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AUTISM WITH AND WITHOUT REGRESSION:
A LONGITUDINAL ANALYSIS OF A CLINICAL COHORT

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Autism with and without regression:

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Autismo com e sem regressão: análise longitudinal duma coorte clínica

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Index

Abstract

Introduction: Autism spectrum disorder is a common neurodevelopmental disorder characterized by impairments in social communication and interaction, with behaviour patterns and desires limited and stereotyped. There is a gender ratio of 4:1 between males and females. Autism with regression is characterized as a trend in which children lose skills which they had previously acquired, namely those of social communication. Our study aims to analyse reported regression in both neurodevelopmental milestones and speech loss. It presents a significant correlation with adaptive function and developmental psychomotor quotients.**:**

Methods: Between 2010 and 2019, we carried out a retrospective analysis of a hospital population of 803 children and adolescents with an autism diagnosis, which was followed at the Pediátrico Hospital, Centro Hospitalar e Universitário de Coimbra.

We evaluated the global psychomotor development, adaptive functioning, and early neurodevelopmental milestones, such as the age of "walking," "first words" and "first phrases" acquisition and compared between children with and without regression.

Results: From the 803 patients, 15,2% had regression with 84% being male patients. As for the development quotient, the GMDS score was significantly lower in children with regression (68.8±20.1) comparing to those without it (74.6±21.0; p=.039). The global Adaptive function in the children with regression had also overall lower scores (mean±SD, 58.2±14.7) while no regression group had a slightly higher score, however still lower than 70 (mean±SD, 59.9±14.4). The VABS subdomains, communication dimension score (mean±SD, 58.5±16.6) and socialization dimension score (mean±SD, 63.7±12.2), were also significantly lower in patients with development regression (p=.034), when comparing with no regression group (mean±SD, 62.4±16.2 and 66.6±13.0 respectively). The exception is the subdomain Daily Living skills that had the same results in both groups [with regression (mean±SD, 60.6±17.9) and without regression (mean±SD, 60.8±15.9)]. In the neurodevelopment milestones, children with regression acquired the first words (13.5 months; p<.001) and phrases (38.0 months; p=.379) at earlier age compared with the non-regression group (24.0 months; p<.001 and 42.0 months; p=.379 respectively). No difference in the walking onset (14.0 months for regression and non-regression groups).

Discussion: The increase in diagnoses *per year* of regression is probably due to better knowledge of the disease and better diagnostic methods. In the regression group, on average, the age of acquisition of the first words, first sentences were at earlier ages, with these children acquiring abilities / skills earlier than other children, but losing them later, with a variable regression time. Furthermore, the child's overall level of development is lower in the regression

group. Finally, the general adaptive function in children with regression is slightly lower, but mainly there is a marked difference in the subtypes of communication and socialization.

Conclusion: It was possible to find a relation between these clinical characteristics and regression. Furthermore, it was possible to confirm that regression is not rare in ASD, however still low prevalence in comparison to others reports. This study enables us to characterize ASD children better with the main aim of an early and direct diagnostic approach during the research of the possible regression so that it is correctly evaluate what treatments/interventions are there to offer to each patient to improve children's and parents/caregivers quality of life.

Keywords: Autism; Autism Spectrum Disorder; Regression; Loss of skills; Neurodevelopmental milestones.

Resumo

Introdução: A perturbação do espetro do autismo é uma patologia do neuro desenvolvimento frequente caracterizada pela presença de dificuldades a comunicação e interação social, com padrão restrito e estereotipado de comportamento e interesses. O ratio de autismo por género é de 4:1 masculino: feminino. Autismo com regressão é descrito como um padrão característico em que as crianças perdem habilidades previamente adquiridas. O objetivo do nosso estudo é analisar casos de crianças previamente diagnosticadas com regressão, avaliar o seu neuro desenvolvimento e analisar a relação da regressão com quociente de desenvolvimento e a função adaptativa.

Métodos: Foi realizada uma análise retrospetiva de um grupo de 803 crianças e adolescentes com diagnóstico de autismo, entre 2010 e 2019, que foram acompanhadas no Centro Hospital e Universitário de Coimbra. Procedemos à análise do nível de desenvolvimento global da criança, da função adaptativa e da idade de aquisição de três etapas do desenvolvimento psicomotor: primeiras palavras, primeiras frases e início da marcha e comparados entre crianças com e sem regressão.

Resultados: Dos 803 pacientes, 15,2% das crianças tinha regressão e 84% do grupo eram do sexo masculino. Quanto ao quociente de desenvolvimento, o score GMDS foi significativamente mais baixo em crianças com regressão (68,8 ± 20,1) em comparação o grupo sem regressão (74,6 ± 21,0; p = 0,039). A função adaptativa global, nas crianças com regressão também apresentaram scores globais mais baixos (média ± DP, 58,2 ± 14,7), enquanto no grupo sem regressão, apesar do score ser ligeiramente mais elevado, continua <70 (média ± DP, 59,9 ± 14,4). Os subtipos VABS, o score de Communication (média ± DP, 58,5 ± 16,6) e o de Socialization (média ± DP, 63,7 ± 12,2) também foram significativamente mais baixos no grupo com regressão do desenvolvimento (p = 0,034), quando comparados com o grupo sem regressão (média ± DP, 62,4 ± 16,2 e 66,6 ± 13,0, respetivamente). A exceção é o subtipo de Daily Living Skills que tiveram os mesmos resultados nos dois grupos [com regressão (média ± DP, 60,6 ± 17,9) e sem regressão (média ± DP, 60,8 ± 15,9)]. Nas etapas do Neurodesenvolvimento, as crianças com regressão adquiriram as primeiras palavras (13,5 meses; p <0,001) e frases (38,0 meses; p = 0,337) em idade mais precoce, em comparação com o grupo sem regressão (24,0 meses; p <0,001) e 42,0 meses; p = 0,379, respetivamente). Não há diferença no início da marcha (14,0 meses para os grupos de regressão e não-regressão).

Discussão: O aumento dos diagnósticos por ano de regressão, deve-se, provavelmente, a um melhor conhecimento da doença e melhores métodos de diagnóstico. No grupo com regressão, em média, a idade de aquisição das primeiras palavras e primeiras frases foi mais

baixa, sendo que estas criança adquirem capacidades/habilidades mais cedo que que as outras crianças, mas perdem-nas mais tarde, com um tempo de regressão variável. Ademais, o nível de desenvolvimento global da criança é mais baixo no grupo de regressão. Por fim, a função geral adaptativa nas crianças com regressão também é mais baixa, principalmente nos subtipos de comunicação e socialização.

Conclusão: Foi possível encontrar uma relação entre essas características clínicas e a regressão. Além disso, foi possível confirmar que a regressão não é rara na perturbação do espetro do autismo, mas ainda com baixa prevalência em comparação com outros estudos. Este estudo permitiu caracterizar melhor as crianças com PEA com objetivo de procurar uma abordagem diagnóstica precoce e direta durante a pesquisa de uma possível regressão, para avaliar corretamente quais tratamentos / intervenções existem para oferecer a cada paciente, a fim de melhorar a qualidade de vida das crianças e dos pais / cuidadores.

Palavras-chave: Autismo; Perturbação Espectro Autismo; Regressão; Perda de capacidades; Marcadores do Neurodesenvolvimento.

Abbreviations

ADI-R – Autism Diagnostic Interview-Revised

ADOS – Autism Diagnostic Observation Schedule

ASD – Autism Spectrum Disorder

CHUC – Centro Hospitalar e Universitário de Coimbra

DQ – Developmental Quotient

DSM-5 – Diagnostic and Statistical Manual of Mental Disorders - fifth edition

EEG - Electroencephalography

F – Female

GDD – Global Developmental Delay

GDQ – Global Developmental Quotient

GMDS – Griffiths Mental Development Scales

HP – Hospital Pediátrico

IDD – Intellectual Developmental Disability

LD – Learning Disability

M – Male

OND – Other Neurodevelopmental Disorders

PMD – Psychomotor Development

SD – Standard Deviation

VABS – Vineland Adaptive Behaviour Scales

Introduction

Autism spectrum disorder (ASD) is a heterogeneous neurodevelopmental disorder with multiple behavioural and biological phenotypes. ASD is characterized by social communication and interaction difficulties as well as presence of restrictive and repetitive behaviour, interests and activities (American Psychiatric Association 2013, Bolte *et al.* 2018). Equally, ASD is a complex chronic multifactorial brain dysfunction whose aetiology is undetermined in approximately 80% of cases (Carter & Scherer 2013). In addition, it has a large genetic contribution, as it is estimated around 40% to 90% the range of heritability (Gaugler *et al.* 2014). The prevalence worldwide of autism is under 0.5 to 1% (in high income countries is estimated to be higher) with a distribution of four males (M) to one female (F) (Oliveira *et al.* 2007; Centers for Disease Control and Prevention 2009). In 2013, diagnostic criteria were reformulated into a broad diagnosis of ASD in the Diagnostic and Statistical Manual of Mental Disorders, 5th edition (DSM-5). There is consensus that substantial heterogeneity underlies the neurobiology of ASD.

ASD is manifested to beyond deficits in social interaction and communication has different levels of intellectual disability. Simultaneously, it can co-occur with other neurological, neurodevelopmental and psychiatric disorders including intellectual disability (most common), anxiety, hyperactivity and attention disorder, epilepsy, and sleep problems (Lord *et al.* 2020).

However, there is also heterogeneity when referring to neurodevelopmental trajectories and how, when, and to what extent regression will occur. Even though research and theories on regression and ASD dates to the middle of the last century, it was only in the past decade that our understanding of the topic changed drastically (Nordahl-Hansen 2019).

Signs and symptoms of autism have a gradual and variable time onset. It has been reported that the mean age of the diagnosis is around 4 to 5 years old, however, most parents express their first concerns to health practitioners at 2 years of age (Zuckerman *et al.* 2015). In most children with autism, symptoms generally arise in the second and third year of life, although, in the DSM-5 onset criteria, it might only be detected when children reaches school age or even later, even though it has always existed.

Furthermore, Pearson *et al.* 2018, conducted a study on a group of children that initially had a normal development followed by a period of loss or stasis, known as regression pattern and plateau/stagnation pattern, respectively. Regression is seen when children have developed and achieved milestones, followed by loss of previously acquired skills. As of plateau or stagnation, it is described as fail to progress after acquisition of initial skills.

Historically, the literature suggests that the behavioral signs of ASD emerge through two major distinct patterns of development: an early onset and a regressive one, in the first and later in the second year, respectively (Boterberg *et al.* 2019). The phenomenon of regression has been discussed in literature for several decades, since firstly reported by Theodor Heller, and the concept of regression has changed.

Nowadays, the interest in the etiological and diagnostic utility and clinical significance of regression in ASD continues (Barger *et al.* 2017, Pearson *et al.* 2018, Thurm *et al.* 2018). Few systematic studies that have been published seem to indicate that regression in autism is (1) not uncommon, but the relative prevalence is not known, and (2) associated with high rated of intellectual developmental disorder (IDD) in follow-up studies (Stefanatos 2008, Bradley *et al*. 2016). It is not consensual, but maybe regressive autism and autism with regression are not the same. The most frequent reported age of regression in ASD cases appears to be about 18 – 24(30) months of age (Baird *et al*. 2008, Stefanatos 2008).

In the present study we report ASD with or without plateau or regression (manifested by loss of language function and/or failure to progress socially) in the second year of life in our population sample, based on parent' information.

Other published reviews (Stefanatos 2008, Barger *et al.* 2013, Williams *et al.* 2015) have also examined rates and correlates of regression in children with ASD, mainly based on parent' reports. This retrospective approach usually has less reported frequencies than prospective studies (Lord *et al.* 2020).

A large meta-analysis on rates and onset of regression in ASD reported 32.1% overall prevalence rate for regression occurring at mean of 1.78 years. Regression prevalence rates differed according to four types of regression: language regression, 24.9 %; language/social regression, 38.1 %; mixed regression, 32.5 %; and unspecified regression, 39.1 %. Results also differed according to sampling method: population-based prevalence was 21.8 %, clinic-based prevalence was 33.6 %, and parent survey-based prevalence was 40.8 %. The risk of regression was equal for M and F (Barger *et al.* 2013).

As for the diagnosis of ASD, the gold standard is clinical judgement by an experienced neurodevelopment paediatrician or child psychiatrist, as there are no specific biomarkers available. In addition, it has been concluded that electroencephalography (EEG) and neuroimaging are not recommended for the diagnosis of ASD but can be in the etiological differential diagnosis with others neurodevelopmental disorders (OND) (Gurau *et al.* 2017, Lord *et al.* 2020). Nowadays, children are been diagnosed at younger ages which is the main goal for a specific, intensive and early psychoeducational intervention.

For assistance of the clinical diagnosis of ASD, clinicians use the best validated instruments: Autism Diagnostic Interview revised (ADI-R) (Le Couteur *et al.* 2003) and Autism Diagnostic Observation schedule second edition (ADOS-2) (Lord *et al.* 2012). In the case of early symptoms, it can be used parent reports and home recorded videos. Both together will improve the diagnosis (Ozonoff *et al.* 2019). Additionally, there is an increase in awareness and acknowledgement of the early signs of ASD. Simultaneously, with continuous neurodevelopmental evaluation in the health care system, the age of diagnosis has been reduced.

Our work is focused in the group of children with ASD and regression pattern. Autism with regression, as mentioned before, is described as a pattern where children loss skills that they had previously acquired. The loss of skills normally occurs between the 15 months (one year and three months) and 30 months (two years and a half), with a mean age of regression of one year and nine months of age (Pearson *et al.* 2018). However, in a more recent study, regression normally started at twelve months old (Ozonoff *et al.* 2019).

The loss of language with 3-5 words lost for three months is the most common regression (Baird *et al.* 2008). There are also other types like loss of social/language skills, i.e., loss of verbalization and other skills, like waving bye-bye, eye gaze, social interaction and playing skills (Werner *et al.* 2005). Gross motor skills loss is rare; however, some parents claim small decrease in manipulative skills (Davidovitch *et al.* 2000). It is also possible to have loss of language before the 5 words stage (like loss of proto-word and of babble) associated or not with loss of social interest and playing skills (Baird *et al.* 2008).

Therefore, it is important to consider at what age the regression occurred, the duration, what type, the amount of loss and at what level of neurodevelopment the child has (Barger *et al.* 2013), so that there is a mindful research of the possible underlying medical conditions that have led to the regression and correctly evaluate the diagnosis. Usually, after the period of loss of skills, there is a regain of skills. Also, for regression, it can be using the ADI-R and retrospective data (Lord *et al.* 2004).

The aetiology of regression is still unknown. It is inconclusive if environmental factors increase the regression process. However, some studies suggested that children with lower language and adaptive levels, and lower developmental or intelligence quotient (DQ/IQ), have higher chance of regression compared to others (Ozonoff *et al.* 2019). Moreover, other studies have not found difference (Davidovitch *et al.* 2000; Lord *et al.* 2004) or have found mixed results (Richler *et al.* 2006).

Many studies have suggested that regression only happened in a minor group of children with ASD, however, Ozonoff *et al.* 2019, that used prospective rather than retrospective methods,

have concluded that regression pattern is more frequent than previously thought and all children that developed ASD lose some skills, but at different rate and age.

The main goal of our study was to evaluate whether early neurodevelopmental milestones and signs of regression based on parent' information were related to development quotient (DQ) and adaptive functioning. Other aim was to provide a description of frequency and type of regression in our sample. In addition, we analysed the reported cases during the ten years period.

Methods

Participants

Participants included 803 patients (84.9% male) with the diagnosis of ASD, ranging in age from 2 to 18 years old, with a current age (years old), mean ± SD, 8.5 ± 3.6.

Participants were part of an outpatient clinic between 2010 and 2019 in the Neurodevelopment and Autism Unit of the Hospital Pediátrico (HP) – Centro Hospitalar e Universitário de Coimbra (CHUC), a referral tertiary hospital.

Our study, a retrospective one, was carried out with a subgroup of patients with ASD and at least one type of regression (n=110). This group of children with ASD derived from a population from our Unit and they were analysed mainly in clinical characteristics that have been related to regression and compared with ASD children without regression.

From the initial ASD group (n=803), there was no clinical information regarding regression in 77 (9.5%). So, in the remaining 726-patients, 110 (15.2%) have been reported with at least one type of regression (Figure 1).
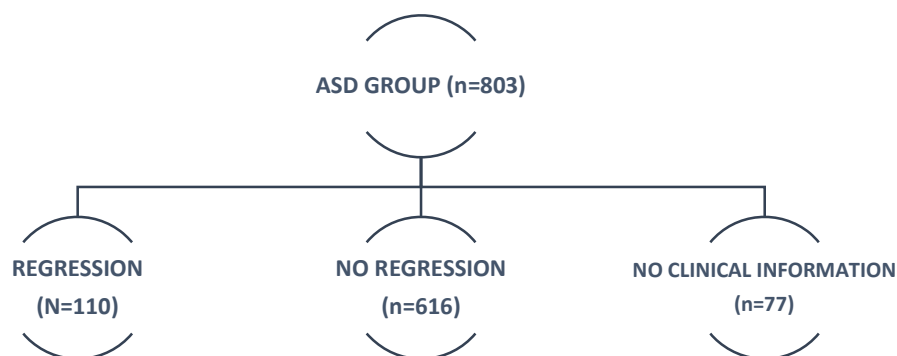

Fig 1. Flowchart of the study sample.

Measures

For the clinical diagnosis of ASD, parents/caregivers were interviewed with the ADI-R (Le Couteur *et al.* 2003) and the child observed with ADOS (Lord *et al.* 1989). Alongside, experienced neurodevelopmental paediatricians integrated in a multidisciplinary team conducted a clinical examination to all patients. Diagnostic criteria for ASD from DSM-5 were also fulfilled. Inclusion criteria was positive results in all evaluations (this means positive results in the ADI-R and ADOS for ASD) and met the criteria for ASD from the DSM-5.

The term gestational age was used to describe the age of the baby at birth and was expressed in weeks of gestation. Premature birth was defined as a delivery before 37 weeks of gestation.

Regression

Regression was defined in 2 ways: the definite language regression group which is a restrict loss of 2 to 5 words previously used for at least 3 months before loss. This loss can be with or without other types of skills losses. And regression was also considered apart from language regression if there was also loss from other skills (Baird *et al.* 2008). The number of children with language regression (105/726, ie, 14.5%) and/or psychomotor development (PMD) regression (19/726, ie, 2.6%).

Assessment of Early Neurodevelopmental Milestones

Furthermore, early neurodevelopmental milestones assessed in our study were: age for onset of independent walking, age for onset of first words and age for onset of first phrases. We considered the definitions of these milestones as described in the ADI-R (Le Couteur *et al.* 2003). Age for onset of independent walking was defined as the age (in months) at which the child takes unaided gait. Age for onset of first words was defined as the age (in months) at which the child first produced single words, in a consistent and meaningful way for the purposes of communication. Age for onset of first phrases was defined as the age (in months) at which the child first produced sentences composed of two or more words, one word being a verb, routinely used (Mouga *et al*. 2019).

ADI-R

ADI-R is a caregiver interview with aim to diagnose autism. It evaluates areas such as (1) communication and language, (2) reciprocal social interactions and (3) repetitive behaviours and interests.

The application of this scale is only recommended if the mental age of the child is at least 18 months. With this interview, we can have access to quantitative measures of behaviour in various areas like language and communication, repetitive behaviours or interests and social interaction.

Also, this instrument has specific questions about regression of language and other skills.

ADOS

ADOS evaluates the ASD severity from the ASD group and characterizes the symptomology of ASD. It is focused-on observation of social communication and behaviour implemented by the examiner and what types of behaviours in each activity during the overall ratings (these activities with standard tools are used to structure the interaction). The main objective is to see what type of activities the patient wants to participate in. It can be used at any age, language skills and development levels. It is an observational evaluation and consists of tasks (structured and semi-structured) given by a professional trained examiner to the patient.

It has 4 modules: 1. is for young children (who do not use phrase speech continuously); 2. is for patients who are not verbally fluent (expressive language used by 4-year-old children, usually), although have some phrase speech.; 3. is for children who are verbally fluent and play with toys appropriate for their age (children up to 12-16 years old); 4. includes tasks from module 3. and questions about daily routines (intended for adolescents and adults).

The patient is evaluated on his performance and behaviour, and it is given a score, that is assessed according to (1) communication and language and (2) reciprocal social interaction (Lord *et al* 1994).

Neurodevelopment assessment with Griffiths Mental Development scales (GMDS)

GMDS (Griffiths 1984) are scales used in Europe and measure the rate of neurodevelopment of children from birth to 8 years old. Children are evaluated in 6 areas which will allow a global evaluation of children's neurodevelopment: locomotor (gross motor skills), personal social (daily tasks, interaction with other children and level of independence), hearing and language, eye and hand coordination (fine motor skills and visual monitoring skill), performance (speed of work and precision), practical reasoning (solve practical problems like basic maths; children between 2 and 8 years old). After the evaluation of the subgroups, a global developmental quotient (GDQ) is calculated given the mental age (sum of all the tasks children reproduced successfully) by the chronological age. Mean value of DQ is 100 [standard deviation (SD) of 15].

In Portugal we use the scales referred to the 1984 UK norms. In the clinical practice, we use a neurodevelopment model in the first years of life. This is important as we identify the needs of each children so that there is an adaptation and planning of the best intervention and an adjustment to be made for the future on what parents and family need to do to take the best care of child with ASD (Mouga *et al*. 2016). For the evaluation of developmental quotient (DQ),

GMDS were used according to child mental or chronological ages. Higher score means higher level of child's global neurodevelopment.

Vineland Adaptive Behaviour Scale (VABS)

The VABS is a semi structured interview originated to assess global adaptive functioning and can be used at any age (Sparrow *et al.* 1984).

It is a combined score with several domains: daily living skills (DLS), communication (COM) and social skills (SOC). For each of the three domains, there are several subdomains that can be classified in five adaptive levels: low, moderately low, adequate, moderately high, and high.

As for the subdomains of DLS we have personal (this is how a person does their hygiene, dresses, put their shoes), domestic (domestic tasks given that are completed by the patient without any help) and community (how they manage their time and money). The subdomains of communication are expressive (this is how and what the patient says) and receptive (this is what the patient understands as the examiner uses both verbal and non-verbal communication). The subdomains of socialization are play and leisure, interpersonal relationships (interaction with others) and coping skills (how they react, respond are sensitive and how they are sensitive with others). (Mouga *et al.* 2015)

The adaptive behaviour composite (ABC) is the total score of the VABS, which is the sum of raw scores of DLS, SOC and COM. The domain scores are expressed as standard scores with a mean of 100 (SD of 15). As for the adaptive Level Domain and ABC Standard Scores: High (130 to 140), moderately high (115 to 129), adequate (86 to 114), moderately low (71 to 85) and low (20 to 70).

To evaluate the adaptive functioning/behaviour (this is daily life activities) we used VABS, standard global score and domains (DLS, SOC and COM). Normal score is 100 ± 15 and the higher the score, the higher the individual adaptive level.

All the patients in this study are followed, routinely, by this team at least 2 times per year and, as mentioned before, all clinical examinations of these patients were performed by an experienced multidisciplinary team in neurodevelopmental paediatrics.

To be included, all participants had to have an ASD diagnosis.

Procedure

Data was gathered from a database according to national policy on the archival research (*Portuguese Data Protection Authority*). The group of participants included in this study represents a subset of patients of our outpatient clinic, whose information is usually collected for clinical and research documentation. A total of 803 records meeting the inclusion criteria (from 2010 until 2019) were included in this study.

ASD groups were divided considering the global development quotient (GDQ), i.e., participants were divided, according to DQ, with Global Developmental Delay (GDD) or Intellectual Developmental Disability (IDD) if DQ < 70 and without if DQ ≥ 70.

Data Analysis

Initially we conducted an exploratory data-analysis using graphical techniques and quantitative analysis to characterize the sample, detect possible extreme outliers and measurement error. All statistical analyses were performed using the Statistical Package for the Social Sciences (SPSS), version 25 (SPSS ®, Chicago, IL, USA).

Descriptive statistics included mean, standard deviation, median and interquartile range for continuous variables, and absolute and relative frequency for categorical variables.

Distribution normality was assessed using the Kolmogorov-Smirnov test. Chi-square test, Fisher's Exact test, independent samples t-test, and Mann-Whitney test were used to establish associations and differences between variables, respectively.

The significance threshold was established at $\alpha < 0.05$.

Ethics statement

This study and all the procedures were reviewed and conducted in accordance with the declaration of Helsinki. Informed consent was obtained from the parents/guardians of all younger participants.

Results

The chronological age, gender distribution and gestational age are summarized in Table 1. It represents the demographics and clinical features of our paediatric ASD sample with 803 patients and a frequency of regression of 15,2% and a 5.6:1 M:F ratio. The mean ± SD age was 8.5 ± 3.6 years. Most patients were born at term (92.0%).

Table 1 – Clinical characteristics of the main clinical group of subjects with ASD (*n* = 803)

Gender, % (n)	
Male	84.9 (682)
Female	15.1 (121)
Current age (years), mean±SD	8.5±3.6
Gestational age (weeks), mean±SD	38.5±2.0
Premature (<37 weeks) % (n)	8.0 (64)
Term (37 to 42 weeks) % (n)	92.0 (738)

ASD Autism Spectrum Disorder; *SD* Standard Deviation.

The distribution of children diagnosed with ASD, according to age and sex is shown in Figure 2.
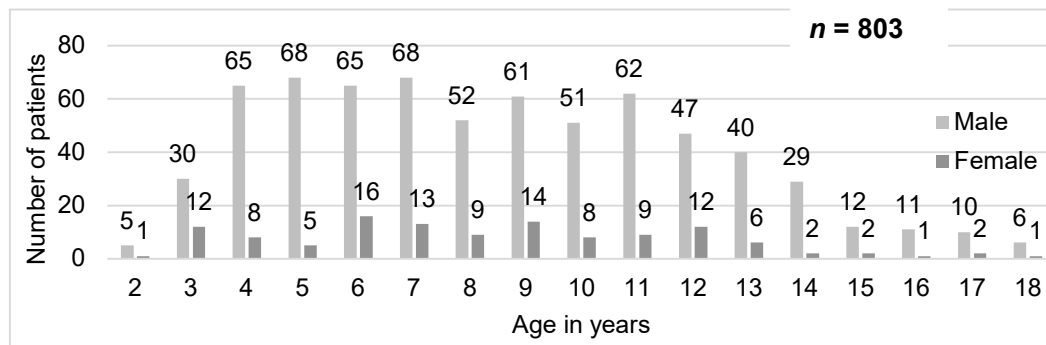

Fig.2 – Age (in years old) and sex distribution of ASD children

The number of ASD diagnosis *per year* is shown in Figure 3. There has been a stable number of diagnosis, with an increase tendency in the last few years.
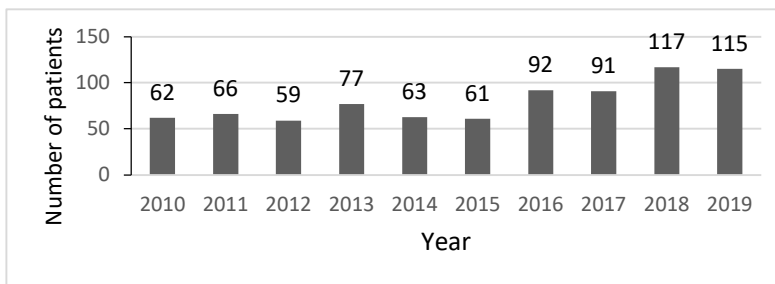

Fig. 3 – Number of ASD diagnosis *per* year.

We performed a quantification analysis of the number of ASD children with regression (*n* = 110/726). There was no information in 77 (9.6%) ASD patients regarding regression.

Associations between clinical variables of interest and neurodevelopmental regression are shown in Table 2. Furthermore, ASD cases with regression are mainly in children with language regression (*n* = 105). No significant associations or differences were found in terms of sex and gestational age.

Table 2 Association between neurodevelopmental regression and clinical variables between clinical subgroups (Regression and No Regression). (*n* = 726)

| Variables | Regression | | Test statistics |
	Yes (*n* = 110)	No (*n* = 616)	P-value
Sex			χ^2=0.273; *p* = .601
Male	86.4 (95)	84.4 (520)	
Female	13.6 (15)	15.6 (96)	
GA			χ^2=3.355; *p* = .084
Premature	3.6 (4)	8.8 (54)	
Term	96.4 (106)	91.2 (561)	

GA Gestational age; Premature <37 weeks GA; Term 37 to 42 weeks GA.

Concerning GMDS, 566/726 children have been evaluated. The DQ average score of all ASD group was 73.7 ± 21.1. The GMDS score was significantly lower in children with regression comparing to those without it (χ^2=4.870; p =.039). as shown in Table 3.

Moreover, GMDS mean value was significantly different between subgroups (with and without regression) and a GMDS score lower than 70 was more likely to be found in patients with regression (t=2.066; p = .037).

Table 3 – DQ association between subgroups (*Regression* and *No Regression*).

Variables	Regression		Test statistics
	Yes (n=110)	No (n=616)	*P*-value
GMDS, mean ± SD	68.8 ± 20.1	74.6 ± 21.0	χ^2=4.870; p=.039 *
<70	55.1 (38)	41.0 (204)	t=2.066; p=.037 *
≥70	44.9 (31)	59.0 (293)	t=2.066; p=.037 *

All comparisons signalled with * are significant. *GMDS* Griffiths Mental Development scales; *DQ* Developmental quotient; *SD* Standard Deviation

Concerning VABS, 339/726 children have been evaluated. The adaptive behaviour composite (ABC) is the total score of the VABS, which is the sum of raw scores of communication, socialization and daily living skills represented in Table 4. The global score, this is the adaptive behaviour composite, was <70 therefore children with ASD had low adaptive function.

Table 4 VABS scores' descriptive statistics	
VABS (*n* = 339)	Mean ± SD
Communication	61.5±16.5
Daily living skills	60.7±16.5
Socialization	65.8±12.8
Global (ABC)	59.5±14.4

VABS Vineland Adaptive Behaviour Scale; *SD* Standard Deviation.

When looking at the subgroups, we had a significantly lower adaptive level in communication subdomain in patients with neurodevelopmental regression (Mann-Whitney U = 12030.5; p = .034). Others VABS subdomains did not significantly differ between subgroups (with and without regression) (Table 5).

Table 5 VABS adaptive levels association between regression subgroups

VABS (n = 339)	Regression		Test statistics
	Yes (n=110)	No (n=616)	P-value
VABS	mean ± SD	mean ± SD	
Communication	58.5±16.6	62.4±16.2	U=12030.5$; p$ =.034*
Daily living skills	60.6±17.9	60.8±15.9	U=13703.5; p =.641
Socialization	63.7±12.2	66.6±13.0	U=12201.0; p =.051
Global (ABC)	58.2±14.7	59.9±14.2	t=0.999; p =.318

All comparisons signalled with * are significant. *VABS* Vineland Adaptive Behaviour Scale; *SD* Standard Deviation.

Regarding neurodevelopmental milestones, clinical characteristics of all ASD children (803), diagnosed between 2010-2019 are shown in Table 6. Most patients were able to walk (97.8%) and had spoken his or her first words (90.3%).

Table 6- Samples' descriptive statistics, ND milestones in the ASD group.

Walking, % (n)	97.8 (785)
Age at onset of walking (months), median (IQR)	14.0 (5.0)
First words, % (n)	90.3 (700)
Age at onset of first words (months), median (IQR)	24.0 (16.0)
First phrases, % (n)	63.6 (472)
Age at onset of first phrases (months), median (IQR)	42.0 (12.0)

IQR Interquartile Range.

Children with neurodevelopmental regression had a significantly lower median age in terms of onset of the first words (13.5 months; p <.001) and phrases (38.0 months; p=.379) when compared to non-regression group shown in Table 7. On the other hand, the age of onset of walking was the same for both clinical groups (14.0 months for regression and non-regression groups).

Table 7- Association between development regression and neurodevelopment milestones between the two main clinical groups (*Regression and No Regression*).

ND Milestones	Regression		Test statistics
	Yes (n=110)	No (n=616)	P-value
Walking, % (n)	100.0 (110)	98.9 (609)	p =.039* (Fisher's Exact test)
Age at onset of walking (months), median (IQR)	14.0 (4.0)	14.0 (5.0)	*U*= 31986.5*; p* =.715
First words, % (n)	94.5 (104)	89.8 (544)	χ^2=2.471; p =.155
Age at onset of first words (months), median (IQR)	13.5 (7.0)	24.0 (13.0)	*U*=14712.0*;* p=<.001*
First phrases, % (n)	44.9 (48)	67.2 (391)	χ^2=19.480; p=<.001*
Age at onset of first phrases (months), median (IQR)	38.0 (16.0)	42.0 (12.0)	*U*=8063.0*;* p=.379

All comparisons signalled with * are significant. *ND* Neurodevelopment; *IQR* Interquartile Range.

Discussion

Regression involves a loss of speech or social adjustment but may include both. Contemporaneous studies evidenced that regression is more common than previously thought. However, this evidence came mostly from prospective studies. In our study, a retrospective analysis, we found 15.2% frequency of regression of a subgroup of ASD children from a large ASD population sample regularly followed in our outpatient clinic, in accordance of other retrospective data published. For this purpose, we used a subgroup of 803 children with ASD diagnosed over the last ten years, between 2010 and 2019, although it remained a very large sample, with clinical features well characterized.

Other strengths of our study were the consistency of followed up throughout the years and a diagnosis based on gold standard instruments and a well-trained multidisciplinary team. The ASD diagnosis are accurate using gold standard and validated instruments such as ADI-R and ADOS, and DSM-5 criteria fulfilment as internationally preconized.

Throughout the years, more children have been diagnosed with ASD, maybe by an increase of awareness, but also due to an increment of referrals for diagnosis, which corroborates a global tendency.

As mentioned, prevalence rate of regression in our clinical sample (15,2%) was in the lower range of previous studies that reported the rate of regression ranging between 12,5 to 50% (Rogers *et al* 2004, Stefanatos *et al* 2008). The prevalence is influenced by the variation of methods used and operational definition of regression used by researchers (Ozonoff 2019, Lord 2020). Also, the sample size and type influence drastically the prevalence rate [smaller samples or samples from clinical referrals have higher percentages, suggested by Roger 2004, whereas population samples have lower average prevalence rates (Taylor *et al* 2002)]. All will contribute to increase the knowledge of this changing concept (Stefanatos *et al* 2008, Ozonoff 2019).

Our sample had a M:F ratio superior than 4:1 as commonly accepted sex ratio, yet it is explained by most cases being high function. Higher rates of high function autism have a pronounced M:F ratio, even 7:1. Other series (Goldberg et al 2003, Kurita 1985, Wilson et al 2003), reported a M:F ratio 2,7-6:1.

Like Barger *et al*. 2013, a meta-analytic review, found no significant difference in rates of regression for males and females, we did not find sex difference with significance.

Besides they concluded that language regression had the lowest rate of all regression types evaluated (even lower than unspecified regression). However, in a recent study, loss of communication, as in language regression, was the most frequent type of loss in regressive

ASD (Gadow *et al.* 2017). In our sample, 14,6% of children had language regression. We found a growing language regression cases reported in the last few years using retrospective data from a computerized database, but we cannot conclude if it is a growing notification or a true increment in cases. On the other hand, this difference might be because, as suggested by others (Stefanatos *et al* 2008, Meilleur *et al* 2009), language regression can be easily noticed and detected when compared to other types of regressions. In general, parents seek for medical help if the loss is related to speech, as it is more difficult to understand a specific social skill loss than language regression.

We highlight a systematic neurodevelopmental milestones records, as previously published by Ferreira & Oliveira, 2016. They found that the age of onset of the first phrases in children with ASD was an important clinical milestone as it was concluded that its delay relates to lower non-verbal and verbal intellectual skills.

ASD children with neurodevelopmental regression had a significantly lower median age in terms of onset of the first words (13.5 months; $p<.001$) when compared to children without regression (U=14712.0; $p<.001$). Baird 2008 had already concluded children first words acquisition was at a significantly younger age compared with the non-regression group, however there was no difference in the age of acquiring phrases.

In addition, within language regression group displayed a more typical child's neurodevelopment shown by earlier first words. Furthermore, in our study, there were no differences regarding the age of onset of walking in the two groups, contrasting with others (Jones and Campbell *et al*. 2010, Thompson *et al*. 2019).

Baird (2008), concluded that when comparing regression and non-regression groups, there was no significant difference in terms of DQ. In our study, using the Griffiths GDQ, children with regression, had lower scores (mean ± SD; 68.8 ± 20.1), therefore they were more likely to have GDD or IDD (score <70) than children without regression (mean ± SD; 74.6 ± 21.0; χ^2=4.870; p =.039), as it was expected and has been reported by Kobayashi & Murata 1998, Thompson *et al*. 2019.

Noteworthy, children with neurodevelopmental disorders, including ASD and IDD, have been characterized as having lower scores of verbal intelligence and performance intelligence. Global intellectual level and specific cognitive deficits are characteristic of children with ASD (Mouga *et al*.2016).

Furthermore, it has been concluded that ASD population have lower adaptive scores. Children with ASD fall behind in adaptive functioning compared with children with other

neurodevelopmental disorder, and socialization domain remains as a distinctive factor of primary diagnosis of ASD (Mouga *et al.*2015).

When looking at the scores of the subdomains of VABS, we concluded that children with regression have lower scores of socialization (mean ± SD; 63.7±12.2) and communication (mean ± SD; 58.5±16.6) comparing with the non-regression group (mean ± SD; 66.6 ± 13.0 and 62.4 ± 16.2, respectively). It was possible to find a relation between these clinical characteristics and regression; however, further studies would help in deepening our ASD understanding.

A limitation of our study was the fact that the information about the early neurodevelopmental milestones and possibility of regression was obtained by caregivers, which is subject to interpretation and memory bias (Ozonoff *et al*. 2019), maybe inflating symptoms severity and reporting later onset of language loss (Jones *et al*. 2015, Pearson *et al*. 2018). In the future we recommend analysing a precision data, at what time regression was noticed.

Nevertheless, we studied a large group of patients and the data was systematically obtained by experienced neurodevelopmental paediatricians, which reduces inaccuracy. Furthermore, as the diagnostic tools for this study were based on retrospective observation, it has been reported that, regression can be underreported (Ozonoff *et al*. 2018).

Conclusion

This research with a very large sample highlights that regression is not rare in ASD, although our study has a low frequency compared to others. A continuous characterization of ASD population, paves the way for helping early psychoeducational intervention programs.

Conflict of Interest

All authors have declared that no conflict of interests exists.

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