

Article

Sex-Based Asymmetry in the Association between Challenging Behaviours and Five Anxiety Disorders in Autistic Youth

Vicki Bitsika, Christopher F. Sharpley * , Kirstan A. Vessey and Ian D. Evans

Brain-Behaviour Research Group, University of New England, Armidale, NSW 2350, Australia; vickibitsika@une.edu.au (V.B.); kvessey@une.edu.au (K.A.V.); ievands3@une.edu.au (I.D.E.)

* Correspondence: csharp13@une.edu.au

Abstract: The presence of sex-based asymmetry in the behaviours of youths with Autism Spectrum Disorder (ASD) is currently under research scrutiny. ASD is characterised by challenging behaviour (CB) and is often accompanied by anxiety, both of which often exacerbate social interaction difficulties. The present study examined the presence of sex-based asymmetry in the *prevalence* of CB and anxiety and in the *association* between CB and anxiety in a sample including 32 male autistic youths (M age = 10.09, SD = 3.83, range = 6–18 yr) and 32 female autistic youths (M age = 10.31, SD = 2.57, range = 6–15 yr) matched for age, IQ, and ASD severity ($p > .101$). While the prevalence and severity of behavioural characteristics across males and females with ASD were similar ($p = .767$), representing symmetry, there was asymmetry in the ways that CBs and anxiety were associated with each other across the two sexes. Specifically, there were 3 instances of symmetry ($r > .3, p < .05$), but there were also 10 occurrences of sex-based asymmetry ($r < .3, p > .05$) in the association between five aspects of CB and five anxiety disorders. These findings emphasise the underlying sex-based *symmetry* in the prevalence of ASD-related behaviours, also highlighting unique sex-based *asymmetry* in the association between CBs and anxiety in autistic youths.

Keywords: asymmetry; autism; sex; challenging behaviour; anxiety



Citation: Bitsika, V.; Sharpley, C.F.; Vessey, K.A.; Evans, I.D. Sex-Based Asymmetry in the Association between Challenging Behaviours and Five Anxiety Disorders in Autistic Youth. *Symmetry* **2024**, *16*, 591. <https://doi.org/10.3390/sym16050591>

Academic Editor: Meimei Xia

Received: 7 April 2024

Revised: 28 April 2024

Accepted: 6 May 2024

Published: 10 May 2024



Copyright: © 2024 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

1. Introduction

It is well established that anxiety is often comorbid with the features of Autism Spectrum Disorder (ASD) in autistic youths [1–3] and that the presence of anxiety has negative implications for a young autistic person’s academic and social progress [4]. Additionally, anxiety can precede minor illnesses [5], interpersonal issues such as demoralisation, hostility, and mistrust [6], as well as an elevated risk of suicide [7,8]. An investigation of the possible correlates (sometimes referred to as “contributors”) of this anxiety is thus a high research priority because it may lead to advances in diagnosis and treatment protocols.

Some of the major possible correlates of anxiety in autistic youths are the characteristics of ASD themselves [9]. One of these is the “challenging” behaviour (CB) that is sometimes exhibited by autistic children [10]. CBs include children’s extreme distress in reaction to changes in their environment, adverse responses to sounds and textures, and behavioural outbursts such as hitting, kicking, screaming, breaking objects, running away, and extreme irritability [10]. These have been observed to cause elevated stress in the parents of autistic children [11–13], but relatively little attention has been given to how these behaviours are associated with the anxiety states of the autistic children themselves.

As suggested above, CBs may be conceptualised as *contributors* to anxiety because repeated displays of CBs can result in greater social stigma and social isolation compared to most neurotypical adolescents, or they may be conceptualised as the *results* of anxiety because they may be manifestations of anxiety that arise from social interactions. It is probable that both of these models of CB–anxiety relationships are credible when observed in anxious autistic youths, and the exact definition of a CB as a “cause” or a “result”

of anxiety is ethically difficult to determine. Instead, it may be sufficient to set aside the causal chain of events and simply define CBs and anxiety as “correlates” for the purpose of reflecting upon possible treatment approaches, thereby arguing that they may act in different directions within different persons, at different times, and under different circumstances. Thus, possible treatment approaches would consider the “CB–anxiety” association as a target itself rather than isolating either CB or anxiety as the consistent initiator of the other.

Two issues arise when investigating the CB–anxiety association: first, the way that CBs and anxiety are defined and measured, and second, the possible sex differences that may exist in the way that CBs and anxiety are associated. The first of these issues is important for psychometric and clinical treatment reasons wherein the choice of validated and reliable instruments for measuring CBs and anxiety is a basic requirement of sound methodology. Several major reviews of CB measurement have been reported (e.g., [14–16]), and a number of standardised valid and reliable instruments have been identified [17]. Similarly, several valid and reliable instruments for assessing anxiety in autistic youths have been reviewed [18]. Relatedly, although the issue of which source of information about an autistic child’s anxiety state is appropriate has received some attention, with some data suggesting that an autistic child’s self-reports of anxiety are more valid when compared with a physiological indicator of chronic stress (i.e., cortisol) than those given by their carers [19], CBs exhibited by autistic children are most often measured by parents’ ratings of their children’s behaviours on standardised scales because parents are the primary caregivers and often spend the most time with their autistic children in home settings. Finally, in terms of measuring CBs and anxiety, although it is common practice to use singular indices of each of these variables derived from the total scores of inventories that represent the range of diagnostic criteria for each, this process can obscure differences in the association between the underlying aspects of CBs (e.g., subscales on a standardised inventory) and various anxiety disorders [10]. For example, in one of the commonly used measures of CBs (the Aberrant Behavior Checklist [20]), there are five subscales, each of which represents a different type of CB, including irritability, lethargy, stereotypy, hyperactivity, and inappropriate speech. Similarly, the Diagnostic and Statistical Manual of Mental Disorders, 5th Ed, Text Revision [10] describes a number of anxiety disorders, five of which (Generalised Anxiety Disorder, Specific Phobia, Panic Disorder, Social Anxiety, and Separation Anxiety) have been shown to vary in their association with at least one of the major diagnostic criteria for ASD [21]; these five anxiety disorders will be the focus of this investigation.

The second issue (sex differences) is relevant because of the increasing focus upon the differences in the way that ASD and its associated behaviours are manifested in autistic male and female youths. From a search of the literature (PubMed, Google Scholar, PsycInfo) conducted in February 2024, using the descriptors “autism”, “child”, “adolescent”, “Challenging Behavior”, and “anxiety”, we identified the most recent research studies on these topics that were relevant to the current investigation (e.g., [22–26]), in addition to the recent review of sex differences in diagnosis, imaging, autistic behaviour, and sex-dependent uniqueness that was reported by Napolitano, Schiavi et al. [27]. Those authors noted that males tended to show more repetitive behaviours and restricted interests than females during childhood and adolescence, but females may have been unnoticed due to their ability to “camouflage” their ASD-related behaviours. By contrast, other data confirm the absence of sex-based differences in anxiety among autistic children and adolescents [28]. However, the relative prevalence of repetitive and restricted behaviours and anxiety between autistic males and females was not the primary focus of this research. Instead, this study focussed on the possible sex-based differences in the *relationship between* one of the major features of ASD (CBs) and several anxiety disorders. Although asymmetry in cognition and behaviour has been noted between human males and females for some time [29], the presence of sex-based asymmetry in the association between ASD-related

behaviours and anxiety has only received limited attention [30], and this limitation includes the CB–anxiety association [27,31] investigated here.

Therefore, the present study focussed on the *association* between CBs and anxiety in young autistic males and females rather than the *presence* of sex-based differences in CBs or anxiety in an attempt to explore whether that association was symmetrical or asymmetrical between males and females and, if so, in what way. On the basis of previous research on the prevalence of CBs and anxiety among autistic male and female youths, it was hypothesised that (1) males would exhibit higher rates of CBs than females, but (2) there would be no significant sex-based differences in anxiety. Due to the lack of previous research on the association between CBs and anxiety, the null hypothesis of there being (3) no significant differences in the significance or direction of the association between CBs and anxiety across autistic male and female youths was set for testing. The total CB scores and subscales from a CB inventory were also examined in addition to five anxiety disorders in order to provide a more comprehensive description of any sex-based asymmetry in the associations between these CB and anxiety variables. Because this is a study in a series about school-age autistic youths, the sample was restricted to an age range of 6yr to 18yr with an IQ of at least 70 and attendance in a mainstream school. In the local clinical categorisation used where these children resided, they were characterised as “mildly impaired”.

2. Materials and Methods

2.1. Participants

A total of 32 autistic males and 32 autistic females (see Table 1 for age, IQ, and ADOS data) were recruited from local parent support groups on the Gold Coast, Queensland, Australia. There were no significant differences between the males and females in age, WASI-II IQ, or ADOS-2 score (all $p > .101$). Because of the presence of sex dysphoria in autistic children [32], these males and females were defined by their parents’ identifications of their children’s sexes at birth, plus the males and females themselves agreeing to participate in “a study about autism in males” or “a study about autism in females” (recruited separately). Eleven of the thirty-two females had reached menarche, but apart from age, there were no significant differences between these females and those who had not reached menarche in any of the defining variables (age, IQ, and ASD severity), the CASI anxiety disorders, or the ABC subscales. Serendipitously, all of the females’ parents and 28 of the males’ parents were mothers, perhaps reflecting the finding that mothers have the highest levels of parenting stress from caring for a child with ASD [33]. All of the autistic children had been diagnosed with ASD via clinical interviews with a paediatrician or psychiatrist and a clinical psychologist, which was later confirmed with the administration of the Autism Diagnostic and Observation Schedule-2nd edition (ADOS-2) [34] by a research-reliable assistant as part of the recruitment process for this study; all of the males and females had total ADOS-2 scores of 7 or more. These children’s Full Scale IQs were all 70+ based on the Wechsler Abbreviated Scale for Intelligence-2nd edition (WASI-II) [35], which was administered by the same research-reliable assistant during recruitment. None of the parents reported that these sons or daughters were currently taking medication for anxiety or that their autistic children had been formally diagnosed with anxiety. The autistic children were able to be classified as “mildly impaired” because of their IQs, ASD severity scores, their abilities to self-manage, and because they were all attending mainstream schools.

Table 1. Mean (SD), range data for age, WASI-2 Full Scale IQ, and ADOS-2 scores for 32 autistic females and 32 autistic males.

Sex	Age	IQ ¹	ADOS ²
Males	10.09 yr (3.82 yr)	95.78 (13.67)	12.41 (2.62)
	6–18 yr	74–125	7–17
Females	10.31 yr (2.57 yr)	99.97 (12.85)	11.25 (2.91)
	6–15 yr	79–128	7–17

¹ Wechsler Abbreviated Scale of Intelligence (2nd ed.); ² Autism Diagnostic Observation Schedule (2nd Ed.).

2.2. Instruments

2.2.1. Anxiety

The Child and Adolescent Symptom Inventory (CASI) was used [36,37]. The CASI is designed to assess children and adolescents on a range of DSM-based disorders. Previously applied in a sample of 103 autistic children [38] and another sample of 67 children with ASD [39], the CASI Test Manual describes satisfactory subscale psychometric data for Generalised Anxiety Disorder (GAD), Specific Phobia, Panic Disorder, Social Phobia, and Separation Anxiety for this population [36,37]. Participants self-rated the frequency of their responses to CASI items as 0 (never), 1 (sometimes), 2 (often), or 3 (very often), thus providing a measure of severity beyond that obtained from categorical assessment procedures.

2.2.2. Challenging Behaviour

The Aberrant Behavior Checklist (ABC) was used to assess challenging behaviour [20]. Parents completed their 58-item community version of the ABC about their autistic children. The ABC provides a total score, plus scores for five subscales (Irritability, Lethargy, Stereotypy, Hyperactivity, and Inappropriate Speech) relevant to ASD. All of these subscales have internal consistencies (Cronbach's alpha) between .77 and .94. Parents rated their children's CBs on a 4-point Likert scale for each ABC item (0 = "not at all", 1 = "slight", 2 = "moderate", and 3 = "severe") for the past four weeks. Because the number of ABC items used in these five subscales ranges from 4 to 16, mean subscale scores were used. Norms for 1893 autistic youths aged 6 to 12 years have been previously reported for the ABC [40].

2.3. Procedure

Parents and children were each given a suitable Information Statement and Consent Form. Parents were also given the ABC, and their children were given the five anxiety subscales described above. Parents and their children completed all of these scales with reference to the last four-week period for all participants in order to reduce the influence of possible history effects. Responses were checked for completion.

2.4. Statistical Analyses

SPSS 27 was used to obtain descriptive data and check for normality, although both a MANOVA [41] and Pearson's Product Moment Correlation [42] are relatively robust to the effects of non-normality. A MANOVA confirmed the absence of significant differences between the male and female data regarding age, the ADOS-2 score, and the WASI-II Full Scale score; in addition, the effect of sex on the ABC and CASI scales was determined by a MANOVA. Pearson correlation coefficients were used to test for significant associations between the males' and females' ages, IQs, and ASD severity and between the males' and females' ABC total and subscale scores and the five anxiety disorder scores from the CASI. Although it has been suggested that when sample sizes are restricted, the alpha level might be set at .10 instead of .05 [43], a more stringent criterion was set for the presence of meaningful effects here. Thus, for these correlational analyses to be considered meaningful, they must have reached at least $p < .05$ plus (at minimum) an effect size of $r \geq .3$ [44].

3. Results

Table 1 presents the descriptive data for the autistic males' and females' ABC total and subscale scores and their five CASI anxiety disorder scores. There was little evidence of skewness or kurtosis in all of these scores; additionally, an inspection of the histograms and the normal Q-Q plots plus the Kolmogorov–Smirnov test for normality indicated that there was no need to transform the data [41]. The internal consistencies for the ABC total score and subscales and the CASI anxiety disorders scale were between .81 and .92, all of which are acceptable values. There were no significant correlations between the autistic females' ages, IQs, or ASD severity and any of their ABC or CASI scores, nor between the males' IQs or ASD severity and their ABC or CASI scores, but the males' ages were significantly

inversely associated with their mean ABC Irritability ($r = -.500, p = .004$) and Hyperactivity ($r = -.561, p = .001$) scores. Therefore, in the following ANOVA and MANOVA, age was covaried out of the analysis.

Sex Effects

The ANOVA indicated that there was no significant difference in the ABC total scores between the autistic females and males ($F(1,63) = .088, p = .767, \eta^2 = .001$). The MANOVA on the ABC subscales' mean scores and the CASI anxiety disorder scores also indicated that there were no significant sex differences in any of these variables (all p values $> .056$). In addition to the data presented in Table 2, Figure 1 shows the means and standard errors for these data for the males and females, plus the results of the MANOVA, suggesting the presence of sex-based symmetry in severity for the ABC subscales (Figure 1A) and the five anxiety disorders (Figure 1B). These results refute hypothesis (1) but confirm hypothesis (2).

Table 2. Mean (SD) data for 32 autistic males' and 32 autistic females' ABC¹ total and subscale scores and five CASI² anxiety disorders.

	Total Score	ABC					CASI				
		Irritability	Lethargy	Stereotypy	Hyperactivity	Inappropriate Speech	GAD ³	Specific Phobia	Panic Disorder	Social Anxiety	Separation Anxiety
Males	49.62 (32.97)	0.85 (0.70)	0.78 (0.53)	0.65 (0.59)	0.91 (0.82)	1.08 (0.67)	9.17 (4.63)	1.40 (1.06)	0.67 (0.92)	4.73 (3.24)	6.80 (6.21)
Females	50.72 (28.05)	1.13 (0.66)	0.68 (0.55)	0.44 (0.53)	0.91 (0.57)	0.74 (0.63)	11.78 (5.79)	1.34 (1.15)	0.72 (0.96)	5.41 (4.14)	7.68 (6.32)

¹ Aberrant Behavior Checklist; ² Child and Adolescent Symptom Inventory; ³ Generalised Anxiety Disorder.

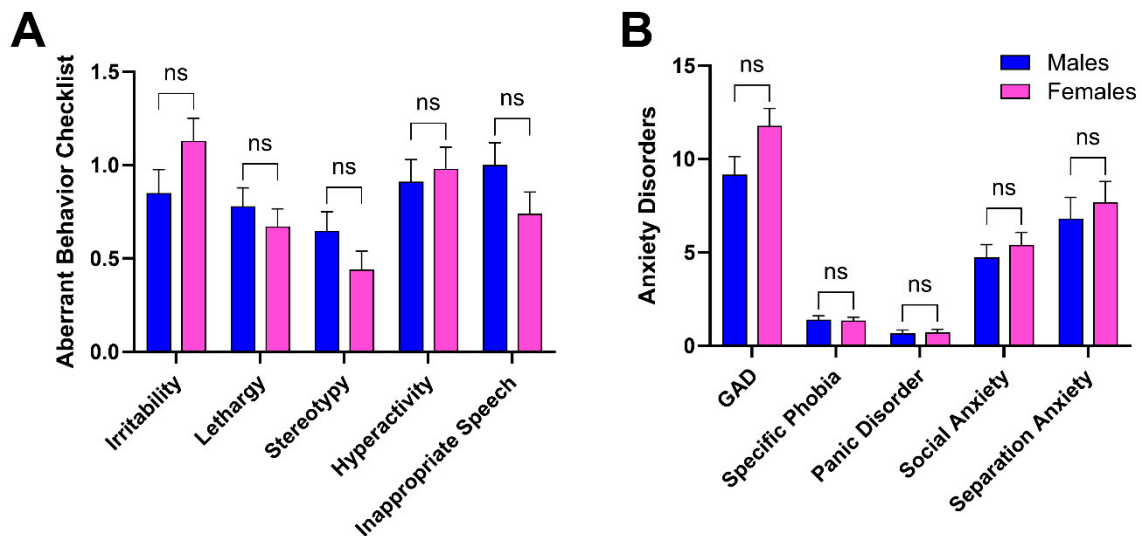


Figure 1. Mean scores and standard errors of five Aberrant Behavior Checklist subscales (A) and five anxiety disorders (B) for 32 young autistic males (blue) and 32 young autistic females (pink) with results of MANOVA univariate tests for sex-based differences.

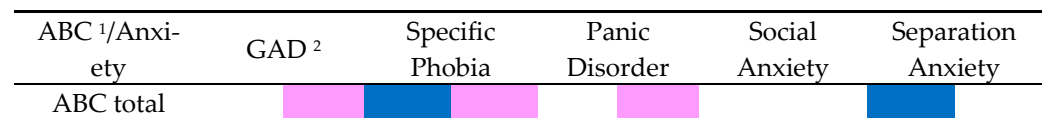
However, the symmetry between males and females regarding their severity of CBs and anxiety disorders did not necessarily imply that the same level of symmetry existed in terms of how the CB and anxiety disorder variables were associated with each other, which was the major focus of this investigation. To test the presence of symmetry in the associations between anxiety and CBs, a Pearson correlational analysis was performed separately on the males' and females' data. Table 3 shows the results of those correlations, and Figure 2 provides a heat map to portray the differences between the males' and females' CB-anxiety associations more clearly via the colour-coding of meaningful associations for males (blue squares) and females (pink squares) for each possible association. It is apparent in Figure 2 that the males' and females' associations between CBs and anxiety

were generally, but not consistently, asymmetrical, providing only partial confirmation of hypothesis (3).

Table 3. Pearson correlations * between Aberrant Behavior Checklist total and subscale scores and five anxiety disorders for 32 autistic males and 32 autistic females.

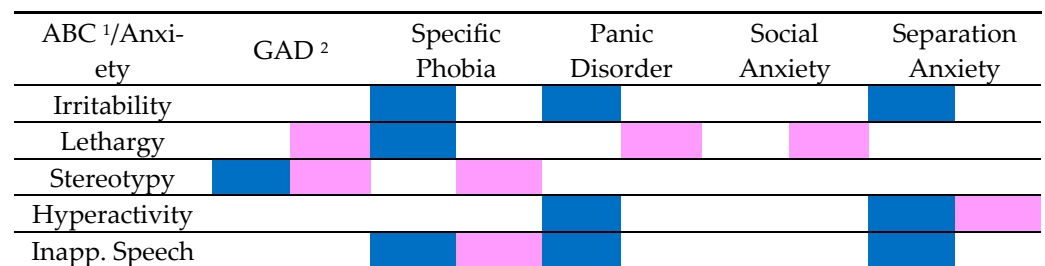
Males	GAD ¹	Specific Phobia	Panic Disorder	Social Anxiety	Separation Anxiety
ABC total	.128	.416 *	.331	.317	.367 *
Irritability	.234	.389 *	.387 *	.309	.417 *
Lethargy	.167	.392 *	.036	.200	.084
Stereotypy	.352 *	.350	.207	.303	.276
Hyperactivity	.249	.305	.364 *	.258	.369 *
Inappropriate Speech	.262	.355 *	.416 *	.334	.457 *
Females	GAD ¹	Specific Phobia	Panic Disorder	Social Anxiety	Separation Anxiety
ABC total	.424 *	.363 *	.393 *	.311	.261
Irritability	.249	.216	.294	.188	.168
Lethargy	.579 *	.241	.417 *	.395 *	.031
Stereotypy	.356 *	.445 *	.306	.066	.248
Hyperactivity	.248	.290	.275	.142	.391 *
Inappropriate Speech	.199	.534 *	.209	.277	.278

* $p < .05$ and $r > .300$; ¹ Generalised Anxiety Disorder.



¹ ABC= Aberrant Behavior Checklist; ² GAD = Generalised Anxiety Disorder.

(A)



¹ ABC= Aberrant Behavior Checklist; ² GAD = Generalised Anxiety Disorder.

(B)

Figure 2. (A) Comparison of meaningful correlations between Aberrant Behavior Checklist total score and five anxiety disorders for 32 autistic males (blue) and 32 autistic females (pink). (B) Comparison of meaningful correlations between Aberrant Behavior Checklist subscales and five anxiety disorders for 32 autistic males (blue) and 32 autistic females (pink).

As shown in Figure 2A, there was sex-based symmetry in the associations between the ABC total score and one of the five anxiety disorders (Specific Phobia), but there was sex-based asymmetry between the ABC total score and GAD, Panic Disorder, and Separation Anxiety. Although the lack of either sex having meaningful associations between the ABC total score and Social Anxiety could appear to be symmetrical, the focus of this investigation was on the presence of such meaningful associations rather than their absence.

When considered in terms of the ABC subscales (Figure 2B), there was sex-based symmetry between three different ABC subscale scores and three of the five anxiety disorders

(Stereotypy and GAD, Inappropriate Speech and Specific Phobia, and Hyperactivity and Separation Anxiety), but there was sex-based asymmetry between the ABC irritability subscale and Specific Phobia, Panic Disorder, and Separation Anxiety (only males had meaningful associations between these CBs and anxiety disorders); Lethargy and GAD, Panic Disorder, and Social Anxiety (females only) and Lethargy and Specific Phobia (males only); Stereotypy and Specific Phobia (females only); Hyperactivity and Panic Disorder (males only); and Inappropriate Speech and Panic Disorder and Separation Anxiety (males only). These instances of sex-based asymmetry between various ABC subscales and particular anxiety disorders were almost evenly spread between the males (seven meaningful associations) and females (six meaningful associations), emphasising the sex-based nature of these asymmetrical associations between CBs and anxiety disorders

4. Discussion

The major aim of this research was to investigate the presence of sex-based symmetry/asymmetry in the associations between five aspects of CB and five anxiety disorders. Although there were no significant differences between the males and females regarding the severity of CBs (rejecting hypothesis (1)) or anxiety (confirming hypothesis (2)) (indicating the presence of *symmetry*), the associations between CBs and anxiety indicated that there was inconsistent *asymmetry* (partially confirming hypothesis (3)). Figure 2 shows this sex-based asymmetry in associations between CBs and anxiety disorders most clearly, while Figure 1 demonstrates the sex-based symmetry in the severity of CBs and anxiety. Thus, both symmetry and asymmetry are present in these data, arguing for a more complex structure than what might initially be understood from simply examining only the severity or prevalence of these major features of ASD according to the sexes of respondents.

At the ABC total score level (i.e., the omnibus index of CBs), the presence of sex-based asymmetry in the associations between CBs and different anxiety disorders is clear, particularly with GAD and Panic Disorder for females, with Separation Anxiety for males, and with Specific Phobia for both males and females; there was no association between CBs and Social Anxiety for either sex. Although tentative at this stage, these results suggest that autistic males and females share common links between CBs and fear of specific objects or situations, but they differ in the way that CBs are associated with more generalised anxiety, the distinctive physiological manifestations that accompany panic, and the fear of being removed from important individuals. It appears that neither sex experiences a link between their CBs and fear of social situations, which is one of the more common anxiety disorders in autistic youths [45]. The “causal” roles of CBs and these anxiety disorders are yet to be defined in terms of directionality, but the finding that those roles vary according to sex and type of anxiety suggests that there is a more complex series of relationships, than has been suggested in the past.

When considered at the ABC subscale level, the sex–CB subtype–anxiety disorder associations become even more complex, displaying instances of symmetry and asymmetry. As described above, each of the five ABC subscales was associated with at least two of the five anxiety disorders for either males or females (asymmetry) or both (symmetry). It is noteworthy that none of the five ABC subscales were consistently associated with all the anxiety disorders for neither males nor females, further emphasising the asymmetry in these associations. This sex-based asymmetry shown at the ABC subscale and anxiety disorder levels of the CB–anxiety association also confirms the invalidity of using global CBs or anxiety as measures of how these two aspects of the ASD diagnostic criteria are related. Instead, these results affirm the need to consider sex and the particular types of CBs and anxiety when attempting to diagnose that relationship and plan for its treatment in autistic youths.

These findings are congruent with the fact that, although traditional investigations of these variables in the ASD population have been almost completely restricted to males, more recent research has reported some major differences in the ways that core ASD features manifest (i.e., their severity) in young autistic males and females [22–26]. However,

none of those studies, or any of the nearly 30 studies of sex differences in ASD that were reviewed by Napolitano Schiavi et al. [27], examined the nature of the associations between ASD features and different forms of anxiety across sexes. Vasa, Keefer et al. [1] reviewed the comorbidity of anxiety and some aspects of CBs, but they did not examine sex differences in those associations. While research which reports solely on the prevalence or severity of ASD features is valuable in understanding how autistic males and females may differ in the way that these features are demonstrated, it does not provide any insights into the ways that these ASD features may correlate with the very common comorbid condition of anxiety that has been found to occur in over 60% of autistic youth samples [1], nor how those features of ASD might be differently associated with different forms of anxiety across males and females. Thus, the issue of sex-based symmetry vs. asymmetry in the associations between CB and anxiety has not been the specific target of previous research.

Further, although it is most often reported as a unitary variable, “anxiety” occurs in several major forms, some of which are included in this study, and each of which is assessed via different symptoms, with only a minor overlap across these disorders that differ from each other in terms of the objects or situations that induce anxiety [10]. Of interest, the commonly quoted statement that there is a 2:1 ratio of anxiety occurrence in girls/boys in the general population [10] was not found here for any of the five anxiety disorders examined nor in some previous studies of anxiety prevalence in autistic males and females [28], which is possibly due to the elevated prevalence of anxiety in both male and female autistic youths compared to their non-autistic peers [46].

Therefore, by examining the association between these different forms of anxiety and five forms of CB, and by conducting examinations across autistic male and female youth, this study has enabled a more complex understanding of the way that one aspect of ASD is associated with the commonly comorbid disorder of anxiety rather than the understanding that can be derived from a simple global score for CBs and anxiety. The delineation of different associations between the five aspects of CB and the five anxiety disorders for males and females that is shown in Figure 2 provides a further basis for the development of individualised treatment planning, a goal that has been set for some time [47] but has yet to be realised [48].

Possible explanations of why ASD-related features may differ between autistic males and females (and perhaps why CBs and anxiety are differently associated for males and females) have been argued to rely, to a very large extent, upon differences in the ways that autistic male and female brains process information [49,50]. Although some attention was given to the “extreme male brain” hypothesis [51], that model has largely been refuted by fMRI data [52] as well as experimental data regarding testosterone concentrations [53]. Alternatively, more recent hypotheses have argued that autistic females may “camouflage” their ASD-related behaviours [54], although this was not found to be the case here as regards the severity of ABC subscale data across the sexes. It has been suggested that “sex-related biology may interact with peripheral processes, in particular the stress axis and brain arousal system, to produce distinct neurodevelopmental patterns in males and females with ASD” ([55], p. 1), and this hypothesis may be of use in explaining the sex-based asymmetry found here for the ways that CBs and anxiety disorders were associated. For example, Calderoni [56] has argued that neurological differences may occur in autistic males and females from in utero stages of development and continue during life, and Walsh, Pagni et al. [57] found different neural connectivity patterns between autistic male and female adults, arguing that these may account for differences in the male/female presentation of ASD features. However, while these are potentially invaluable in accounting for the sex-based differences in ASD feature presentation per se, they do not directly consider the underlying associations between specific aspects of CBs and particular anxiety disorders, leaving this field open to further investigation. The present study is a first step in that process and emphasises the presence of sex-based asymmetry in the ways that CBs and anxiety are associated in autistic youths. Further steps to investigate these asymmetrical sex-

based associations between CBs and anxiety might benefit from focusing on neurological differences between autistic males and females.

Although not directly comparable to some previous studies due to differences in age and specific neuropsychiatric disorders, these results add to the understanding of how sex differences may occur across a range of these disorders, including depression [58], anxiety [59], and Alzheimer's [60]. It has been argued that the sex differences found for many psychiatric disorders may be artifacts of a lack of sex-balanced participant recruitment [61,62], although there have been some suggestions that sex differences in some psychiatric disorders are the results of differences in brain development [63,64]. The current study directly addressed the issue of participant recruitment by matching males and females based on their ages, IQs, and ASD severity, thus adding some degree of validity to the comparison, which may not have been present in previous research. Future research into sex differences in brain functions while keeping participant recruitment constant will extend these findings.

This study is not without limitations, one of which was argued in the opening sections of this ms: no attempt was made here to derive causality in the association between CBs and anxiety. This would require a longitudinal study and major ethical considerations (i.e., how to initiate the kind of stressor experience that would lead to anxiety or CBs). Instead, the association between anxiety and CBs was the major research focus of this study, but further investigation (perhaps via observational studies) into whether CBs initiate anxiety, or vice versa, is a logical and valuable next step. The sample sizes in this study were limited, but the application of a more stringent criterion to obtain meaningful results for the correlational analysis argued for the validity of the results reported here. However, a further extension of this study with larger samples would enhance its generalisability. As stated, the autistic youths recruited here were only mildly impaired, and so no comment can be made regarding these findings and more severely impaired individuals with ASD, including the possible influence of comorbid intellectual impairment, ASD severity, ADHD, and chronic anxiety that may influence CB occurrence [65–67]. For reasons given in the Introduction, this study only recruited relatively high-functioning autistic youths, but the extension of this research to young autistic males and females with other psychiatric comorbidities is necessary before a confirmed model of the association between CBs and anxiety can be established. Although the focus of this study was on children and adolescents who were of school age, younger or older autistic males and females would be of interest in future research. There were some geographical and cultural limitations in sampling, and the fact that very few fathers volunteered for this study also limits this study's generalisability to them. Data were collected at a single point in time, and no examination was made of the possible changes that might occur in the association between CBs and anxiety over time, although age was not significantly correlated with CBs nor anxiety. In contrast to these limitations, the measures of anxiety and CBs used here are well established in the ASD research literature, and the identification of IQ and ASD severity was performed with gold standard instruments. The examination of CBs and anxiety from the perspective of different subscales and anxiety disorders contributes to the comprehensiveness of these findings.

5. Conclusions

These findings extend our understanding of the ways that males and females differ in their experiences of ASD features and comorbid forms of anxiety. The results indicating that there are different connections between specific CBs and various anxiety disorders across autistic male and female youths argue for a careful consideration of these differences when planning treatments for either CBs or anxiety in young people with ASD. Although this is a first step in such a process of developing individualised treatment options, the demonstration that such individualised models are necessary is of value.

Author Contributions: V.B. and C.F.S. designed the study and collected the data. C.F.S. analysed the data. K.A.V. and I.D.E. contributed to the data analysis and the writing of the ms in all drafts. C.F.S. completed the final draft. All authors have read and agreed to the published version of the manuscript.

Funding: This study received no funding.

Institutional Review Board Statement: This study was approved by the Bond University Human Research Ethics Committee, constituted according to the Helsinki Declaration of 1964 and recent amendments, approval no. RO1516.

Informed Consent Statement: Written informed consent to participate was provided by these parents for themselves and for their children, and the children themselves gave verbal assent.

Data Availability Statement: The raw data supporting the conclusions of this article will be made available by the authors upon request.

Conflicts of Interest: None of the authors have any conflicts of interest to declare.

References

1. Vasa, R.; Keefer, A.; McDonald, R.; Hunsche, M.; Kerns, C. A Scoping Review of Anxiety in Young Children with Autism Spectrum Disorder. *Autism Res.* **2020**, *13*, 2038–2057. [[CrossRef](#)] [[PubMed](#)]
2. White, S.W.; Oswald, D.; Ollendick, T.; Scahill, L. Anxiety in children and adolescents with autism spectrum disorders. *Clin. Psychol. Rev.* **2009**, *29*, 216–229. [[CrossRef](#)]
3. Settapani, C.A.; Puleo, C.M.; Conner, B.T.; Kendall, P.C. Characteristics and anxiety symptom presentation associated with autism spectrum traits in youth with anxiety disorders. *J. Anxiety Disord.* **2012**, *26*, 459–467. [[CrossRef](#)] [[PubMed](#)]
4. Ambrose, K.; Simpson, K.; Adams, D. The relationship between social and academic outcomes and anxiety for children and adolescents on the autism spectrum: A systematic review. *Clin. Psychol. Rev.* **2021**, *90*, 102086. [[CrossRef](#)]
5. Fries, E.; Hesse, J.; Hellhammer, J.; Hellhammer, D. A new view on hypocortisolism. *Psychoneuroendocrinology* **2005**, *30*, 1010–1016. [[CrossRef](#)]
6. Langewitz, W.; Ruddell, H. Spectral analysis of heart rate variability under mental stress. *J. Hypertens.* **1989**, *7*, 32–33. [[CrossRef](#)] [[PubMed](#)]
7. Malone, K.; Haas, G.; Sweeney, J.; Mann, J. Major depression and the risk of attempted suicide. *J. Affect. Disord.* **1995**, *34*, 173–185. [[CrossRef](#)]
8. Zimmerman, M.; McDermt, W.; Mattia, J. Frequency of anxiety disorders in psychiatric patients with Major Depressive Disorder. *Am. J. Psychiatry* **2000**, *157*, 1337–1340. [[CrossRef](#)]
9. Lau, B.Y.; Leong, R.; Uljarevic, M.; Lerh, J.W.; Rodgers, J.; Hollocks, M.J.; South, M.; McConachie, H.; Ozsivadjian, A.; Van Hecke, A.; et al. Anxiety in young people with autism spectrum disorder: Common and autism-related anxiety experiences and their associations with individual characteristics. *Autism* **2020**, *24*, 1111–1126. [[CrossRef](#)] [[PubMed](#)]
10. APA. *Diagnostic and Statistical Manual of Mental Disorders*, 5th ed.; Text Revision; American Psychiatric Association: Washington, DC, USA, 2022.
11. O’Nions, E.; Happé, F.; Evers, K.; Boonen, H.; Noens, I. How do Parents Manage Irritability, Challenging Behaviour, Non-Compliance and Anxiety in Children with Autism Spectrum Disorders? A Meta-Synthesis. *J. Autism Dev. Disord.* **2018**, *48*, 1272–1286. [[CrossRef](#)]
12. Estes, A.; Munson, G.; Dawson, E.; Koehler, E.; Zhou, X.-H.; Abbott, R. Parenting stress and psychological functioning among mothers of preschool children with autism and developmental delay. *Autism* **2009**, *13*, 357–387. [[CrossRef](#)] [[PubMed](#)]
13. Lecavalier, L.; Leone, S.; Wiltz, J. The impact if behavior problems on caregiver stress in young people with Autism Spectrum Disorders. *J. Intellect. Disabil. Res.* **2006**, *50*, 172–183. [[CrossRef](#)] [[PubMed](#)]
14. Carr, E.G.; Innis, J.; Blakeley-Smith, A.; Vasdev, S. Challenging behavior: Research design and measurement issues. In *The International Handbook of Applied Research in Intellectual Disabilities*; John Wiley & Sons Ltd.: Hoboken, NJ, USA, 2004; pp. 423–441.
15. Cohen, I.L.; Yoo, J.H.; Goodwin, M.S.; Moskowitz, L. Assessing challenging behaviors in Autism Spectrum Disorders: Prevalence, rating scales, and autonomic indicators. In *International Handbook of Autism and Pervasive Developmental Disorders*; Springer: New York, NY, USA, 2011; pp. 247–270.
16. Hanratty, J.; Livingstone, N.; Robalino, S.; Terwee, C.B.; Glod, M.; Oono, I.P.; Rodgers, J.; Macdonald, G.; McConachie, H. Systematic review of the measurement properties of tools used to measure behaviour problems in young children with autism. *PLoS ONE* **2015**, *10*, e0144649. [[CrossRef](#)] [[PubMed](#)]
17. Reyes-Martín, J.; Simó-Pinatella, D.; Font-Roura, J. Assessment of challenging behavior exhibited by people with intellectual and developmental disabilities: A systematic review. *Int. J. Environ. Res. Public Health* **2022**, *19*, 8701. [[CrossRef](#)] [[PubMed](#)]
18. Lecavalier, L.; Wood, J.; Halliday, A.; Jones, N.; Aman, M.; Cook, E. Measuring anxiety as a treatment endpoint in youth with Autism Spectrum Disorder. *J. Autism Dev. Disord.* **2014**, *44*, 1128–1143. [[CrossRef](#)]

19. Bitsika, V.; Sharpley, C.; Andronicos, N.; Agnew, L. Agreement between self- vs parent-ratings of General Anxiety Disorder symptoms and salivary cortisol in boys with an ASD. *J. Dev. Phys. Disabil.* **2015**, *27*, 467–477. [[CrossRef](#)]
20. Aman, M.; Singh, N.; Stewart, A.; Field, C. The Aberrant Behavior Checklist: A behavior ratings scale for the assessment of treatment effects. *Am. J. Ment. Defic.* **1985**, *89*, 485–491. [[PubMed](#)]
21. Bitsika, V.; Sharpley, C.; Mills, R. How are Sensory Features associated with seven anxiety disorders in boys with Autism Spectrum Disorder? *Int. J. Dev. Neurosci.* **2016**, *50*, 47–54. [[CrossRef](#)] [[PubMed](#)]
22. De Giambattista, C.; Ventura, P.; Trerotoli, P.; Margari, F.; Margari, L. Sex Differences in Autism Spectrum Disorder: Focus on High Functioning Children and Adolescents. *Front. Psychiatry* **2021**, *12*, 539835. [[CrossRef](#)]
23. Dillon, E.F.; Kanne, S.; Landa, R.J.; Annett, R.; Bernier, R.; Bradley, C.; Carpenter, L.; Kim, S.H.; Parish-Morris, J.; Schultz, R. Sex differences in autism: Examining intrinsic and extrinsic factors in children and adolescents enrolled in a national ASD cohort. *J. Autism Dev. Disord.* **2021**, *53*, 1305–1318. [[CrossRef](#)]
24. McFayden, T.C.; Putnam, O.; Grzadzinski, R.; Harrop, C. Sex differences in the developmental trajectories of autism spectrum disorder. *Curr. Dev. Disord. Rep.* **2023**, *10*, 80–91. [[CrossRef](#)]
25. Kaat, A.J.; Shui, A.M.; Ghods, S.S.; Farmer, C.A.; Esler, A.N.; Thurm, A.; Georgiades, S.; Kanne, S.M.; Lord, C.; Kim, Y.S. Sex differences in scores on standardized measures of autism symptoms: A multisite integrative data analysis. *J. Child Psychol. Psychiatry* **2021**, *62*, 97–106. [[CrossRef](#)] [[PubMed](#)]
26. Siracusano, M.; Postorino, V.; Riccioni, A.; Emberti Gialloreti, L.; Terribili, M.; Curatolo, P.; Mazzone, L. Sex differences in autism spectrum disorder: Repetitive behaviors and adaptive functioning. *Children* **2021**, *8*, 325. [[CrossRef](#)] [[PubMed](#)]
27. Napolitano, A.; Schiavi, S.; La Rosa, P.; Rossi-Espagnet, M.C.; Petrillo, S.; Tagliente, E.; Longo, D.; Valeri, G.; Piemonte, F.; Trezza, V. Sex differences in autism spectrum disorder: Diagnostic, neurobiological, and behavioral features. *Front. Psychiatry* **2022**, *13*, 889636. [[CrossRef](#)]
28. Ambrose, K.; Adams, D.; Simpson, K.; Keen, D. Exploring profiles of anxiety symptoms in male and female children on the autism spectrum. *Res. Autism Spectr. Disord.* **2020**, *76*, 101601. [[CrossRef](#)]
29. Gray, J.A.; Buffery, A.W.H. Sex differences in emotional and cognitive behaviour in mammals including man: Adaptive and neural bases. *Acta Psychol.* **1971**, *35*, 89–111. [[CrossRef](#)]
30. Hull, L.; Mandy, W.; Petrides, K. Behavioural and cognitive sex/ gender differences in autism spectrum condition and typically developing males and females. *Autism* **2017**, *21*, 706–727. [[CrossRef](#)] [[PubMed](#)]
31. Bölte, S.; Neufeld, J.; Marschik, P.B.; Williams, Z.J.; Gallagher, L.; Lai, M.-C. Sex and gender in neurodevelopmental conditions. *Nat. Rev. Neurol.* **2023**, *19*, 136–159. [[CrossRef](#)]
32. Glidden, D.; Bouman, W.; Jones, B.; Arcelus, J. Gender Dysphoria and Autism Spectrum Disorder: A Systematic Review of the Literature. *Sex. Med. Rev.* **2016**, *4*, 3–14. [[CrossRef](#)]
33. Dabrowska, A.; Pisula, E. Parenting stress and coping styles in mothers and fathers of pre-school children with autism and Down syndrome. *J. Intellect. Disabil. Res.* **2010**, *54*, 266–280. [[CrossRef](#)]
34. Lord, C.; Rutter, M.; DiLavore, P.; Risi, S.; Gotham, K.; Bishop, S. *Autism Diagnostic Observation Schedule*, 2nd ed.; (ADOS-2); Western Psychological Services: Los Angeles, CA, USA, 2012.
35. Wechsler, D. *The Wechsler Abbreviated Scale of Intelligence*, 2nd ed.; Pearson: Bloomington, MN, USA, 2011.
36. Gadow, K.; Sprafkin, J. *Child and Adolescent Symptom Inventory 4R: Screening and Norms Manual*; Checkmate Plus: Stony Brook, NY, USA, 2010.
37. Gadow, K.; Sprafkin, J.; Carlson, G.; Schneider, J.A.; Nolan, E.; Mattison, R.; Rundberg-Rivera, V. A DSM-IV-referenced adolescent self-reporting scale. *J. Am. Acad. Child Adolesc. Psychiatry* **2002**, *41*, 671–679. [[CrossRef](#)] [[PubMed](#)]
38. Gadow, K.; Devinent, C.; Pomeroy, J.; Azizian, A. Comparison of DSM-IV symptoms in elementary school-age children with PDD versus clinic and community samples. *Autism* **2005**, *9*, 392–415. [[CrossRef](#)] [[PubMed](#)]
39. Weisbrot, D.; Gadow, K.; DeVincent, C.; Pomeroy, J. The presentation of anxiety in children with Pervasive Developmental Disorders. *J. Child Adolesc. Psychopharmacol.* **2005**, *15*, 477–496. [[CrossRef](#)] [[PubMed](#)]
40. Kaat, A.; Lecavalier, L.; Aman, M. Validity of the Aberrant Behaviour Checklist in children with Autism Spectrum Disorder. *J. Autism Dev. Disord.* **2014**, *44*, 1103–1116. [[CrossRef](#)] [[PubMed](#)]
41. Tabachnik, B.; Fidell, L. *Using Multivariate Statistics*, 6th ed.; Pearson Education: Boston, MA, USA, 2013.
42. Cohen, J.; Cohen, P.; West, S.; Aiken, L.S. *Applied Multiple Regression/Correlation Analysis for the Behavioral Sciences*; Lawrence Erlbaum Associates: Mahwah, NJ, USA, 2003.
43. Stevens, J. *Applied Multivariate Statistics for the Social Sciences*, 4th ed.; Erlbaum: Hillsdale, NJ, USA, 2002.
44. Cohen, J. *Statistical Power for the Behavioural Sciences*; Erlbaum: Hillsdale, NJ, USA, 1988.
45. Bitsika, V.; Sharpley, C. Variation in the profile of Anxiety Disorders in boys with an ASD according to method and source of assessment. *J. Autism Dev. Disabil.* **2015**, *45*, 1825–1835. [[CrossRef](#)]
46. Hallett, V.; Ronald, A.; Colvert, E.; Ames, C.; Woodhouse, E.; Lietz, S.; Garnett, T.; Gillan, N.; Rijdsdijk, F.; Scahill, L.; et al. Exploring anxiety symptoms in a large-scale twin study of children with autism spectrum disorders, their co-twins and controls. *J. Child Psychol. Psychiatry* **2013**, *54*, 1176–1185. [[CrossRef](#)] [[PubMed](#)]
47. Stahmer, A.C.; Schreibman, L.; Cunningham, A.B. Toward a technology of treatment individualization for young children with autism spectrum disorders. *Brain Res.* **2011**, *1380*, 229–239. [[CrossRef](#)] [[PubMed](#)]

48. Klinger, L.G.; Cook, M.L.; Dudley, K.M. Predictors and Moderators of Treatment Efficacy in Children and Adolescents with Autism Spectrum Disorder. *J. Clin. Child Adolesc. Psychol.* **2021**, *50*, 517–524. [[CrossRef](#)] [[PubMed](#)]
49. Lai, M.-C.; Lerch, J.; Floris, D.; Ruigrok, A.; Pohl, A.; Lombardo, M.; Baron-Cohen, S. Imaging sex/gender and autism in the brain: Etiological implications. *J. Neurosci. Res.* **2017**, *95*, 380–397. [[CrossRef](#)]
50. Mottron, L.; Duret, P.; Mueller, S.; Moore, R.; d’Arc, B.; Jacquemont, S.; Xiong, L. Sex differences in brain plasticity: A new hypothesis for sex ratio bias in Autism. *Mol. Autism* **2015**, *6*, 33. [[CrossRef](#)]
51. Baron-Cohen, S.; Auyeung, B.; Norgaard-Pedersen, B.; Hougaard, D.; Abdullah, M.; Melgaard, L.; Cohen, A.; Chakrabarti, B.; Ruta, L.; Lombardo, M. Elevated fetal steroidogenic activity in autism. *Mol. Psychiatry* **2014**, *20*, 369–376. [[CrossRef](#)] [[PubMed](#)]
52. Alaerts, K.; Swinnen, S.P.; Wenderoth, N. Sex differences in autism: A resting-state fMRI investigation of functional brain connectivity in males and females. *Social Cognit. Affect. Neurosci.* **2016**, *11*, 1002–1016. [[CrossRef](#)] [[PubMed](#)]
53. Sharpley, C.F.; Bitsika, V.; Andronicos, N.; Agnew, L. Age-related variations in comparative testosterone concentrations between boys with autism spectrum disorder and their typically-developing peers: A challenge to the ‘extreme male brain’ hypothesis of ASD. *J. Dev. Phys. Disabil.* **2017**, *29*, 353–367. [[CrossRef](#)]
54. Hull, L.; Petrides, K.; Mandy, W. The Female Autism Phenotype and Camouflaging: A Narrative Review. *Rev. J. Autism Dev. Disord.* **2020**, *7*, 306–317. [[CrossRef](#)]
55. Walsh, M.J.M.; Wallace, G.L.; Gallegos, S.M.; Braden, B.B. Brain-based sex differences in autism spectrum disorder across the lifespan: A systematic review of structural MRI, fMRI, and DTI findings. *NeuroImage Clin.* **2021**, *31*, 102719. [[CrossRef](#)] [[PubMed](#)]
56. Calderoni, S. Sex/gender differences in children with autism spectrum disorder: A brief overview on epidemiology, symptom profile, and neuroanatomy. *J. Neurosci. Res.* **2023**, *101*, 739–750. [[CrossRef](#)] [[PubMed](#)]
57. Walsh, M.J.M.; Pagni, B.; Monahan, L.; Delaney, S.; Smith, C.J.; Baxter, L.; Braden, B.B. Sex-related brain connectivity correlates of compensation in adults with autism: Insights into female protection. *Cereb. Cortex* **2022**, *33*, 316–329. [[CrossRef](#)] [[PubMed](#)]
58. Eid, R.S.; Gobinath, A.R.; Galea, L.A. Sex differences in depression: Insights from clinical and preclinical studies. *Prog. Neurobiol.* **2019**, *176*, 86–102. [[CrossRef](#)]
59. Jalnapurkar, I.; Allen, M.; Pigott, T. Sex differences in anxiety disorders: A review. *J. Psychiatry Depress Anxiety* **2018**, *4*, 3–16. [[CrossRef](#)]
60. Irvine, K.; Laws, K.R.; Gale, T.M.; Kondel, T.K. Greater cognitive deterioration in women than men with Alzheimer’s disease: A meta analysis. *J. Clin. Exp. Neuropsychol.* **2012**, *34*, 989–998. [[CrossRef](#)]
61. Rechlin, R.K.; Splinter, T.F.L.; Hodges, T.E.; Albert, A.Y.; Galea, L.A.M. An analysis of neuroscience and psychiatry papers published from 2009 and 2019 outlines opportunities for increasing discovery of sex differences. *Nat. Commun.* **2022**, *13*, 2137. [[CrossRef](#)]
62. Farhane-Medina, N.Z.; Luque, B.; Taberner, C.; Castillo-Mayén, R. Factors associated with gender and sex differences in anxiety prevalence and comorbidity: A systematic review. *Sci. Prog.* **2022**, *105*, 00368504221135469. [[CrossRef](#)]
63. Knouse, M.C.; McGrath, A.G.; Deutschmann, A.U.; Rich, M.T.; Zallar, L.J.; Rajadhyaksha, A.M.; Briand, L.A. Sex differences in the medial prefrontal cortical glutamate system. *Biol. Sex Diff.* **2022**, *13*, 66. [[CrossRef](#)] [[PubMed](#)]
64. Szadvári, I.; Ostatníková, D.; Durdiaková, J.B. Sex differences matter: Males and females are equal but not the same. *Physiol. Behav.* **2023**, *259*, 114038. [[CrossRef](#)] [[PubMed](#)]
65. Edirisooriya, M.; Dykiert, D.; Auyeung, B. IQ and Internalising Symptoms in Adolescents with ASD. *J. Autism Dev. Disord.* **2021**, *51*, 3887–3907. [[CrossRef](#)] [[PubMed](#)]
66. Peña-Salazar, C.; Arrufat, F.; Santos López, J.-M.; Fontanet, A.; Roura-Poch, P.; Gil-Girbau, M.; Carbonell-Ducastella, C.; Serrano-Blanco, A. Intellectual disability, autism spectrum disorders, psychiatric comorbidities and their relationship with challenging behavior. *J. Ment. Health Res. Intellect. Disabil.* **2022**, *15*, 77–94. [[CrossRef](#)]
67. Kerns, C.M.; Winder-Patel, B.; Iosif, A.M.; Nordahl, C.W.; Heath, B.; Solomon, M.; Amaral, D.G. Clinically significant anxiety in children with autism spectrum disorder and varied intellectual functioning. *J. Clin. Child Adolesc. Psychol.* **2021**, *50*, 780–795. [[CrossRef](#)]

Disclaimer/Publisher’s Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.