



Early Identification of Autism Using Cry Analysis: A Systematic Review and Meta-analysis of Retrospective and Prospective Studies

Sandra Pusil¹ · Ana Laguna¹ · Brenda Chino^{2,3} · Jonathan Adrián Zegarra^{2,4} · Silvia Orlandi^{5,6}

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Abstract

Cry analysis is emerging as a promising tool for early autism identification. Acoustic features such as fundamental frequency (F0), cry duration, and phonation have shown potential as early vocal biomarkers. This systematic review and meta-analysis aimed to evaluate the diagnostic value of cry characteristics and the role of Machine Learning (ML) in improving autism screening. A comprehensive search of relevant databases was conducted to identify studies examining acoustic cry features in infants with an elevated likelihood of autism. Inclusion criteria focused on retrospective and prospective studies with clear cry feature extraction methods. A meta-analysis was performed to synthesize findings, particularly focusing on differences in F0, and assessing the role of ML-based cry analysis. The review identified eleven studies with consistent acoustic markers, including F0, phonation, duration, amplitude, and voice quality, as reliable indicators of neurodevelopmental differences associated with autism. ML approaches significantly improved screening precision by capturing non-linear patterns in cry data. The meta-analysis of six studies revealed a trend toward higher F0 in autistic infants, although the pooled effect size was not statistically significant. Methodological heterogeneity and small sample sizes were notable limitations across studies. Cry analysis holds promise as a non-invasive, accessible tool for early autism screening, with ML integration enhancing its diagnostic potential. However, the findings emphasize the need for large-scale, longitudinal studies with standardized methodologies to validate its utility and ensure its applicability across diverse populations. Addressing these gaps could establish cry analysis as a cornerstone of early autism identification.

Keywords Autism spectrum disorder · Early identification · Cry analysis · Infant cry · Acoustic analysis

Introduction

Autism Spectrum Disorder (ASD) is a neurodevelopmental condition affecting approximately 1 in 36 children in the United States (CDC, 2022; Mughal et al., 2024). It is characterized by challenges in social communication, behavior, and interaction, along with restricted and repetitive behaviors (RRBs) and atypical sensory processing, which vary widely in severity. While the etiology of ASD is linked to a combination of genetic and environmental factors (Chaste & Leboyer, 2012), no single definitive cause has been identified. Early signs of autism can emerge as early as two months of age (Okoye et al., 2023; Salgado-Cacho et al., 2021). However, diagnosis often occurs much later, relying on the criteria outlined in the *Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-5)* (Wiggins et al., 2019), alongside gold-standard tools like the Autism Diagnostic Observation Schedule (ADOS) (Lebersfeld et al., 2021) and the Autism Diagnostic Interview-Revised (ADI-R) (Lim

✉ Sandra Pusil
sandra.pusil@zoundream.com

¹ Zoundream AG, Novartis Campus - SIP Basel Area AG, Lichtstrasse 35, 4056 Basel, Switzerland

² Achucarro Basque Center for Neuroscience, Leioa, Spain

³ Department of Psychology, Faculty of Health Sciences, University of Deusto, Bilbao, Bizkaia, Spain

⁴ Universidad Señor de Sipán, Chiclayo, Perú

⁵ Department of Electrical, Electronic, and Information Engineering “Guglielmo Marconi” – DEI, Alma Mater Studiorum University of Bologna, Bologna, Italy

⁶ IRCCS Istituto Delle Scienze Neurologiche Di Bologna, Bologna, Italy

et al., 2018). Screening tools such as the Modified Checklist for Autism in Toddlers, Revised (M-CHAT) (Guthrie et al., 2019) are also commonly used in clinical practice.

The diagnostic process can begin with developmental screenings during routine well-child visits, followed by comprehensive evaluations by multidisciplinary teams. However, delays in diagnosis are common, driven by subjective assessments and resource limitations, especially in underserved communities (Vu et al., 2023). While it is possible to reliably diagnose ASD by the age of two (Cor-sello et al., 2013), many children are not diagnosed until after age four (Baio et al., 2018). Such delays hinder timely access to early interventions, which are crucial for leveraging neuroplasticity and improving developmental outcomes. Without early support, families often face greater reliance on intensive therapies or pharmacological treatments, which may have limited long-term efficacy (DeFilippis & Wagner, 2016). Early intervention has proven to be a foundation in improving the quality of life and developmental trajectories for autistic children (Dawson, 2008).

The search for innovative early detection and screening methods remains critical to enhancing long-term clinical outcomes for autistic children. Emerging technologies, such as Artificial Intelligence (AI)-driven algorithms, hold promise in bridging gaps in current diagnostic practices by enabling earlier and more precise identification of autism (Song et al., 2019). Early identification can support timely interventions that promote developmental progress, enhance adaptive skills, and optimize learning opportunities during critical periods of neurodevelopment (Boyd et al., 2010; Nadel & Poss, 2007).

Recent studies have highlighted the potential of physiological signals, including eye-tracking (Ahmed et al., 2022; Kanhirakadavath & Chandran, 2022; Meng et al., 2023; Wen et al., 2022), electroencephalogram (EEG) in infants starting at 3 months (Gabard-Durnam et al., 2019), MRI changes associated with social deficits (Shen et al., 2022), abnormal connectivity patterns identified via functional near-infrared spectroscopy (fNIRS), and cry acoustics (Khozaei et al., 2020), in the early identification of autism. Furthermore, recent Machine Learning (ML) techniques have demonstrated high accuracy in identifying behavioral patterns associated with autism using acoustic and visual data, such as video (Kojovic et al., 2021) and retina image analysis (Kim et al., 2023)). Notably, several researchers have found differences in the cry acoustics of autistic infants younger than 12 months, suggesting these may function as reliable early indicators of autism (Esposito & Venuti, 2010b; Orlandi et al., 2012; Sheinkopf et al., 2012).

In this context, cry analysis is particularly compelling due to its accessibility, non-invasive nature, and its association with neurodevelopmental conditions (Esposito et al., 2017). Infant cries reflect the neurological and physiological state

of the infant, offering a unique opportunity to identify early markers of autism through differences in acoustic features. Previous studies have demonstrated that infants later diagnosed as autistic produce cries with distinct acoustic characteristics, including variations in fundamental frequency (F0, defined as the frequency of vocal fold vibrations measured in Hertz, typically ranges from 350 to 600 Hz in infants, and is commonly perceived as the "pitch"), duration, and variability (Esposito & Venuti, 2010a; Khozaei et al., 2020). These findings suggest that crying is a promising signal for exploring early vocal biomarkers of autism, warranting its inclusion as a central focus in this review.

While several reviews have examined early autism identification through physiological signals, including neuroimaging and behavioral cues (Shan et al., 2023), none have systematically reviewed or conducted a meta-analysis specifically on cry acoustics. For instance, prior reviews have focused on the general role of cry features in developmental conditions or pathologies (Jeyaraman et al., 2018; Ji et al., 2021), leaving the specific application of cry analysis in autism underexplored. Furthermore, no meta-analysis has been conducted to consolidate findings related to cry acoustics in infants who later receive an autism diagnosis.

Consequently, this review aims to highlight cry analysis as a promising, non-invasive, and accessible vocal biomarker with significant potential supporting early autism identification, advocating for its future integration into neonatology and pediatric practice. Specifically, this systematic review analyzes the specific role of infant cry analysis in the early detection of autism, focusing on two main research approaches. The first approach involves retrospective studies that compare the acoustic features of cries from autistic infants (*AutI*) to those from typically developing (TD) infants, using statistical and ML techniques. The second approach includes prospective studies that follow infants with an elevated likelihood (EL) and a decreased likelihood (DL) of autism longitudinally, analyzing their cries and diagnosing autism later in life, again employing statistical analysis and ML techniques for comparison. Moreover, a meta-analysis will be conducted as part of this systematic review to examine whether the F0 of infant cries differs between *AutI* or EL and TD or DL infants. The primary goal is to synthesize findings from multiple studies reporting F0 measurements to assess its viability as an early vocal biomarker for autism identification.

Methods

Study Design

This systematic review included all levels of research evidence (e.g., randomized controlled trials, observational

studies, longitudinal studies, cross-sectional studies, and quantitative research) and aimed to integrate best practice systematic review methodology, the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines (Moher et al., 2010).

Search Strategy

A comprehensive search strategy was employed to identify relevant studies on cry analysis in relation to early autism identification. The PICO (Participant, Intervention, Comparison, Outcome) framework was used to structure the search related to the research questions (Cooke et al., 2012). We systematically searched multiple electronic databases that cover key biomedical, psychological, and multidisciplinary research areas, including PubMed, Scopus, Web of Science, and PsycINFO, using keywords and subject headings related to cry analysis and autism. Other databases, such as EMBASE, were excluded due to significant overlap with PubMed and Scopus, ensuring comprehensive yet efficient study identification. The search terms included "autism spectrum," "cry," and "screening;" and the search was conducted on 09 September 2024. The keyword combinations used in the literature search are presented in Table 1. Electronic database searches were performed using the key terms related to "Population" (Newborns with high-risk or with a diagnosis of ASD), "Intervention" (early detection of ASD), and "Outcome" (acoustical cry analysis). For the outcome, we included only cry terms to capture all potentially relevant articles. The search strategy did not specify a comparison/control group. These terms were considered in the inclusion and exclusion criteria. After retrieving studies from the searches, duplicates were removed, and the paper titles, abstracts, and associated meta-data were compiled into a single table for further review. We also reviewed reference lists of relevant articles to identify additional studies.

Inclusion and Exclusion Criteria

All research within Oxford levels of evidence I–IV (Howick, 2011), including case studies and single-case experimental design studies reporting objective outcome measures were eligible for inclusion, if they met the following criteria: a)

Studies focusing on cry analysis; b) Studies conducted on children under 60 months of age (this age range encompass early autism identification and the period of significant neuroplasticity, aligning with the typical timeline of diagnosis and recognition by caregivers and educators); c) The evaluation process follows a uniform diagnostic protocol (including the last three versions of the *Diagnostic and Statistical Manual of Mental Disorders* (DSM) criteria (DSM-IV, DSM-IV-TR, DSM-V)); or diagnosis made by a specialist in neurology, psychiatry, and pediatrics; or based on an extensive, rigorous and detailed neuropsychological evaluation; d) Reported in full text; and e) English language manuscripts.

The exclusion criteria taken into account were as follows: a) Participants with other medical conditions (included neurological, neurodevelopmental, genetic, perinatal, metabolic, and chronic illnesses that could confound cry analysis); b) Information relative to statistical analysis, ML, or AI was not reported; c) Utterance analysis was based on other types of infant vocalizations such as speech or babbling, because they do not directly assess cry-related biomarkers.

From the systematic review, the final eligible studies included were assessed to evaluate crying as a potential vocal biomarker for autism. For inclusion in the meta-analysis, only studies that reported sufficient data on F0, such as the mean and standard deviation for both *Aut/EL* and *TD/DL* groups, were retained. The studies were included if they met the following criteria:

Reported quantitative measures of F0 (e.g., mean and standard deviation) for both *Aut/EL* and *TD/DL* groups. Included infants aged between 0 and 18 months (to capture a critical developmental period before a reliable diagnosis is made and to ensure sufficient data for robust statistical analysis while maintaining clinical relevance). Provided data sufficient to calculate Standardized Mean Differences (SMD) and standard errors. Studies that did not report F0 metrics or failed to provide data enabling the calculation of effect sizes were excluded. Gray literature and unpublished works were not eligible for inclusion (research and materials that are not formally published in traditional peer-reviewed journals, includ-

Table 1 PICO Search terms

Key concept	Search terms used
Population: autism spectrum disorder	"Autism spectrum" or "autism" or "autistic spectrum disorder" or "autistic disorder" or "child development" or "development disorder"
Population: newborn	"newborn" or "infant" or "toddler" or "neonate"
Intervention: early screening	"early screening" or "early detection" or "early diagnosis" or "screening" or "assess" or "test"
Outcome: cry	"cry" or "cries" or "crying" or "atypical cry"

ing reports, theses, government documents, white papers, and other non-commercial publications). Strictly qualitative research, book chapters, and review articles were excluded.

Screening Process

To ensure reliability, prevent duplication, and adhere to standard systematic review protocols, this review was registered in PROSPERO (publication code: CRD42024592154). Selected articles were imported into Rayyan (Ouzzani et al., 2016), used to facilitate the screening process through its collaborative and filtering functionalities. This was done following the revision process by two reviewers (BC and SP), as recommended in PRISMA guidelines (Moher et al., 2010), based on the established selection criteria. The manuscripts selected in the title and abstract review were thoroughly reviewed. The disagreements regarding the selection were resolved through discussion or a third reviewer (screening protocols and final selection are depicted in Fig. 1). The third reviewer (AL) arbitrated disagreements that could not be resolved by discussion. An almost perfect level of agreement was obtained for title, abstract screening,

and full-text screening (Cohen's kappa coefficient, $k = 0.983$, $SE\text{-}kappa = 0.017$, $CI: 0.949\text{--}1.00$).

Quality Assessment

The included studies' quality and risk of bias were assessed using the National Heart, Lung and Blood Institute (NHLBI) quality assessment tool for observational cohort and cross-sectional studies (NHLBI, 2021). The NHLBI tool is a widely recognized framework designed to systematically evaluate the internal validity and reliability of studies, focusing on factors such as study design, sample size, and measurement methods. Two different reviewers (JAZV and SP) independently applied the protocol as mentioned earlier. Discrepancies were solved through discussion or by a third reviewer (BC).

Data Extraction

Data were extracted using a standardized form to ensure consistency and accuracy. The information extracted included study characteristics such as Study Design, Audio Features studied, Statistical and ML Analysis, Findings, Sample size and Study limitations. Two reviewers (AL and

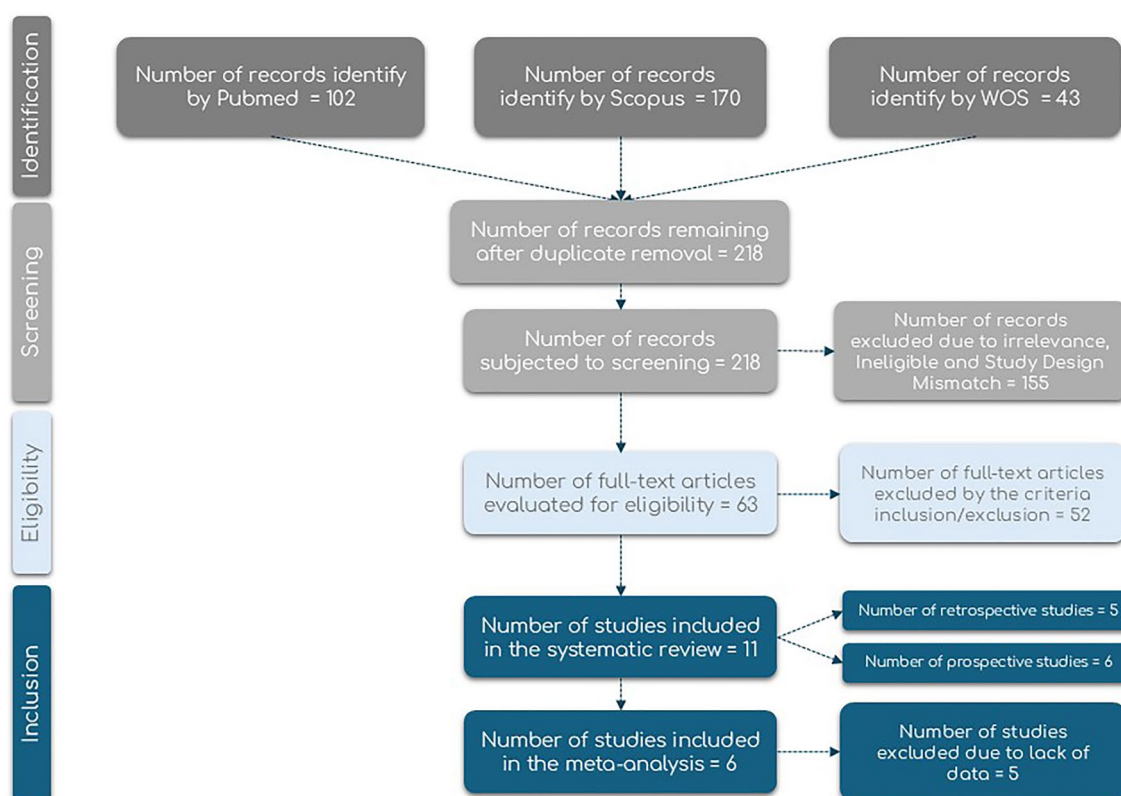


Fig. 1 Selection process overview. Flow chart of included and excluded articles through the screening process following the PRISMA presentation guidelines for the systematic review and for the meta-analysis

SP) performed this extraction process and discrepancies during data extraction were resolved by consensus between the two reviewers. The information is summarized in Table 2 and Table 3 according to the study type (retrospective vs. prospective studies). The Study limitations presented in Tables 2 and 3 reflect those identified by each author in their respective manuscripts. Most of these limitations are common challenges in this type of research, such as heterogeneity, sample size, and issues related to audio recordings.

Data for the meta-analysis were independently extracted by two reviewers (SP and AL) to guarantee accuracy and consistency following the full-text review conducted during the systematic review. Discrepancies during meta-analysis data extraction were resolved by consensus between the two reviewers. The extracted information included (Table 4):

The mean and standard deviation of F0 for *AutI*/EL and TD/DL groups.

Sample sizes for both *AutI*/EL and TD/DL groups.

Study-level characteristics, such as the infant age range (0–18 months), methods used to elicit crying, and the conditions under which recordings were made.

Statistical Analysis

The meta-analysis was conducted using Jamovi software (version 2.4.14) and its meta-analysis module (MAJOR) to assess the variability and reliability of findings across studies. SMD were calculated to compare F0 values between *AutI*/EL and TD/DL groups, enabling the integration of data from studies with different measurement scales. Positive SMD values indicated higher F0 in *AutI*/EL infants, while negative values indicated higher F0 in TD/DL infants. The random-effects model, following the DerSimonian and Laird method (DerSimonian & Laird, 2015), was used to account for variability in study designs, populations, and methodologies, providing a weighted average effect size while assuming true effect sizes varied across studies.

Heterogeneity among the included studies was evaluated using Cochran's Q test to determine statistical significance, the I^2 index to quantify the proportion of variation due to heterogeneity rather than chance (with values interpreted as low, moderate, or high at 25%, 50%, and 75%, respectively). The tau² statistic was used to estimate the variance in effect sizes. Model fit was assessed using Akaike Information Criterion (AIC) and Bayesian Information Criterion (BIC), where lower values indicated better fit and model parsimony.

The overall summary effect and 95% confidence intervals (CI) were calculated and visualized using forest plots, which displayed individual study effect sizes alongside the pooled effect size. Publication bias was assessed through

funnel plot analysis (Fig. 2) and further evaluated statistically using Egger's regression test and Begg's rank correlation test, with significant results indicating potential bias (Table 5). To ensure robustness, sensitivity analyses were performed by excluding studies with small sample sizes, outliers, or methodological differences, and any changes in effect size or heterogeneity were reported.

Results

This systematic review evaluated both retrospective and prospective studies to assess the role of cry analysis in early autism identification. The primary goal was to identify key acoustic features distinguishing the cries of *AutI* or EL infants from those of TD or DL infants and to evaluate the efficacy of statistical and ML techniques in these analyses. After a thorough literature search, 11 studies involving 736 participants were included in the systematic review (Fig. 1).

Quality Assessment

Risk of bias assessment revealed that 7 out of 11 studies were of fair quality, while the remaining four were rated as good (Esposito et al., 2014, p. 201; Esposito & Venuti, 2010b; Manigault et al., 2023; Unwin et al., 2017). None were classified as poor quality. Key strengths included appropriate sample size justification and power analysis, though limitations were noted in reporting methods for cry measurements and blinding of outcome assessors. Additionally, few studies provided validation for cry measurement protocols, and inconsistencies in the reporting of diagnostic and exposure data were common (Tables 2 and 3).

Literature Review on Early Autism Identification—Retrospective Studies

Table 2 summarizes five retrospective studies comparing *AutI* and TD infants. These studies analyzed cry features across a wide age range (5 months to 4 years). Statistical methods, including ANOVA, GLM, and correlation analysis, were employed in four studies (Esposito & Venuti, 2009, 2010b, 2010a; Moffitt et al., 2022), while one applied ML using Support Vector Machines (SVM) (Khozaei et al., 2020). Two studies include 20 infants (Esposito & Venuti, 2009, 2010a), one includes 28 children (Esposito & Venuti, 2010b), and the most recent studies report 62 (Khozaei et al., 2020) and 61 participants (Moffitt et al., 2022), respectively. ADOS-2 is the primary diagnostic tool used in 4 out of 5 studies. Notably, one study (Khozaei et al., 2020) has employed ML to distinguish *AutI* cries from TD ones. Specifically, SVM demonstrated classification accuracies exceeding 85% when distinguishing *AutI* from TD infants

Table 2 Characterization of cry in Autl versus TD infants

Research article	Groups, gender & recording age	Age and test for diagnosis of ASD	Audio features	Type of data analysis	Statistical analysis findings	Machine learning findings	Number of cry episodes (CEs) & cry types	Limitations	Quality assessment
(Esposito & Venuti, 2009)	Groups: Autl (n = 10) TD (n = 10) Gender: Autl (5 m/5f) TD (5 m/5f) Recording Age: 12 months	Age: 38.4 months: Diagnosis: - DSM-4 - ADOS-2 - Griffiths Mental Development Scale	- Duration - Waveform modulation - Dysphonation	Statistical analysis Test: -ANOVA - Tukey HSD post hoc tests	In Autl: - Longer hyperphonation cry periods (M = 0.14, SE = 0.04, $p \leq 0.05$) - Shorter aspiration/expiration (less waveform modulation and more dysphonation) (M = -0.24, SE = 0.05, $p \leq 0.05$) - Shorter pauses, more arousal (M = -0.08, SE = 0.05, $p \leq 0.05$) - No differences in means	N/A	CEs: 32 Cry types: Feeding, changing diaper and startled	- Sample size - Heterogeneity in Autl traits - Low quality of audio recording in amateur settings - Home videos	Fair
(Esposito & Venuti, 2010a)	Groups: Autl (n = 10) TD (n = 10) Gender: Autl (5 m/5f) TD (5 m/5f) Recording Age: 5 and 18 months	Age: 35.2 months: Diagnosis: - DSM-4 - ADOS-2 - Griffiths Mental Development Scale	- F0	Statistical analysis Test:—GLM - Tukey HSD post hoc tests	In Autl: - No change of F0 trajectory - Higher F0 at five and at 18 months (M = 40.35, SE = 15.82, $p \leq 0.05$)	N/A	CEs: 160 Cry types: Non-elicited cries during infant daily routine	- Sample size - Heterogeneity in Autl traits—Low quality of audio recording in amateur settings - Home videos	Good

Table 2 (continued)

Research article	Groups, gender & recording age	Age and test for diagnosis of ASD	Audio features	Type of data analysis	Statistical analysis findings	Machine learning findings	Number of cry episodes (CEs) & cry types	Limitations	Quality assessment
(Esposito & Venuti, 2010b)	Groups: Autl (n = 14) TD (n = 14) Gender: Autl (7 m/7f) TD (7 m/7f) Recording Age: 18 months	Age: 36.7 months Diagnosis: - DSM-4 - ADOS-2 - Griffiths Mental Development Scale	- F0	Statistical analysis Test: - GLM - ANOVA - Tukey HSD post hoc tests	In Autl: - Higher F0 (M = 65.44, SE = 19.85, P < 0.05) - No differences in the cry episodes length - No significant among feeding, changing diaper, pain cries	N/A	CEs: 140 Cry types: Feeding, changing diaper and pain	- Sample size - Heterogeneity of Autl traits- Low quality of audio recording in amateur settings - Home videos	Fair
(Khozaei et al., 2020)	Groups: Autl (n = 31) TD (n = 31) Gender: Autl (24 m/7f) TD (24 m/7f) Recording Age: 18–53 months	Age: - 35.6 months (Autl) - 30.8 months (TD) Diagnosis: - DSM-5 - GARS-2 questionnaire	- F0 - MFFC - other quality voice features	Machine Learning Analysis	N/A	RBF-SVM The sensitivity, specificity, and precision for boys were 85.71%, 100%, and 92.85%; 71.42%, 100%, and 85.71% for girls, respectively	CEs: 367 Cry types: uncomfortable, unwilling, sleepy, scared, thirsty, complaining	- Sample size for training (10 Autl / 10 TD) - Unbalanced dataset - Tested by gender	Fair
(Moffitt et al., 2022)	Groups: Autl (n = 61) Gender: Not reported Recording Age: 24–66 months	Age > 24 months Diagnosis: - DSM-5 - ADOS-2 - CSS - RRB - SA	- F0 - proportion of time crying	Statistical analysis Test: Correlation (Regression)	In Autl: - F0 predicted by ADOS-2, RRB and CSS ($\beta = 0.40$, t(63) = 3.83, p < 0.001)	N/A	CEs: Not reported Cry types: Not reported	- No TD group	Fair

Retrospective studies. Gender (m: male; f: female); F0: fundamental frequency – pitch, MFCC Mel frequency cepstral coefficients, GLM General linear model, RBF-SVM Radial basis function-support vector machine, ADOS-2: Autism diagnostic observation schedule – second edition, DSM IV and V Diagnostic and statistical manual of mental disorders, Fourth and Fifth Edition; RRBs Restricted and repetitive behaviors, CSS Calibrated severity scores, GARS-2 Gilliam autism rating scale, second edition

Table 3 Characterization of cry in elevated likelihood (EL) and decreased likelihood (DL) infants

Research article	Groups & age (months)	Age & test for diagnosis of ASD	Audio features	Type of data analysis	Statistical analysis findings	Machine learning findings	Number of cry episodes (CEs) & cry type	Limitations	Quality assessment
(Orlandi et al., 2012)	Groups: EL (n = 7) DL (n = 17) Gender: Not reported Recording Age: 10 days, 6 weeks and 12 weeks	Age: Not reported Diagnosis: Not reported	- Number of voiced and unvoiced segments - Cry Duration - F0 - F1 - F2 - Melody	Statistical Descriptive Analysis (mean, std)	In EL: - Lower F0 (10 days (EL: f0mean = 401.0, std = 108.2; TD: f0mean = 428.2, std = 145.9) 6 weeks (EL: f0mean = 417.7, std = 89.9; TD: f0mean = 447.9, std = 166.8) 12 weeks (EL: f0mean = 398.0, std = 93.3; TD: f0mean = 477.2, std = 188.7) - Higher F1 and F2 - Less and shorter cries - Similar melody patterns in hunger and boring cries In DL: - Large differences in the melody between hunger and boring cries	N/A	CEs: - 74 (10 days) - 75 (6 weeks) - 80 (12 weeks) Cry types: hunger and boring cry	- Sample size - Not reporting diagnosis information	Fair

Table 3 (continued)

Research article	Groups & age (months)	Age & test for diagnosis of ASD	Audio features	Type of data analysis	Statistical analysis findings	Machine learning findings	Number of cry episodes (CEs) & cry type	Limitations	Quality assessment
(Sheinkopf et al., 2012)	Groups: EL (n = 21) DL (n = 18) Gender: EL (6 m/15f) DL (8 m/10f) Recording Age: 6 months	Age: 36 months Diagnosis: - DSM-IV - ADOS - MSEL - MacArthur	- Phonation - Cry Utterance duration - Average amplitude (loudness) - Amplitude variation (range) - F0 - F0 variation - F1 and F2 - Hyperphonation	Statistical Analysis: t-test	In EL: - Higher F0 (t (10) = 2.82, P = 0.018) and F0 variation trend in pain related cries (t (10) = 2.14, P = 0.058) - Smaller amplitude trend (t (10) = 2.02, P = 0.070) - No differences in phonation	N/A	CEs: Not reported Cry types: pain and non-pain cries	- Small sample size - Difficulties distinguishing the causes of crying - Problems in F1 and F2 estimation - Manual analysis for CEs extraction	Fair
(Esposito et al., 2014)	Groups: EL (n = 13) DL (n = 14) Gender: EL (9 m/4f) DL (7 m/7f) Recording Age: 15 months	Age: 36 months Diagnosis: - DSM-IV-TR - ADOS	Time domain:— Cry duration Frequency domain: - F0 mean - F0 max - F0 variation (range)	Statistical Analysis: ANCOVA	In EL: - Higher F0 (F = 16.82, p < 0.01) and F0 max (F = 6.12, p < 0.05) - Shorter duration (F = 7.62, p < 0.05) - No significant differences in F0 variation	N/A	CEs: 159 Cry types: Cries elicited using the SSP, a standard infant attachment assessment	- Small sample size	Good

Table 3 (continued)

Research article	Groups & age (months)	Age & test for diagnosis of ASD	Audio features	Type of data analysis	Statistical analysis findings	Machine learning findings	Number of cry episodes (CEs) & cry type	Limitations	Quality assessment
(Unwin et al., 2017)	Groups: EL (n = 22) DL (n = 27) Gender: EL (18 m/4f) DL (12 m/15f) Recording Age: 12 months	Age: 12 months Diagnosis: - MSEL - ADEC Age: 24 months Diagnosis: - MSEL - ADOS-G	- F0 - Cry Duration - Amplitude - Formants (F1, F2)	Statistical Analysis (ANOVA, Pearson)	In EL: - Lower F0 trend (F = 0.01, p = 0.92) - A trend in shorter cry duration (F = 4.43, p = 0.04) correlating with more severe ADOS-G (p = 0.08) score and poorer performance on MSEL receptive language (p = 0.07)	N/A	CEs: 146 Cry types: Parent-reported cause of infant distress (e.g. hunger, fatigue, frustration)	- Sample size - Multiple comparison - Limited reporting on infant positioning	Good
(Santos et al., 2013)	Groups: EL (n = 23) DL (n = 20) Gender: EL (15 m/8f) DL (13 m/7f) Recording Age: 18 months	Age: 36 months Diagnosis: - ADOS-2 - ADI-R - Medical History	- F0 - Formants - Harmonics - Energy - CPP - hnr - Jitter - Shimmer - Voiced ratio	Machine Learning Analysis	N/A	PNN 97.7% SVM 79.1	CEs: 2187 Cry types: Not reported	- Sample size - No follow up information	Fair
(Manigault et al., 2023)	Groups: EL (n = 363) Gender: EL (202 m/161f) Recording Age: < 1 month	Age: 24 months Diagnosis: Bayley III M-CHAT CBCL	- Energy - F0 - Formants - Cry utterances - Signal quality	Machine Learning and Correlation Analysis	Estimates of models trained using acoustic cry characteristics were associated with clinical and developmental assessments at 2 years	Random Forest. 56 acoustics characteristics per utterance. Energy, F0, formants, and signal quality were the most variables used by the models	CEs: 14,701 Cry types: Elicited cries during routine caregiving in the NICU, such as diaper change	- Improve cry collection - Lacking specific intervention strategies - No accuracy reported	Good

Prospective studies. Gender (m: male; f: female); F0 fundamental frequency – pitch, F1 and F2 First and Second resonance frequencies, Hyperphonation: F0 > 1000 Hz; MFCC Mel frequency cepstral coefficients, CPP Cepstral peak prominence, ADOS-2 Autism diagnostic observation schedule—second edition, DSM IV and V Diagnostic and Statistical Manual of Mental Disorders, Fourth and Fifth Edition; M-CHAT Modified checklist for autism in toddlers, CBCL Child behavior checklist, Bayley III Bayley scales of infant and toddler development, Third Edition; ADI-R Autism diagnostic interview-revised, MSEL Mullen scales of early learning, ADEC Autism detection in early childhood, SSP Strange situation procedure

Table 4 Acoustic features—Fundamental frequency (F0) for meta-analysis selected studies

Research article	Aut/EL		TD/DL	
	n	F0 (mean $\pm$ std)	n	F0 (mean $\pm$ std)
(Esposito & Venuti, 2010b)	14	530.56 $\pm$ 91.30	14	465.12 $\pm$ 48.63
(Esposito & Venuti, 2010a)	10	535.33 $\pm$ 86.16	10	456.33 $\pm$ 59.74
(Orlandi et al., 2012)	7	398 $\pm$ 93.30	17	477.2 $\pm$ 188.70
(Sheinkopf et al., 2012)	21	504.87 $\pm$ 57.42	18	420.28 $\pm$ 40.32
(Esposito et al., 2014)	13	370.54 $\pm$ 30.48	14	329.96 $\pm$ 26.84
(Unwin et al., 2017)	22	420.91 $\pm$ 65.48	27	444.11 $\pm$ 88.65

based on cry features like F0, Mel Frequency Cepstral Coefficients (MFCC), and quality voice features.

Regarding statistical analysis, in general, the studies highlight key audio features, specifically the cry duration and F0. These retrospective studies consistently found that *Aut/*

exhibited more prolonged hyperphonation ($F0 > 1000$ Hz) (Esposito & Venuti, 2009) periods, fewer silence or pauses and shorter cry vocalizations (Esposito & Venuti, 2009), more dysphonation or irregular patterns in cry amplitude (Esposito & Venuti, 2009), and higher F0 (Esposito & Venuti, 2010a, 2010b), which are statistically significant markers. Additionally, F0 was also predicted by developmental assessments and diagnostic tools. Specifically, (Moffitt et al., 2022) suggest that both acoustic and phonological features of child vocalizations correlate with expert clinician evaluations of autism-related traits.

Literature Review on Elevated Likelihood of Autism—Prospective Studies

Six prospective studies examining EL and DL infants are detailed in Table 3. These six studies encompass a diverse age range, spanning from neonates to 18 months infants. Two of them used ML (SVM, Random Forest), and four applied Statistical Analysis (ANCOVA, ANOVA, t-test,

Fig. 2 Meta-analysis Summary Estimations. A. Forest plot for effect sizes for the included studies. B. Funnel plot for publication bias assessment

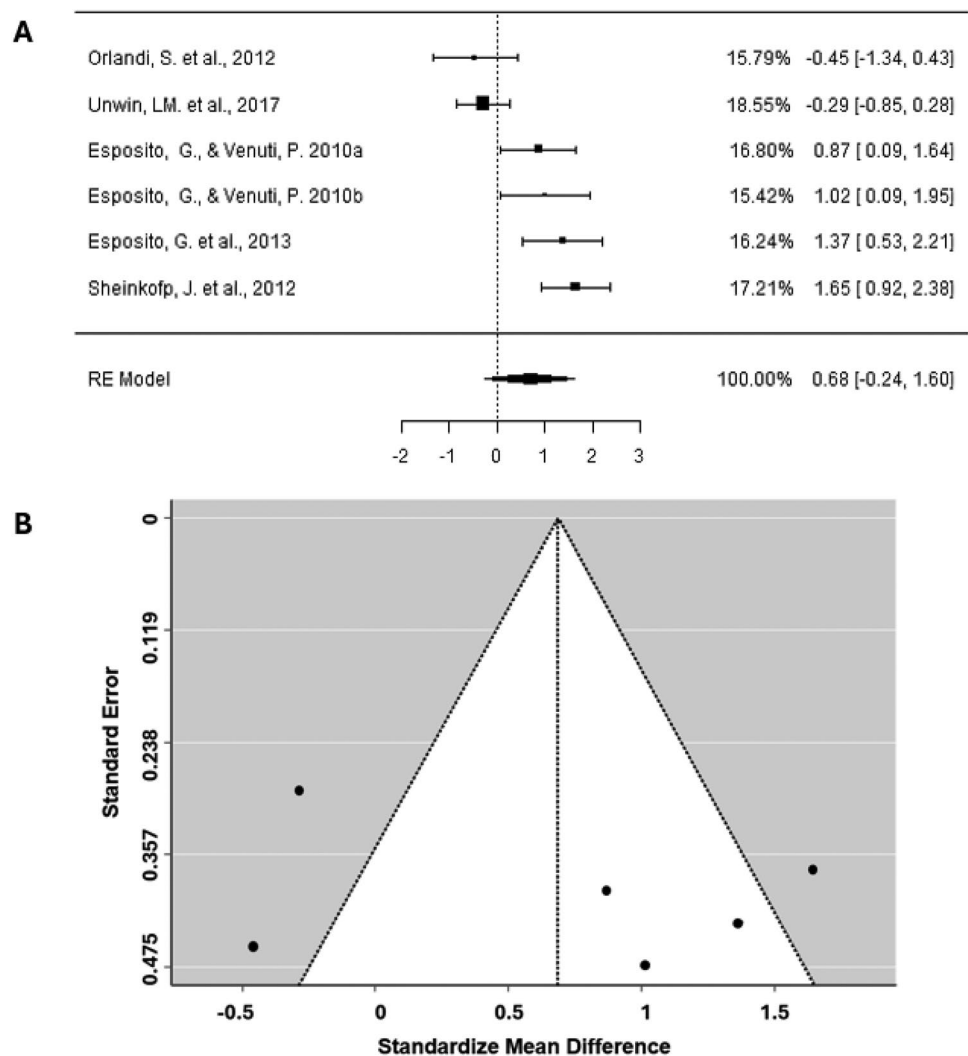


Table 5 Meta-analysis statistics for F0 across all different studies

Random-effects model (k = 6)							
	SMD	SE	Z	p	CI lower bound	CI upper bound	
Intercept	0.685	0.358	1.91	0.114	− 0.235	1.604	
Heterogeneity statistics							
Tau	Tau ²	I ²	H ²	R ²	df	Q	p
0.787	0.6196 (SE=0.4943)	80.2%	5.05		5.000	27.332	< .001
Model fit statistics and information criteria							
	log-likelihood		Deviance		AIC	BIC	AICc
Maximum-likelihood	− 7.238		14.534		18.477	18.060	22.477
Restricted maximum-likelihood	− 6.442		12.884		16.884	16.103	22.884
Publication bias assessment							
Test name				Value		p	
Fail-safe N				31.000		< .001	
Begg and Mazumdar rank vorrelation				0.067		1.000	
Egger's regression				0.509		0.638	
Trim and fill number of studies				0.000			

Tau² Estimator: Restricted Maximum-Likelihood. Knapp and Hartung (2003) adjustment used

Fail-safe N calculation using the rosenthal approach

Pearson). One study included the smallest sample size of 24 infants (Orlandi et al., 2012), while four studies enrolled between 40 and 50 infants (Esposito et al., 2014; Santos et al., 2013; Sheinkopf et al., 2012; Unwin et al., 2017). The largest sample size is reported by Manigault et al., 2023, who included 363 children. The ADOS was utilized in most studies, with the exception of Orlandi et al., 2012, which did not report any diagnostic assessment, and Manigault et al., 2023, which employed a broader developmental assessment approach combined with parental questionnaires (Bayley Scales of Infant and Toddler Development: Bayley-III; M-CHAT; and Child Behavior Checklist: CBCL, respectively).

These studies generally supported the findings of the retrospective studies, with EL infants showing distinct cry features, particularly increased F0 and shorter cry vocalizations. Prospective studies also identified more hyperphonation (F0 > 1000 Hz) on pain cries (Sheinkopf et al., 2012), higher resonance frequencies (F1 and F2) (Orlandi et al., 2012), smaller amplitude or energy (Sheinkopf et al., 2012), with no difference in phonation (Sheinkopf et al., 2012) and melodic patterns (Orlandi et al., 2012). Some studies (Esposito et al., 2014; Sheinkopf et al., 2012) show discrepancies in F0 variation. The same studies show F0 higher in EL infants while F0 is reported lower in (Orlandi et al., 2012) and (Unwin et al., 2017) although the studies included a small sample size.

One noteworthy AI model (Santos et al., 2013) achieved a 97.796% accuracy in effectively differentiating cries between EL and DL infants. Additionally, the study conducted by (Manigault et al., 2023), introduces a Random Forest algorithm capable of discerning EL cries in infants as young as 1 month. Although the accuracy of this model is not explicitly reported (Manigault et al., 2023), the model trained using acoustic cry characteristics was associated with clinical and developmental assessments at 2 years, being amplitude, F0, resonance frequencies, and signal quality as the most relevant variables.

Vocal Biomarkers for Differentiating Cries on Autism

Several consistent trends were observed across both retrospective (see Table 2) and prospective studies (see Table 3) for specific audio features that may help differentiate cries of *AutI*/EL from TD/DL infants:

1. F0, which refers to the basic rate of vibration of the vocal folds and is perceived as the pitch of a sound (Porter et al., 1988), was consistently a relevant feature in *AutI* and EL infants. Most of the studies show that F0 in cries is one of the most reliable early indicators of autism across multiple studies, as atypical pitch modulation can signal developmental differences. In this regard, within the prospective studies, (Orlandi et al., 2012)

and (Unwin et al., 2017) found a F0 decrease in the EL cohort compared to the DL one, while (Esposito et al., 2014) and (Sheinkopf et al., 2012) highlight an increase in F0.

2. Cry duration, where short cry vocalizations (cry during the expiratory phase of respiration) with shorter pauses (brief intervals of silence between voiced periods), (Laguna et al., 2023a, 2023b) less unvoiced segments and irregularities in cry intervals (inconsistent time gaps between cry bursts), and prolonged hyperphonation periods, has been identified as a key distinguishing feature of autism. This pattern is significant because more continuous crying and irregular timing have been linked to atypical regulatory processes in infants with an increased likelihood of neurodevelopmental differences (Esposito & Venuti, 2009; Esposito et al., 2014; Orlandi et al., 2011; Sheinkopf et al., 2012; Unwin et al., 2017).
3. Hyperphonation, defined as the presence of abnormally high-pitched ($F0 > 1000$ Hz) or intense vocalizations, especially in distress-related cries (Zeskind et al., 2011), emerged as a significant feature in both *AutI* and EL infants (Esposito & Venuti, 2009; Sheinkopf et al., 2012).
4. Jitter and shimmer, which are acoustic measures of voice stability (Teixeira et al., 2013), were frequently found to be higher in EL infants, correlating with later diagnoses of autism or behavioral issues (Santos et al., 2013). They are often associated with hoarseness and roughness of the voice (Teixeira & Fernandes, 2015). Jitter refers to the cycle-to-cycle variation in F0, while shimmer measures the cycle-to-cycle variation in amplitude.
5. Formants (F1, F2, etc.) are the prominent spectral peaks observed in the acoustic signal, created by resonance frequencies shaped by articulation in speech. They are derived from the resonance frequencies but represent the acoustic outcome specific to the configuration of the vocal tract during sound production (Vorperian & Kent, 2018). Several ML studies (Manigault et al., 2023; Santos et al., 2013; Unwin et al., 2017) highlight the formants' relevance as input features for the models. Additionally, even though (Sheinkopf et al., 2012) did not find differences among EL and DL infants, (Orlandi et al., 2011) did, promoting future research using these features.
6. Cry Melody, such as rising or falling patterns, often associated with different cry types characterization (Laguna et al., 2023a, 2023b), is defined as the contour of the F0 in time (Mampe et al., 2009; Manfredi et al., 2019; Wermke et al., 2002).
7. Amplitude, energy, or loudness (dB) produced by EL infants during pain-related cries presents a smaller range of amplitude than the DL infants. This finding is intriguing and suggests that vocal control or regulation of the

force of vocalizations may be affected in EL infants (Sheinkopf et al., 2012).

ML models, particularly SVM and Random Forest classifiers, consistently outperformed traditional statistical methods, achieving classification accuracies upwards of 90% across various studies (Khozaei et al., 2020; Manigault et al., 2023; Santos et al., 2013), validating their use in cry analysis supporting early autism identification.

Commonly used Diagnostic Tools for Autism

The gold standard for ASD diagnosis combines DSM-5 criteria with the ADOS, developmental assessments, and parental questionnaires (McMorris et al., 2013; Wiggins et al., 2019). The reviewed studies consistently relied on established tools, including the DSM-5 criteria, ADOS (assessment for social interaction and communication), developmental assessments such as the Mullen Scales of Early Learning (Akshoomoff, 2006), Griffiths Mental Development Scales (Pino et al., 2024), MacArthur Communicative Development Inventory (Charman et al., 2003), Child Behavior Checklist (CBCL) (Rescorla et al., 2019) or Bayley-III (evaluation for cognitive, motor, and developmental milestones) (Torras-Mañá et al., 2016). These tools were often used in combination with parental input and medical history to provide a comprehensive diagnostic framework. These methods align with international practices (Lordan et al., 2021), though some regions also utilize tools like Childhood Autism Rating Scale (CARS) (Rellini et al., 2004), Gilliam Autism Rating Scale (GARS) (Samadi & McConkey, 2014) or rely more on clinical judgment due to limited access to specialized instruments.

Most studies diagnosed autism between 18 and 54 months, with a peak around 36 months. While early signs were detected as young as 12 months, delays in formal diagnosis—often beyond age 4—highlight the need for earlier developmental screenings to address this gap.

Meta-analysis of F0 Values Between *AutI*/EL and TD/DL

A total of six studies were included in the analysis of F0 comparing *AutI*/EL and TD/DL infants (Table 4). The estimated overall SMD for F0 differences was 0.685 (standard error = 0.358), yielding a Z-value of 1.91. However, the result was not statistically significant at the 5% level ($P = 0.114$), as the CI: -0.24 to 1.60 included zero, indicating a lack of strong evidence for differences in F0 between EL and DL groups (see Table 5 for more details on statistics).

The studies demonstrated substantial heterogeneity, with a τ^2 value of 0.6196 ($SE = 0.4943$) and an I^2 of 80.2%,

reflecting high variability in effect sizes across studies. This variability likely stems from differences in study designs, sample sizes, methodologies, and the age ranges of infants studied (0 to 18 months). Model fit statistics, including an AIC of 18.477 and aBIC of 22.477, indicated a reasonable fit but highlighted the need for greater consistency in study designs (see Table 5).

Individual study findings varied, as illustrated in the forest plot (Fig. 2A). Studies by (Esposito & Venuti, 2010a, 2010b; Esposito et al., 2014) reported positive effect sizes with a CI that did not cross zero, indicating significant differences in F0 between EL and DL groups. In contrast, studies by (Orlandi et al., 2012) and (Unwin et al., 2017) found non-significant differences.

Despite the trend towards higher F0 in EL infants (combined SMD = 0.68), the results for this meta-analysis were not statistically significant, underscoring the need for further research. The symmetrical funnel plot (Fig. 2B) suggested no significant publication bias, and the distribution of study results appeared consistent, supporting the reliability of the findings despite high heterogeneity.

Discussion

The findings of this systematic review and meta-analysis highlight the significant potential of cry analysis as an accessible, automatic and non-invasive tool supporting early autism identification within the first months of life, often before observable behavioral traits emerge. Both retrospective and prospective studies demonstrate that features like F0, hyperphonation, amplitude, voice quality measures, and cry duration as reliable vocal biomarkers for distinguishing *Aut*/EL from TD/DL groups. Among these, hyperphonation is often associated with heightened arousal or stress, potentially reflecting sensory or emotional regulation differences common in autism (White et al., 2014; Zeskind et al., 2011), while voice quality irregularities in jitter and shimmer may signify underlying motor or neurological differences (Teixeira & Fernandes, 2015), underscoring the potential of infant cries as a biomarker for early screening. Moreover, the consistency of these findings across different ages and cry contexts (e.g., distress-related or spontaneous cries), also suggests their robustness as vocal markers of early autism-related neurodevelopmental markers. Additionally, the integration of ML models enhances the precision of cry analysis. ML approaches surpass traditional statistical methods by capturing complex, nonlinear relationships within acoustic data, enabling the differentiation of autism traits with high accuracy surpassing 90%. This emphasizes the critical role of advanced analytics in optimizing the predictive power of cry-based screening tools.

The Pivotal Role of Cry Analysis in Early Identification

The role of cry analysis in early autism identification is particularly important given the current challenges in diagnosing autism, which often depends on behavioral assessments that are not feasible, accessible and often challenging in very young infants. Early identification during infancy aligns with the critical developmental window when neuroplasticity is most pronounced, optimizing the effectiveness of early interventions (Dawson, 2008; Jacobson et al., 1998; Nadel & Poss, 2007; Towle et al., 2020). Evidence strongly supports the benefits of these interventions, ranging from cognitive and language improvements to better social outcomes, underscoring their transformative impact on children, families, and society (Boyd et al., 2010; Okoye et al., 2023; Pickles et al., 2016).

In brief, cry analysis, especially when combined with ML algorithms, represents a significant advancement in the field of early autism screening, potentially enabling clinicians to detect early signs or identify infants with an increased likelihood during routine pediatric check-ups, thereby facilitating timely early intervention programs.

Exploring F0 Differences in Cries as an Early Vocal Biomarker for Autism

The meta-analysis examined differences in the F0 of crying between *Aut*/EL and TD/DL infants, synthesizing data from six studies. The pooled effect size indicated a trend toward higher F0 in *Aut*/EL infants, but the result was not statistically significant. This finding aligns with hypotheses suggesting that atypical vocal characteristics could serve as early markers for autism (Sheinkopf et al., 2012). However, the lack of statistical significance and substantial heterogeneity among studies underscore the need for caution in drawing definitive conclusions. Variability in methodologies, including differences in F0 measurement techniques and the inclusion of infants at varying developmental stages, contributed to the inconsistency in findings. Notably, studies with significant results (Esposito & Venuti, 2010a, 2010b) highlight the importance of further research into these early acoustic markers. While no significant publication bias was detected, the substantial variability suggests a need for standardized methodologies in future research to improve comparability and robustness.

The Importance of Longitudinal Large-scale Studies and Population-Level Screening

While the existing research provides a strong foundation for the use of cry analysis in early autism screening, there is a growing need for longitudinal large-scale studies that

follow-up infants from the general population from birth through early childhood. Most prospective studies reviewed here focused on EL of autism, such as infants with siblings already diagnosed. While these studies provide valuable insights, they may not capture the full spectrum of autism traits or generalize to the broader population. Additionally, it is important to note that, despite the accuracy demonstrated by cry acoustic ML models in this review, they are not yet widely used in this field. This is likely due to the difficulty of gathering a sufficiently large, heterogeneous, and diverse sample size, as well as the low prevalence of relevant cases, which makes it challenging to train and validate robust ML models.

Therefore, future multicentric large-scale longitudinal studies following infants from various demographic, education, genders, races, ethnicities and socioeconomic backgrounds populations are essential to validate cry analysis as a universal screening automatic tool for early autism identification. Such studies would allow researchers to track the developmental trajectory of acoustic cry features over time, providing a more comprehensive understanding of how cry patterns evolve in both *AutI* and TD infants. Furthermore, identifying critical developmental windows where cry analysis is most predictive of later autism diagnoses could enhance early intervention strategies. The potential of cry analysis in early autism identification is supported by the success of similar technologies in clinical settings. For example, automated auditory screening tools like otoacoustic emission testing for newborn hearing loss (Akinpelu et al., 2014) have been seamlessly integrated into standard pediatric care due to their reliability and non-invasive nature. Furthermore, advanced diagnostic tools employing data from eye-tracking systems (Jones et al., 2023) and video-based behavioral analyses (Megerian et al., 2022), combined with ML algorithms, have demonstrated high accuracy and clinical utility in identifying early autism traits.

These approaches highlight how technology-driven, multimodal data-rich methodologies can be effectively incorporated into routine screenings, laying the foundation for cry analysis to become a practical tool in pediatric healthcare.

Limitations of the Systematic Review and Meta-analysis

The studies analyzed in this systematic review and meta-analysis had several limitations that should be considered when interpreting the findings. Small sample sizes were a common issue, reducing statistical power and generalizability, particularly in studies involving ML and acoustic cry analysis. Additionally, there was significant heterogeneity in participants' age, autism characteristics, and cry elicitation methods, complicating comparisons across studies. The variability in recording quality, often influenced by

non-standardized environments, further compromised data reliability. The lack of standardized methods for eliciting and recording cries also posed challenges, with some studies not differentiating between cries triggered by pain, or other needs or emotions. Moreover, missing or incomplete developmental follow-up information in several studies hindered the ability to draw long-term conclusions about the relationship between early acoustic markers and later autism diagnosis. Furthermore, limited demographic reporting restricted insights into the influence of socioeconomic, cultural, and biological factors. In fact, details related to gender, ethnicity, race, educational, socio-economic background, and co-occurring conditions that could influence cry patterns are usually not reported. Missing developmental follow-up data in many studies precluded definitive conclusions about the long-term predictive value of early acoustic markers. Finally, the meta-analysis itself was constrained by the substantial heterogeneity in study design, population characteristics, and cry measurement methods, emphasizing the need for standardization in future research. Specifically, the meta-analysis of F0 exhibited high heterogeneity, driven by differences in sample demographics, cry recording methods, and analytical approaches. The limited number of studies further constrained subgroup analyses and precision of effect size estimates. The non-significant pooled result highlights the potential for underpowered conclusions. These challenges emphasize the need for more robust, large-scale, and standardized studies to validate cry analysis as a screening tool for autism.

Conclusion

This systematic review and meta-analysis highlight the potential of cry analysis as a pivotal vocal biomarker supporting early autism identification. Specific acoustic features, such as F0, phonation, duration, amplitude, melody, resonance frequencies, and voice quality measures, consistently emerge as reliable indicators of early neurodevelopmental differences. ML models have further enhanced the diagnostic potential, outperforming traditional methods in accuracy and scalability.

The meta-analysis identifies F0 as a promising vocal biomarker, although high heterogeneity and a lack of statistical significance highlight the necessity for methodologically robust studies to confirm its clinical utility. Variability in methodologies and participant demographics underscores the need for standardized cry analysis protocols to establish reliable, generalizable findings.

In summary, cry analysis offers an objective, accessible, and non-invasive screening tool with the potential to support health assessments and developmental outcome prediction in early infancy. To realize this potential, future research

must focus on large-scale, multicentric, longitudinal studies encompassing diverse populations. These efforts should aim to refine ML models, establish standardized datasets, and delineate developmental trajectories of acoustic markers. By addressing these gaps, cry analysis could become a cornerstone of early autism detection, paving the way for timely interventions and significantly improving long-term outcomes for children and families worldwide.

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Declarations

Competing Interest The authors declare competing interests (Funding, Employment or Confidentiality interests) in relation to the work described herein. Sandra Pusil, Ana Laguna, are employed by Zoundream AG. Ana Laguna is also co-founder of the company and owns stock in Zoundream AG. Silvia Orlandi and Jonathan Adrian Zegarra-Valdivia receive compensation for the collaboration as members of the scientific advisory board of Zoundream AG. Brenda Chino declared no potential conflict of interest.

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