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# Autism Spectrum Disorder Profile in Neurofibromatosis Type I

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**Abstract** Neurofibromatosis Type 1 (NF1) is a common autosomal dominant single-gene disorder, in which the co-occurrence of autism spectrum disorder (ASD) has attracted considerable research interest recently with prevalence estimates of 21–40 %. However, detailed characterization of the ASD behavioral phenotype in NF1 is still lacking. This study characterized the phenotypic profile of ASD symptomatology presenting in 4–16 year old children with NF1 (n = 36) using evidence from parent-rated Social Responsiveness Scale and researcher autism diagnostic observation Scale-2. Compared to IQ-matched reference groups of children with autism and ASD, the NF1 profile shows overall similarity but improved eye contact, less repetitive behaviors and better language skills.

**Keywords** NF1 · ASD · Neurofibromatosis Type 1 · Autism spectrum disorder · SRS · ADOS

## Introduction

Autism spectrum disorder (ASD) is a common neurodevelopmental disorder with a population prevalence of about 1 % (Baird et al. 2006). It is behaviorally defined and diagnosed, characterized by deficits in the clinical dyad of social communication and interaction along with restricted and repetitive behaviors and interests. It is highly heritable with monozygotic twin concordance rates of about 60–90 %. Over the last three decades, concerted research efforts into the etiology of ASD have found striking genetic heterogeneity, with a large number of highly penetrant de novo germline mutations and rare inherited ASD variations distributed acro
ss many genes (Pinto et al. 2014). Alongside this, there is wide variation in the phenotypic presentation, not only in the pattern and severity of symptoms but also in the occurrence of psychiatric comorbidities.

The considerable genetic and phenotypic heterogeneity has been a limiting factor in furthering our insights into causal mechanisms in autism and in the search for targeted

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interventions. Two parallel approaches have been used to reduce this heterogeneity. One approach, which has proved useful, has been to subset the ASD sample according to specific phenotypes; by sex of proband, language, or using behavioral features such as autistic regression (Abrahams and Geschwind 2008). The other has been to study behavioral profiles of genetically homogenous groups, which manifest ASD symptoms at higher than expected frequencies compared to the general population. This so-called 'syndromic autism' accounts for approximately 10–20 % of all ASD cases (Geschwind 2011). Examples include Fragile X, Tuberous Sclerosis, Cornelia de Lange syndrome, Timothy syndrome, Angelman's syndrome—all of which have a documented high prevalence of ASD (Oliver et al. 2011).

Recently, there has been considerable research interest in the co-occurrence of ASD in another single gene disorder, Neurofibromatosis Type 1 (NF1). NF1 is a common autosomal dominant single-gene disorder caused by a mutation in the *NF1* gene located on chromosome 17q11.2. It has an estimated birth incidence of one in 2,700 and prevalence of one in 4,560 (Evans et al. 2010). NF1 is clinically diagnosed based on the presence of distinctive cutaneous features such as café-au-lait spots, skin-fold freckling and neurofibromas. It is a clinically heterogeneous disorder with marked inter and intra-familial variability. Genotype phenotype correlation studies have largely failed to explain the phenotypic variability; 5 % of the NF1 (Table 1) gene mutations are microdeletions which are linked to a more severe clinical phenotype including dysmorphic features, intellectual impairment, cardiovascular malformations and a higher burden of benign and malignant tumours (Mautner et al. 2010).

The commonest associated complication of NF1 in childhood relates to cognitive dysfunction. Mean intelligence quotient scores of children with NF1 fall within the average range; nevertheless, approximately 70 % of children underachieve academically. The cognitive-behavioral phenotype of NF1 is characterized by specific learning difficulties, neuropsychological deficits, attention deficit hyperactivity disorder (ADHD), and social, emotional and behavioral problems (Lehtonen et al. 2013). ADHD is most commonly reported with prevalence estimates ranging from 30–67 % (Coude et al. 2007).

The co-occurrence of ASD in NF1 was first noted over three decades ago; Gillberg and Forsell 1984 reported simultaneous occurrence of NF1 in three out of 51 children with infantile autism in the community. A retrospective case note analysis of 341 children with infantile autism seen at child psychiatry clinics over a 25 year period found only one case of co-occurrence of NF1 and autism (Mouridsen et al. 1992). Similarly Fombonne et al. (1997) conducted an epidemiological survey among 300,000 children and found NF1 in 0.6 % of children with autism. Published literature over the last 10 years has described social and communication impairments in the NF1 population but without integrating this within an ASD syndromic framework. Barton and North (2004) found children with NF1 had significantly poorer social outcomes as compared to their unaffected siblings with the presence of ADHD as an important mediator of social functioning. Noll et al. (2007) found that children with NF1 were perceived as having more social problems by parent, teachers and classroom peers. Huijbregts et al. (2010) found that as compared to controls, children with NF1 have problems in social information processing, perform poorly on tasks such as identification and matching facial emotions and have significant parent reported autistic symptomatology. In a two phase population based epidemiological study ($n = 109$) with gold standard assessment instruments of autism diagnostic inventory-revised (ADI-R) and Autism Diagnostic Observation Schedule (ADOS), Garg et al. (2013) demonstrated prevalence estimates of 25 % ASD in NF1 with a further 21 % broader ASD phenotype. In a retrospective, cross-sectional design

**Table 1** Demographic characteristics of NF1 + ASD children and autism and non-autism ASD reference groups obtained from the ADOS manual

| | NF1 + ASD study group ($n = 36$) | Autism reference group ($n = 129$) | Non-autism ASD reference group ($n = 186$) |
|---|---|---|---|
| Mean age (*SD*) | 11.09 (3.19) | 8.41 (2.45) | 8.38 (2.51) |
| Gender | | | |
| Male | 75 | 87 | 81 |
| Female | 25 | 13 | 19 |
| Mean VIQ (*SD*) | 94.00 (14.60) | 85.46 (23.35) | 99.61 (21.52) |
| Heritability | | | |
| Familial | 50 | – | – |
| De novo | 47 | – | – |
| Missing | 3 | – | – |

*NF1* neurofibromatosis type 1, *ASD* autism spectrum disorder, *VIQ* verbal IQ

($n = 66$), with parent-reported measures, Walsh et al. (2013) suggested 40 % prevalence of ASD symptomatology. In a hospital-based NF1 pediatric population ($n = 82$) using the children's social behavior questionnaire (CSBQ) and social responsiveness scale (SRS), Plasschaert et al. (2014) found a high prevalence of ASD characteristics, more frequently reported above the age of 8 years. Replicating the previous studies, a prevalence estimate of 26 % was reported by extensive diagnostic ASD assessment in a subgroup of these children, clinically selected based on parental concerns and observations by a multidisciplinary team. Item-level analyses suggested that these NF1 + ASD children may have a distinctive social behavioral profile compared to general ASD.

It is well known that the prevalence of ASD rises with the severity of intellectual impairment. Indeed Skuse (2007) suggests that intellectual impairment rather than any other syndrome-specific cause largely explains the increased autism prevalence in genetic syndromes. Disorders such as Cornelia de Lange and Angelman's syndrome are associated with severe intellectual impairment (Deardorf et al. 2005). By contrast in NF1, the mean IQ of children is within the average range, which suggests that this is a true co-occurrence and intellectual impairment alone fails to explain the increased prevalence rates. NF1 has a well-described neurobiology explaining the cognitive phenotype (Diggs-Andrews and Gutmann 2013). The loss of neurofibromin in NF1 is associated with increased rat sarcoma (Ras)/MAPK (mitogen-activated protein kinase) signaling impairing synaptic plasticity leading to the cognitive phenotype (Costa et al. 2002). A number of putative interventions such as statins (Krab et al. 2008; Acosta et al. 2011; van der Vaart et al. 2013) and calcium channel blockers (Kallarackal et al. 2013) have been studied in the *NF1* mouse model capable of reversing the cognitive phenotype. Evidence is also emerging implicating the Ras/MAPK pathway as being important in the etiology of ASD. Recently Pinto et al. (2014) showed that genes most affected by copy number variations (CNVs) and single nucleotide variations (SNVs) converge on three neural system functional networks including the MAPK signaling pathway. Adviento et al. (2014) have shown increase prevalence of autism traits in the disorders on the Ras/MAPK pathway (collectively known as the Rasopathies).

NF1 may thus be highly rewarding to study in the context of ASD; however detailed characterization of the ASD behavioral phenotype is lacking. We also need to understand how the *NF1* mutation influences the ASD phenotypic presentation on the core domains; whether the diagnosis is driven by extreme scores in any particular domain. To our knowledge this is the first study to thus characterize the NF1 + ASD phenotype. The aim of this study is twofold: characterize the ASD profile in NF1 and to compare it to idiopathic ASD.

## Methodology

### Recruitment

For the current study, participant data were collected from the Regional genetics service at Central Manchester University Hospitals Foundation NHS Trust in the United Kingdom and the Center of Human Genetics, University Hospital of Leuven in Belgium. Participant data were gathered from existing databases with the following inclusion criteria (1) Meeting the ASD cut-off on the social responsiveness scale ($T$ scores $\geq 60$) (2) Scoring above ASD cut-off score of the autism diagnostic observation schedule–second edition (ADOS-2) module 3 (overall total of social affect and restricted and repetitive behaviors $\geq 7$). NF1 diagnosis was made clinically using the National institutes of health (NIH) diagnostic criteria (National Institutes of Health Consensus Development Conference 1988).

For the Manchester group, NF1 children aged 4–18 years were originally recruited and assessed for ASD as part of two independent studies: an earlier published epidemiological study of ASD prevalence in NF1(Garg et al. 2013) and an ongoing randomized controlled trial of Simvastatin in children with NF1 + ASD. For the Leuven group, NF1 children aged 5–17 years were recruited from the NF1 clinic and assessed for ASD on the basis of clinical suspicion and/or parental concerns (Plasschaert et al. 2014).

### Participants

In this study, a total of 36 children with NF1 + ASD met the inclusion criteria. The mean age of the sample was 11.09 years (*SD* 3.19, range 4.6–16.1 years) and the sample consisted of 27 boys (75 %) and 9 girls (25 %). Familial (50 %) and de novo (47 %) NF1 cases were equally divided in the NF1 + ASD group, with the manner of inheritance in one child remaining unknown, since this child was adopted. All children were administered with the Weschsler's Abbreviated Scales for Intelligence (WASI) (Wechsler 2004) and vocabulary and similarities subtest were used to present the level of verbal intellectual abilities. Their mean verbal IQ (VIQ) was 94.00 (*SD* 14.60, range 64.0–126.0).

For the SRS, standardized $T$-scores on the total problem scale and the different subdomains are reported. When analyzing ADOS scores, NF1 + ASD children were compared to normative data of children with autism ($n = 129$) and children with ASD ($n = 83$), as reported in the ADOS-2 manual (Lord et al. 2012). No statistically significant difference was observed in the sex ratio or the VIQ of the NF1 + ASD group compared to the autism and

ASD normative groups (VIQ in NF1 + ASD/autism ($t$ (212) = 1.60; $p = .110$ and NF1 + ASD/ASD ($t$ (284) = −1.90; $p = .06$); sex ratio in NF1 + ASD/autism ($\chi^2$ (1) = 1.43, $p = .23$ and NF1 + ASD/ASD ($\chi^2$ (1) = 0.04, $p = .84$). The NF1 sample was significantly older (NF1 + ASD/autism: $t$ (212) = 6.27, $p < .01$ and NF1 + ASD/ASD: $t$ (284) = 5.45, $p < .01$).

## Measures

The *Social Responsiveness Scale* (SRS) (Constantino et al. 2003) for children and adolescents aged 4–18 years, is a normed 65-item rating scale developed to screen for a wide range of behaviours characteristic of ASD. The scale is completed by parents rating the social interactions of their child in their naturalistic social context(s). Ratings are given by frequency of occurrence on a scale from 1 (not true) to 4 (almost always true). The SRS generates a *total problem score*, describing the severity of social deficits in the autism spectrum, while also generating five subscale score assessing *social awareness*, *social cognition* (information processing), reciprocal *social communication*, *social motivation* (anxiety/avoidance), and *autistic mannerisms* (preoccupations and traits). Higher scores on the SRS indicate greater severity of social impairment.

The *Autism diagnostic observation schedule–second edition* (ADOS-2) (Lord et al. 2012) is a semi-structured, standardized observational assessment of communication, social interaction, play and imaginative skills, and repetitive behaviors. It consists of 4 modules appropriate to different developmental age and expressive language skills. In the current study, only NF1 + ASD children assessed with the ADOS Module 3 were included, since this was the only group with a reliable sample size ($n = 36$). Thirty NF1 + ASD children were measured with the ADOS. These observations were scored according to ADOS-2 algorithms. Since item 'Amount of social overtures' was only scored when using ADOS-2 observations ($n = 6$), it was excluded from the analyses. Module 3 is intended for verbally fluent children and young adolescents. Verbal fluency was defined as 'producing a range of sentence types and grammatical forms, using language to provide information about events out of the context of the ADOS, and producing some logical connections within sentences'. In the current study, item, algorithm and comparison scores will be used. Scores on individual items range from 0 (no evident abnormality) to 3 (marked abnormality) in domains of social communication, social interaction, imagination and restricted, repetitive behaviors.

Thresholds consistent with ASD diagnosis are obtained from an *overall total algorithm score*, which is the summation of *social affect* (SA: combination of social interaction and social communication scores) and *repetitive and restricted behavior* (RRB) algorithm scores. These scores are compared with cut-off scores to provide one of three classifications: autism, autism spectrum, and non-spectrum. The difference between autism and autism spectrum classifications is one of severity, with the former indicating more pronounced symptoms.

A *comparison score*, which is a continuous metric ranging from 1 to 10, allows to compare a child's overall level of ASD symptoms to that of ASD children with the same age and similar language skills. Consequently, it can be expressed as one of four descriptive categories—from 'no evidence of autism spectrum-related symptoms' to 'a high level of autism spectrum-related symptoms'.

## Procedure

The study was approved by the Ethics committee of the University Hospital of Leuven (ML8552-S54631) and the Greater Manchester West & Central Manchester Ethics committees (REC 11/NW/0838 and 13/NW/0111). The SRS was completed by the parent or primary caregiver. ADOS-2 assessments were video-recorded, administered by trained researchers and scored immediately after administration.

## Statistical Analysis

Data were analyzed in SPSS version 17 and Sigmastat version 4.0. Demographic and clinical characteristics of participants were compared with norm groups by using 2-sample $t$ tests and $\chi^2$ tests. Categorical data are presented as percentages (frequency) and compared by using the $\chi^2$ test. To compare ADOS item and algorithm scores, the effect of group (NF1 vs. TD and NF1 vs. ASD) was investigated. An ANOVA test was applied, with Tukey–Kramer correction for post hoc tests. A $p$ value of $<0.05$ was considered significant.

We also looked at the differences at the data according to study centers (Leuven or Manchester). No significant differences were observed in the SRS data. For the ADOS Total score, a marginally significant difference was observed between study groups ($p = .055$), with the Manchester group having higher average scores than the Leuven group. Having a closer look at ADOS domain scores, this was caused by significantly average higher scores in the Manchester group for the SA domain ($p = .007$). On item level, significant differences were found on few algorithm items: A5 (offers information; $p = .020$), B1 (eye contact; $p = .001$), B2 (facial expressions; $p = .025$) for which the scores were overall higher in the Manchester group than in the Leuven group.

## Results

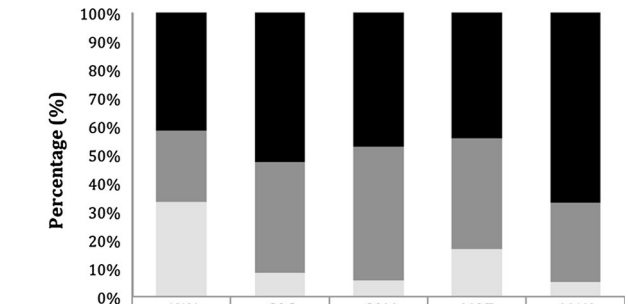
### SRS Questionnaire

The mean SRS total problem *T*-score of the NF1 sample was 78.58 (*SD* 11.37, range 60–107). Forty-four percent of this group presented with mild to moderate problems with *T*-scores ranging from 60 to 75, while another 56 % had severe problems with *T*-scores reaching 76 or higher. On the SRS subscales, the mean *T*-scores were as follows: social awareness 68.89 (*SD* 15.50), social cognition 76.22 (*SD* 13.20), social communication 76.47 (*SD* 13.10), social motivation 73.78 (*SD* 13.41) and autistic mannerisms 79.08 (*SD* 12.66). A significant negative correlation was found between VIQ and the SRS total *T*-score ($r = -553$; $p = .001$). The distribution of SRS subscale scores is shown in Fig. 1.

### ADOS-2 Comparison and Algorithm Domain Scores

As based on selection criteria, all children met ASD cut-off scores on ADOS-2. In terms of severity using the ADOS-2 *comparison* scores, 11 % ($n = 4$) had a low level of autism spectrum symptoms, 66 % ($n = 24$) had moderate symptoms and 23 % ($n = 8$) had a high level of symptoms.

Mean ADOS-2 algorithm domain scores are presented in Table 2. There were statistically significant group differences in the *SA* domain across the three groups with scores being the highest for the autism group, followed by scores of the NF1 group and ASD group ($F$ (2,348) = 40.5, $p < .001$). Statistically significant group differences were also present on the *restricted and repetitive behavior (RRB)* domain with the NF1 group showing the lowest scores as compared to the autism and ASD groups ($F$ (2,348) = 31.0, $p < .001$). The autism group had the highest overall total algorithm scores (mean 14.73, *SD* 0.43) followed by the NF1 group (mean 11.11, *SD* 0.61) and the ASD group (mean 9.34, *SD* 0.32). No significant correlations were found between VIQ and ADOS domain scores (all $p > .05$).




| | AWA | COG | COM | MOT | MAN |
|---|---|---|---|---|---|
| Severe | 42 | 53 | 47 | 44 | 67 |
| Mild/moderate | 25 | 39 | 47 | 39 | 28 |
| Normal | 33 | 8 | 6 | 17 | 5 |

**Fig. 1** The distribution of mild/moderate ($T \geq 60$) and severe ($T \geq 76$) problems on SRS subscales as scored by parents. AWA, Social awareness; COG, Social cognition; COM, Social communication; MOT, Social Motivation; MAN, Social mannerisms

### ADOS-2 Algorithm Item Scores

Algorithm item scores are also shown in Table 2. On the *SA* algorithm items, there were no significant differences between the NF1 + ASD group and the autism group apart from unusual eye contact which was worse in the autism group. On all of the *RRB* items, NF1 + ASD children had significantly lower scores compared to autism children.

The NF1 + ASD group in comparison to the ASD group was more impaired on the *SA* algorithm items for *language and communication* items including conversation and use of gestures and *reciprocal social interaction* items including facial expressions directed to examiner, shared enjoyment in interaction, quality of social overtures, quality of social responses, amount of reciprocal social communication and overall quality of report. In contrast, the ASD group was significantly more likely to have poor eye contact. A borderline significant trend was found for 'reporting of events', with more problems observed in NF1 children compared to ASD children. On the *RRB* algorithm items, no significant differences were found between NF1 + ASD and ASD children, except for the item assessing the use of stereotyped words/phrases, which was significantly more impaired in ASD children.

### ADOS-2 Non-Algorithm Item Scores

Non-algorithm item scores are presented in Table 3. On the non-algorithm items, the NF1 group was less impaired compared to autism children for *language and communication* items related to overall level of non-echoed language, speech abnormalities associated with autism and echolalia. However, they were equally impaired for items assessing offering and asking for information. Nonetheless, for this latter item, the difference was borderline significant. Similarly on *reciprocal social interaction* items, NF1 + ASD children were less impaired than autism group on items concerning language production and linked non-verbal communication, comments on others emotion/empathy and insight into typical social situations and relationships. No statistical differences were found in *imagination/creativity* across the two groups. On *RRB* non-algorithm items, NF1 + ASD children were significantly less impaired to the autism group on presence of compulsions/rituals.

When comparing NF1 + ASD to ASD children, statistical differences were found in only two *language and*

**Table 2** Group comparisons for NF1 versus Autism and NF1 versus ASD children on ADOS-2 algorithm domain and item scores

| ADOS-2 algorithm items | Mean (*SD*) | | | *F* | *p* | Post-hoc pairwise comparisons | |
|---|---|---|---|---|---|---|---|
| | NF1 + ASD, $n = 36$ | Autism, $n = 129$ | ASD $n = 186$ | | | NF1 + ASD/ Autism | NF1 + ASD/ ASD |
| Social affect (SA) | 9.89 (0.56) | 11.40 (0.36) | 7.29 (0.30) | 40.59 | **<.001** | .114 | |
| Language and communication | | | | | | | |
| Reporting of events | 1.22 (0.64) | 1.40 (0.87) | 0.91 (0.81) | 13.97 | **<.001** | .472 | .093 |
| Conversation | 1.25 (0.65) | 1.43 (0.62) | 0.92 (0.70) | 22.83 | **<.001** | .324 | **.018** |
| Descriptive/conventional/instrumental or informational gestures | 0.86 (0.64) | 0.91 (0.81) | 0.52 (0.63) | 12.80 | **<.001** | .924 | **.021** |
| Reciprocal social interaction | | | | | | | |
| Unusual eye contact | 0.61 (0.93) | 1.57 (0.80) | 1.05 (0.95) | 21.42 | **<.001** | **<.001** | **.019** |
| Facial expressions directed to examiner | 0.92 (0.44) | 1.01 (0.59) | 0.63 (0.58) | 17.74 | **<.001** | .681 | **.015** |
| Shared enjoyment in interaction | 0.78 (0.87) | 0.71 (0.73) | 0.37 (0.56) | 12.73 | **<.001** | .841 | **.002** |
| Quality of social overtures | 1.06 (0.58) | 1.08 (0.66) | 0.67 (0.69) | 16.10 | **<.001** | .986 | **.004** |
| Quality of social responses | 1.06 (0.33) | 1.14 (0.56) | 0.76 (0.53) | 21.18 | **<.001** | .698 | **.005** |
| Amount of reciprocal social communication | 1.17 (0.66) | 1.26 (0.70) | 0.77 (0.72) | 19.59 | **<.001** | .778 | **.005** |
| Overall quality of report | 1.08 (0.60) | 1.11 (0.73) | 0.76 (0.67) | 10.98 | **<.001** | .971 | **.028** |
| Restricted and repetitive behaviors (RRB) | 1.25 (0.23) | 3.33 (0.18) | 2.05 (0.11) | 31.01 | **<.001** | **<.001** | **.028** |
| Stereotyped/idiosyncratic use of words and phrases | 0.53 (0.61) | 1.25 (0.78) | 0.87 (0.77) | 16.43 | **<.001** | **<.001** | **.037** |
| Unusual sensory interest in play material/person | 0.11 (0.47) | 0.62 (0.81) | 0.28 (0.54) | 14.21 | **<.001** | **<.001** | .319 |
| Hand and finger and other complex mannerisms | 0.33 (0.59) | 0.70 (0.91) | 0.37 (0.69) | 7.83 | **<.001** | **<.029** | .956 |
| Excessive interest in or references to unusual or highly specific topics or objects or repetitive behaviors | 0.33 (0.63) | 0.85 (0.91) | 0.57 (0.77) | 7.61 | **<.001** | **.002** | .236 |
| Total (SA + RRB) | 11.11 (0.61) | 14.73 (0.43) | 9.34 (0.32) | 54.81 | **<.001** | **<.001** | .078 |

*ADOS-2* autism diagnostic observation scale–second edition, *NF1* neurofibromatosis type 1, *ASD* autism spectrum disorder

$P < 0.05$ considered significant are shown in bold

*communication* non-algorithm items; NF1 + ASD children were significantly less impaired in overall non-echoed spoken language, but more impaired when offering information. No differences were observed in any items on *social interaction, imagination/creativity* or *RRB* non-algorithm items.

No group differences were observed on items measuring other abnormal behaviors, such as overactivity/agitation, anxiety and tantrums, aggression, negative or disruptive behavior.

## Discussion

To our knowledge, this is the first study that has characterized the phenotypic profile of ASD symptomatology in NF1. Using convergent evidence of both the parent-rated SRS questionnaire and clinical interaction/observational assessments of the ADOS-2, we compared the NF1 + ASD group with two reference groups, namely autism and ASD from the original validation samples of ADOS-2. Although these two sub-types of idiopathic autism have now been combined within the current DSM 5 criteria (American Psychiatric Association 2013), this dual comparison is pragmatic and being based on currently available systematic data, is also useful in being able to make a comparison with different severities of idiopathic ASD.

Reviewing the algorithm data in the light of these comparisons we find ASD symptoms were evaluated as moderate in most assessed NF1 + ASD children (66 %). Generally the symptom profile shows similarity between NF1 and the ASD/autism groups. However overall differences are apparent in some areas; these include, comparatively in NF1, overall improved eye contact, less repetitive behaviors and better language skills.

Overall the results suggest that on the *SA* domains, NF1 + ASD children are just as impaired as the idiopathic autism group and more impaired compared to the ASD group. The NF1 + ASD group uses eye contact significantly more than either of the two comparison groups. Parent-reported SRS subscale scores confirm these

**Table 3** Group comparisons for NF1 versus Autism and NF1 versus ASD children on ADOS-2 non-algorithm item scores

| ADOS-2 non-algorithm items | Mean (*SD*) | | | *F* | *p* | Post-hoc pairwise comparisons | |
|---|---|---|---|---|---|---|---|
| | NF1 + ASD, $n = 36$ | Autism, $n = 129$ | ASD $n = 186$ | | | NF1/ Autism | NF1/ ASD |
| Language and communication | | | | | | | |
| Overall level of non-echoed spoken language | 0.19 (0.40) | 0.67 (0.58) | 0.55 (0.54) | 11.03 | **<.001** | **<.001** | **<.001** |
| Speech abnormalities associated with autism | 0.78 (0.64) | 1.23 (0.73) | 0.82 (0.69) | 14.48 | **<.001** | **.002** | .947 |
| Echolalia | 0.08 (0.37) | 0.35 (0.55) | 0.20 (0.47) | 5.73 | **.004** | **<.001** | .374 |
| Offers information | 1.06 (0.72) | 1.01 (0.77) | 0.46 (0.63) | 28.66 | **<.001** | .923 | **<.001** |
| Asks for information | 1.47 (0.74) | 1.88 (0.94) | 1.51 (0.96) | 6.68 | **.001** | .051 | .970 |
| Reciprocal social interaction | | | | | | | |
| Language production and linked nonverbal communication | 0.56 (0.50) | 0.88 (0.56) | 0.54 (0.55) | 15.35 | **<.001** | **.006** | .978 |
| Comment's on others emotions/empathy | 1.31 (0.82) | 1.95 (0.90) | 1.52 (0.93) | 11.45 | **<.001** | **<.001** | .412 |
| Insight into typical social situations and relationships | 1.61 (0.90) | 2.13 (0.86) | 1.59 (0.90) | 15.00 | **<.001** | **.005** | .992 |
| Imagination | | | | | | | |
| Imagination/creativity | 1.31 (0.71) | 1.18 (0.72) | 0.85 (0.67) | 12.19 | **<.001** | .580 | **<.001** |
| Stereotyped behaviors and restricted interests | | | | | | | |
| Self-injurious behaviour | 0.00 (0.00) | 0.02 (0.14) | 0.03 (0.16) | 0.70 | .495 | – | – |
| Compulsions/rituals | 0.06 (0.23) | 0.51 (0.71) | 0.21 (0.47) | 15.03 | **<.002** | **<.001** | .299 |
| Other abnormal behaviours | | | | | | | |
| Overactivity/agitation | 0.56 (0.74) | 0.57 (0.69) | 0.61 (1.21) | .079 | .924 | – | – |
| Tantrums, aggression, negative or disruptive behavior | 0.11 (0.47) | 0.29 (0.52) | 0.17 (0.48) | 3.03 | **.049** | .13 | .783 |
| Anxiety | 0.36 (0.64) | 0.18 (0.42) | 0.28 (0.53) | 2.42 | .091 | – | – |

*ADOS-2* autism diagnostic observation scale–second edition, *NF1* neurofibromatosis type 1, *ASD* autism spectrum disorder

$P < 0.05$ considered significant are shown in bold

observed difficulties in communication and interaction; >90 % have clinically relevant difficulties in interpreting social cues in interactions (cognition), while also having troubles with social communication. Also, social motivation was impaired in about 80 %. Sixty percent had problems in picking up on social cues (awareness).

Our results also show that using the ADOS, the NF1 + ASD group is statistically less likely to score on RRBs as compared to the autism group, however having comparable scores to the ASD group. Curiously though, parent ratings on the autistic mannerisms subscales of the SRS questionnaire were very high with two-third of NF1 + ASD children scoring in the severe problem range. Other studies which have used the SRS questionnaire to measure autistic symptomatology in the NF1 population have similarly found elevated scores on the 'autistic mannerisms' domain This subscale measures problems with flexibility, repetitive behaviors and being regarded by others as 'odd'. This apparent discrepancy between parent-rated (SRS) and clinician-rated child observational measures (ADOS) suggests that the RRBs are qualitatively different or not as severe as in idiopathic autism such that they are not being picked up within the ADOS assessment context. The ADOS is a short clinical interaction/observational measure lasting 30–40 min which may not be enough to observe repetitive behaviors (Rudacille 2011). The NF1 + ASD sample was significantly older than the reference groups. But this is unlikely to offer a plausible explanation for the fewer observed RRBs as there is good reported stability of core ASD traits in children across diverse ages. Indeed, Gotham et al. (2012) have shown the relative stability of ADOS standardized scores over 8–12 years. Or if RRBs are truly absent, does the new DSM 5 diagnosis of 'social pragmatic communication disorder' more accurately describe the social impairment in the NF1 group? (American Psychiatric Association 2013).

On non-algorithm ADOS domains of imagination/creativity and other abnormal behaviors, some specific findings should be underlined. On the items measuring *imagination/creativity*, NF1 + ASD children are just as impaired as the autism group and significantly more impaired than ASD children. Additionally, no group differences were found in

item measuring *overactivity*, which is an interesting finding since ADHD prevalence estimates in NF1 range from 30–67 %. This was perhaps unsurprising since high levels of comorbidity with ADHD are also observed in idiopathic ASD (Simonoff et al. 2008). There was also no evidence of group differences in the item measuring *anxiety* which is also a feature of some genetic disorders such as fragile X (Rogers et al. 2001) and Cornelia de Lange syndrome (Moss et al. 2012).

Based on our study, it is possible that with fewer observed RRB's and better eye contact these children do not present in clinic as idiopathic ASD and hence more likely to be overlooked by clinicians. Further research should look into detail at social cognition in NF1 including, theory of mind and affect recognition and explore RRBs using detailed parental interview such as the Autism Diagnostic Inventory-Revised (ADI-R) and observational measures. Understanding differences in children with NF1 + ASD and NF1 without ASD will also be important in furthering our understanding of the neurobiology of autism spectrum disorders.

Phenotypic profiles of syndromic ASD have been reported in other genetic disorders such as Fragile X, Williams syndrome, Tuberous Sclerosis, Down syndrome, Angelman syndrome and Rett syndrome. Specificity in profiles is often striking and intriguing in relation to our findings in NF1. For example, Fragile X presents with gaze aversion, stereotypic and repetitive speech, Williams syndrome presents with poor shared enjoyment in interaction and impaired non-verbal communication (Tordjman et al. 2012), excessive sociability in Angelman's syndrome (Oliver et al. 2007). Cross syndrome comparison is a promising approach to understanding the neurobiology of ASD and development of targeted interventions. This sort of comparative phenotyping has been used by Bruining et al. (2014) to stratify idiopathic ASD cases according to behavioural signature patterns associated with genetic disorders.

These findings should be considered in light of the study's limitations. Sample size was relatively small and future studies with larger sample sizes are needed to replicate these findings. We have used the autism and ASD normative samples reported in the ADOS-2 manual as a comparison groups. These normative children were closely matched in terms of VIQ but were significantly younger than the NF1 sample. Only a parent-rated questionnaire was used in the study rather than a clinician-rated interview like the Autism Diagnostic Interview-Revised (ADI-R), which along with the ADOS is considered gold standard in the assessment of ASD and was not available on all cases. Finally the participant data were gathered from two different centers with some identified differences as elaborated earlier.

## Conclusion

Behavioural phenotyping in genetic syndromes is a promising approach to illuminating the pathogenesis for ASD. NF1 is an important single gene disorder model for studying ASD with well-described neurobiology and the exciting possibility of interventions capable of reversing the phenotype. Within this context, our study sheds light on the profile of ASD in NF1 thus continuing the pursuit of a biological underpinning of the disorder. Using two standardized and well-validated ASD instruments, the findings support previous suggestions of subtle but significant overall differences in ASD symptomatology between NF1 and polygenic autism. Results highlight that the use of clinical cut-off scores and total scores alone may mask more subtle, but potentially significant, differences in the precise nature of ASD symptomatology in different genetic syndromes.

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