis either prenatally or postnatally. Their patients either belonged to the region of John Radcliffe hospital, and thus had a pre-booked place of birth at the hospital, or they were referred for a second opinion from another district general hospital. Choudhry et al. concluded that a small or absent stomach bubble is too sparse to make a precise prenatal diagnosis of EA (41).

"*The main finding in this study is the difficulty in identifying TOF/OA based on subjectively small/absent stomach bubble at prenatal scanning. Of the 32 cases of TOF/OA, 10 (31%) were suspected prenatally because of a small/absent stomach bubble on scan"(41).*

Gestational age (GA) for ultrasound diagnosis is reported from week 18 until birth. The median GA is shown in table 3.5.1.

	PPV	**NPV**	**Se**	**Sp**	**Ultrasound Findings that led to referral to a specialist**	**Findings leading to a prenatal diagnosis**	**Median GA (weeks)**
Borsellino (38)	73%	-	-	-	Small or absent stomach AND polyhydramnios	Confirmation of the same findings on two or more ultrasound examinations	
Bradshaw (12)	35%	99%	26%	99%	Small or absent stomach AND/OR polyhydramnios	Confirmation of the same findings on two or more ultrasound examinations	20-24
Choudhry (41)	25%	-	31%	-	Small or absent stomach	Small or absent stomach	24 (19-31)
Garabedian (46)	67%	-	-	-	Small or absent stomach AND/OR polyhydramnios AND MRI findings	Confirmation of the findings on ultrasound and/or findings in MRI or amniocentesis	28+5 (24-36)
Langer (53)	50%	-	-	-	Small or absent stomach	Confirmation of the findings on ultrasound and/or findings in MRI	23 (17-33)
Stringer (64)	34%	-	42%	-	Small or absent stomach AND/OR polyhydramnios	Small or absent stomach AND polyhydramnios	27 (18-39)

Table 3.5.1: *Prenatal diagnosis using ultrasound, including GA (gestational age) at ultrasound scan. PPV (Positive predictive value), NPV (Negative predictive value), Se (sensitivity) and Sp (specificity).*

3.5.1 Polyhydramnios

In a series of 158 patients with polyhydramnios, the most frequent malformation causing polyhydramnios was esophageal atresia, seen in 6 patients (3.7%) (32).

Stringer et al. described a population of 87 patients with small or absent stomach. Twenty-one of the 87 fetuses had oligohydramnios, and none had a postnatal EA diagnosis. Oligohydramnios refers to a condition characterized by too little amniotic fluid. It is the opposite of polyhydramnios(64). Twenty-eight of the 87 had normal amniotic fluid volume and two of them had a postnatal EA diagnosis. The remaining 38 of the 87 had polyhydramnios. Ten of these 38 pregnancies had a postnatal EA diagnosis. Since all the patients had small or absent stomach, this study shows the importance of polyhydramnios(64).

Tracy et al. evaluate polyhydramnios as a sign for EA. The results are listed in table 3.5.2. Polyhydramnios by itself is considered too unspecific to diagnose EA. It is, however, present in more than 50% of the fetuses with EA and it is easy to detect on an ultrasound scan (11, 44, 67). In addition, other fetal anomalies may also present with polyhydramnios. Therefore, it is a sign that leads to further examinations (66).

Tracy (66)	**PPV**	**NPV**	**Se**	**Sp**	**Median GA**
Polyhydramnios	61%	56%	56%	61%	-
Small or absent stomach	59%	58%	67%	50%	-
The pouch sign	95%	75%	62%	97%	32 (25-38)
Polyhydramnios AND Small or absent stomach	70%	56%	41%	81%	-
Garabedian (46)					
The pouch sign	100%	46%	40%	100%	32 (24+4-33)

Table 3.5.2: *Ultrasonographic signs as reported by Tracy et al. and Garabeidan et al. This table lists the PPV (positive predictive value), NPV (negative predictive value), Se (sensitivity) and Sp (specificity) of the signs, and the median GA (gestational age) at detection of the sign.*

3.5.2 Small or absent stomach

Borsellino et al. focus especially on false positives. In the study, 11 patients had prenatally suspected EA, and three of them were false positives. In the study, the criteria for a prenatal diagnosis was a small or absent stomach bubble and polyhydramnios. *"All 3 FP had polyhydramnios and microgastria, and no FP occurred when diagnosis of EA was based on polyhydramnios and absent gastric bubble"* (38). When restricting the finding to absent stomach, rather than small or absent, the specificity improved, but the sensitivity decreased.

In Garabedian et al. small or absent stomach and polyhydramnios was the criterion to look for the pouch sign. This sign is quite specific for EA (46). In Langer et al., small or absent stomach led to suspicion and further examination with MRI (53). Several other studies also listed small or absent stomach as a criterion for suspicion and further examination, with or without polyhydramnios (41). Small or absent stomach is therefore, similarly to polyhydramnios, a sign that often leads to further examinations.

3.5.3 The pouch sign

The pouch sign is presumed to be specific for EA. In Garabedian et al., all cases with the pouch sign had EA. The specificity was 100%. However, the sensitivity was low, only 40%. The values are listed in table 3.5.2. They list possible reasons for the low sensitivity. The reason, they suggest, was the difficulty in identifying the sign due to fetal position, GA, and the fact that the fetus needed to swallow during the examination for the sign to become visualized. They suggest a longer examination period, 20-30 minutes (46).

Brantberg et al. also include the pouch sign in their study. They found a low sensitivity for the pouch sign in the patients that already had a prenatal diagnosis based on small/absent stomach and polyhydramnios. Only 9/21 (43%) with a prenatal diagnosis had the pouch sign (68).

Another paper that describes the pouch sign is by Solt et al. (69). Due to the fact that it contained no patients born with EA, it was excluded. In their study there was six patients with a prenatal diagnosis of EA, but all were structurally normal at birth. All six of the patients had the pouch sign at some time during the pregnancy, thereby questioning the specificity of the sign. The first visualization of the pouch sign was in gestational week 18-29.

3.5.4 A combination of ultrasound signs

Tracy et al. describe how a combination of signs affect the precision. When polyhydramnios and small or absent stomach was combined, they had a higher specificity and positive predictive value, but lower sensitivity. The negative predictive value is similar. The values are listed in table 3.5.2 (66).

Borsellino et al. required the presence of both polyhydramnios and small or absent stomach. Despite requiring both signs present, they still reported false positives. Three out of 11 (27%) were false positives.

3.6 Prenatal findings on MRI and biochemical tests

3.6.1 MRI diagnosis

The papers discussed in this paragraph used MRI to gain a better visualization of the esophagus and the stomach. The MRI diagnosis is based on the following signs: the pouch sign or incomplete visualization of the esophagus or a small or absent stomach.

Langer et al. published a paper in 2001. They suggested that MRI can be useful in diagnosing EA, because the method is helpful in prenatally diagnosing other congenital malformations. They utilized the MRI to visualize the esophagus and the stomach. They examined 10 fetuses with MRI between gestational weeks 23 to 34. To have an MRI examination, they had to be referred due to small or absent stomach. The evaluation of the MRI findings are listed in table 3.6.1 (53).

Ethun et al. used MRI and ultrasound to diagnose EA. They looked for two signs on the MRI, “the pouch sign” and “incomplete visualisation of the esophagus”. Twenty-seven patients had a prenatal diagnosis of EA, and 15 were postnatally confirmed to have EA. Twelve out of fifteen patients with confirmed EA were examined with MRI. Among these 12 patients, ten had the pouch sign, and six had an incomplete visualization of the esophagus. They concluded that the pouch sign is a more sensitive and specific sign for EA than an incomplete visualization of the esophagus (43).

Garabedian et al. discussed that the pouch sign is easier to visualize on MRI than ultrasound. "*The location of the midline sagittal plane may be easier in MRI; indeed, it avoids the difficulties caused by fetal position or mother echogenicity*" (46).

Both Garabedian et al. and Ethun et al. discussed that MRI could be useful not only to improve prenatal diagnosis of EA, but also to look for other malformations. This is relevant for EA, which is associated with other anomalies.

MRI diagnosis	PPV	NPV	Se	Sp	GA
Ethun (43)	75%	60%	50%	82%	Mean: 30,2+/- 4,5*
Garabedian (46)	100%	71%	80%	100%	32+4 (30+2-36)**
Langer (53)	83%	100%	100%	80%	31 (23-34)**

Table 3.6.1: *Prenatal diagnosis using MRI, listing GA (gestational age) at MRI scan. PPV (Positive predictive value), NPV (Negative predictive value), Se (sensitivity) and Sp (specificity). *Mean GA. **Median GA and range.*

3.6.2 Specific MRI signs

There have been several attempts to find a new and better sign to diagnose EA. Hochart et al. introduced "the bowing of the trachea" sign and compared it to the pouch sign and small or absent stomach on MRI. The pouch sign was more sensitive than the bowing of the trachea sign, as seen in table 3.6.2.

Specific MRI findings	PPV	NPV	Se	Sp	Median (range) GA
Hochart (49) The bowing of trachea sign	100%	50%	50%	100%	32+4 (30+4-36)
Hochart (49) Small or absent stomach	77%	80%	91%	57%	32+4 (30+4-36)
Hochart (49) The pouch sign	100%	78%	82%	100%	32+4 (30+4-36)
Tracy (66) Distended hypopharynx	71%	83%	86%	67%	27 (19-36)

Table 3.6.2: *Specific MRI signs. This table lists the PPV (positive predictive value), NPV (negative predictive value), Se (sensitivity) and Sp (specificity) of the signs, and the median GA (gestational age) at detection of the sign.*

Tracy et al. introduced "The distended hypopharynx" as a sign for EA. They categorized the different signs of EA as primary signs and secondary signs. The primary signs were the pouch sign and the distended hypopharynx. The distended hypopharynx was more sensitive than the pouch sign but less specific (66).

3.6.3 Biochemical signs

Chodirker et al. published a paper in 1994 about "MSAFP levels and oesophageal atresia". The aim of their study was to find the relationship between maternal blood samples for alpha-fetoprotein (MSAFP) levels and EA. They found the levels to be higher than expected in fetuses with EA. Fetuses with EA were 4-5 times more likely to have elevated MSAFP levels than the rest of the population (40).

In 2011, Czerkiewcz et al. suggested an EA index (42). They found that fetuses with EA had high levels of GGTP and AFP, as well as total protein, but normal or low AMP levels in amniotic fluid. Based on this pattern, they created the EA index and used it to compare EA fetuses to normal fetuses with and without polyhydramnios. The EA index is AFP multiplied with GGTP with a cut-off value of 3.0. In Czerkiewcz et al., the EA-index in combination with one sign on ultrasound provided high sensitivity and specificity for EA, as seen in table 3.4.3. The median gestational age at amniocentesis was 32 weeks (23.3-38.5) (42).

Amniocentesis	**Diagnostic criteria**	**PPV**	**NPV**	**Se**	**Sp**	**Median (range) GA (weeks)**
Chodirker(40)	**MSAFP levels above 2.5**	-	-	19%	-	16,5 (14.5-33)
Czerkiewicz(42)	EA-index and one or more ultrasonic findings	-	-	98%	100%	32 (23.3-38.5)
Garabedian(46)	EA index in patients referred to specialist	82%	75%	90%	60%	32 (25-35+5)
Spaggiari(11)	EA-index in fetuses with a prenatal diagnosis	-	-	88,2%	-	-

Table 3.6.3: Prenatal diagnosis using biochemical analyses, listing the GA (gestational age) and PPV (positive predictive value), NPV (negative predictive value), Se (sensitivity) and Sp (specificity).

In 2013, Muller et al. explained why the digestive enzymes in Czerkiewicz et al´s paper showed the specific pattern. Since AMP and GGTP are both produced and secreted below the esophagus, they should both be elevated in the amniotic fluid in the third trimester. Muller et al found that the reason is that GGTP has a longer half-life than AMP, as seen in figure 3.6.1 (37).

In Garabedian et al. from 2014, they used the EA index created by Czerkiewicz et al. (46). Garabedian et al. found the EA-index to be less specific in their population, as seen in table 3.6.3. They explain that gestational age at amniocentesis could influence the results, but they both reported a median gestational age at amniocentesis to be 32 weeks.

Spaggiari et al. also used the EA-index by Czerkiewicz et al. They only included patients with a prenatal diagnosis of EA and therefore only tested the sensitivity. They discussed the previous results from Czerkiewicz and Garabedian and concluded that the sensitivity was high in all three studies (42, 46). However, in Spaggiari et al., amniocentesis was performed in patients with polyhydramnios only, and approximately 50% of their patients had polyhydramnios (11). Therefore, it is only relevant for half of the EA patients.

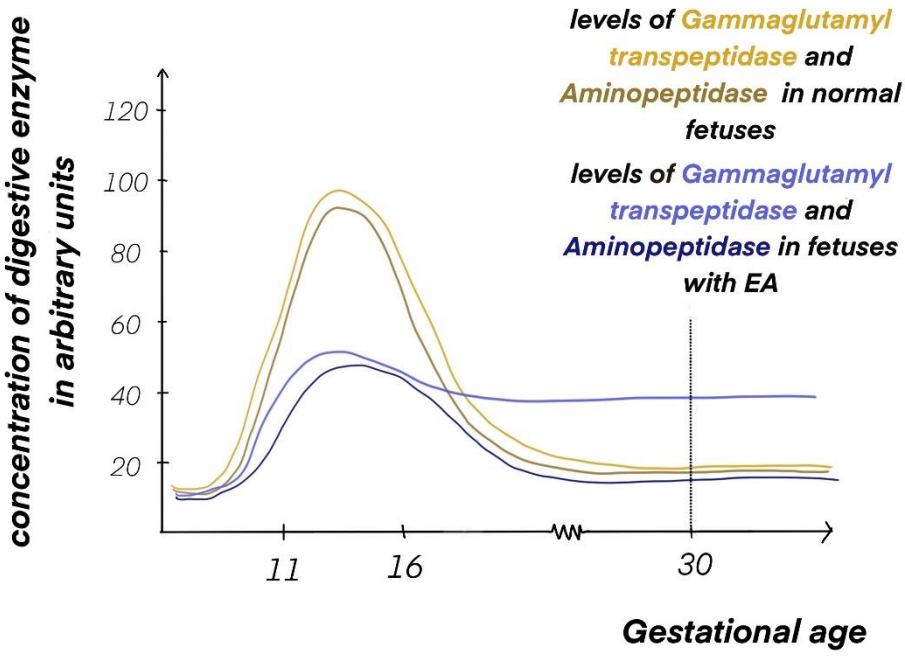

Figure 3.6.1: The theoretical model by Muller et al. The normal fetus (Yellow) compared to the fetus with EA (Blue) based on the levels of AMP (darker hue) and AMP (lighter hue). The values are expressed as the concentration of digestive enzymes as a function of gestational age. In both categories of fetuses, the anal membrane opens at approximatly gestational week 12. Digestive enzymes are leaked into the amniotic fluid. The amount of enzyme released is lower in the fetuses with EA due to reduced swallowing. The normal fetus continues to swallow amniotic fluid, including digestive enzymes. The anal sphincter develops from gestational week 16-20 and the digestive enzymes are either swallowed and stored in the meconium or not swallowed and left in the amniotic fluid. Since AMP has a shorter half-life than GGTP, the AMP levels decrease over time. At gestational week 30 the AMP levels are much lower than the GGTP levels.

3.7 A combination of signs and modalities

Tracy et al. evaluated a combination of signs using ultrasound and MRI. They distinguished between primary and secondary signs. Primary signs included the pouch sign and a distended hypopharynx. The secondary signs were abnormal stomach and/or polyhydramnios. The primary signs, the pouch sign AND/OR the distended hypopharynx, were found to be statistically significant in detecting EA. The combination of secondary signs, abnormal stomach AND polyhydramnios, were also found to be statistically significant. They also concluded that the secondary signs were better at detecting pure EA than EA with fistula (66).

Garabedian et al. performed both MRI, amniocentesis, and ultrasound examination of the pouch sign. They suggested a model with two out of three signs being positive, but the model was not statistically significant when compared to MRI signs alone. They conclude: "*In case of ultrasound suspicion of EA (with or without visualization of the pouch sign), an MRI at 30-32 weeks using fast imaging employing steady-state acquisition should be proposed. Biochemical amniotic fluid may be helpful and should be evaluated in a larger study" (46).*

Combination of signs and modalities	**PPV**	**NPV**	**Se**	**Sp**
Primary signs (66)	78%	76%	70%	82%
Secondary signs (66)	70%	56%	41%	81%
2/3 signs present (46)	100%	71%	80%	100%

Table 3.7.1: *Prenatal diagnosis using a combination of signs and modalities.*

Primary signs are direct signs such as the pouch sign and the distended hypopharynx. Secondary are indirect signs such as polyhydramnios and small or absent stomach.

"2/3 signs present" indicate at least 2/3 signs present as either the pouch sign on ultrasound, MRI diagnosis or amniocentesis analysed using the EA index.

PPV (positive predictive value), NPV (negative predictive value), Se (sensitivity) and Sp (specificity).

3.8 Conclusion

Prenatal diagnosis of esophageal atresia is challenging. Two thirds of the publications were published within the last decade. A prenatal diagnosis of esophageal atresia occurs in a minority of the cases (10-40%) and is usually suspected only because of the presence of indirect or direct signs on targeted ultrasonography. MRI and biochemical evaluation of the amniotic fluid marginally improve the detection of esophageal atresia. Maternal serum samples were not useful in diagnosing EA with a sensitivity of 19% only. Despite a meticulous prenatal diagnostic work-up, details of the malformations and the associated comorbidities can only be diagnosed postnatally.

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