Research article

Effectiveness of the gluten-free, casein-free diet for children diagnosed with autism spectrum disorder: Based on parental report

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Objectives: Studies on the gluten-free and/or casein-free (GFCF) dietary intervention for children with autism spectrum disorders (ASDs) suggest that some children may positively respond to implementation of the dietary intervention. Other research suggests that children diagnosed with ASD can be classified into subpopulations based on various factors, including gastrointestinal (GI) abnormalities and immune function.

Methods: This study analyzes parental report data collected using a 90-item online questionnaire from 387 parents or primary caregivers of children diagnosed with ASD on the efficacy of the GFCF diet. Parents reported on their child’s GI symptoms, food allergy diagnoses, and suspected food sensitivities, as well as the degree and length of their diet implementation.

Results: Overall, diet efficacy among children whose parents reported the presence of GI symptoms, food allergy diagnoses, and suspected food sensitivities included greater improvement in ASD behaviors, physiological symptoms, and social behaviors compared with children whose parents reported none of these symptoms, diagnoses, or sensitivities ($P < 0.05$). Parental report of strict diet implementation, indicated by complete gluten/casein elimination and infrequent diet errors during and outside of parental care, also corresponded to improvement in ASD behaviors, physiological symptoms, and social behaviors ($P < 0.05$).

Discussion: These findings suggest that various intricacies related to diet implementation and GI and immune factors may play a role in differentiating diet responders from diet non-responders and substantiate the importance of further investigations into the various, nuanced factors that influence efficacy of the intervention among children with ASDs.

Keywords: Autism spectrum disorder (ASD), Casein, Gluten, GFCF diet, Parental report

Introduction

Autism spectrum disorders (ASDs) are pervasive developmental disorders (PDDs) characterized and diagnosed by social interaction and communication impairment and restricted, stereotyped, and repetitive patterns of behavior and language. These disorders affect hundreds of thousands of children (an estimated 1 in 110 children) with an increasing incidence. To date, the physiological underpinnings of autism have not been fully elucidated. Similarly, treatments vary in their effectiveness for treating children with these disorders. The etiology of ASDs is complex, and a cure remains elusive. Among the various treatment modalities being evaluated for these individuals, studies on the gluten-free and/or casein-free (GFCF) dietary intervention suggest that some children with ASDs may positively respond to implementation of the diet. Among the various theories to explain the pathological action of gluten and casein within the ASD clinical population is the hypothesis that gluten- and casein-derived peptides trigger an immune response resulting in an excessive gastrointestinal (GI) inflammation. Specifically, researchers have found that children with autism had higher proinflammatory cytokines following challenge with food proteins from gluten, casein, and soy, compared with normal controls and non-autistic children with known symptomatic food allergies. In addition to resolved GI symptoms, parents often report improvements in speech and communication skills, decreases in hyperactive behavior, improvements in their child’s ability to focus, and decreases in sleep problems following implementation of the diet.

Other research suggests that children diagnosed with ASDs can be classified into subpopulations based on various factors, including GI abnormalities and immune function. Some children may positively respond to the GFCF diet, while others may not. This study analyzes parental report data collected using a 90-item online questionnaire from 387 parents or primary caregivers of children diagnosed with ASD on the efficacy of the GFCF diet. Parents reported on their child’s GI symptoms, food allergy diagnoses, and suspected food sensitivities, as well as the degree and length of their diet implementation.

Overall, diet efficacy among children whose parents reported the presence of GI symptoms, food allergy diagnoses, and suspected food sensitivities included greater improvement in ASD behaviors, physiological symptoms, and social behaviors compared with children whose parents reported none of these symptoms, diagnoses, or sensitivities ($P < 0.05$). Parental report of strict diet implementation, indicated by complete gluten/casein elimination and infrequent diet errors during and outside of parental care, also corresponded to improvement in ASD behaviors, physiological symptoms, and social behaviors ($P < 0.05$).

These findings suggest that various intricacies related to diet implementation and GI and immune factors may play a role in differentiating diet responders from diet non-responders and substantiate the importance of further investigations into the various, nuanced factors that influence efficacy of the intervention among children with ASDs.

Keywords: Autism spectrum disorder (ASD), Casein, Gluten, GFCF diet, Parental report
immune function. It is not yet known which subpopulations respond best to the GFCF dietary intervention. Similarly, there have been no studies to date investigating factors related to diet implementation that play a role in the degree of the intervention’s effectiveness. Dietary non-compliance to gluten elimination is known to affect GI outcomes in patients with celiac disease. However, a randomized, controlled study of the dietary intervention noted diet errors by participants taking part in the study. Children with ASD who also have GI and allergy symptoms may be similarly affected by poor diet compliance (i.e. gluten and/or casein elimination). However, no studies to date have analyzed whether diet compliance is a factor in degree of diet effectiveness among children with ASDs.

The present study was designed to examine ASD subpopulations and diet implementation factors related to the GFCF diet. Specifically, parents or primary caregivers of children with ASD were asked to complete a 90-item online questionnaire about their child’s GI symptoms, food allergy diagnoses, and suspected food sensitivities, as well as GFCF diet trial compliance and length in order to examine which subpopulation and implementation factors correlate with diet responsiveness. Based on prior research studies investigating the GFCF diet and GI symptoms common in the pediatric autistic population, we hypothesized that parental report of effectiveness of this diet would differ by degree of diet compliance during implementation as well as by the child’s history of physiological and allergy symptoms prior to the start of the diet. Specifically, we hypothesized that the diet would be more effective for children whose parents strictly eliminated all gluten and casein, compared with those children whose diet was less strict. We also hypothesized that the diet would be more effective for children with a history of physiological and/or allergy symptoms as well as food allergies and/or suspected food sensitivities, compared to those children without these physiological and/or allergy symptoms and food sensitivities.

**Methods**

**Participants and study sample**

Potential participants included parents or primary caregivers of children diagnosed with ASDs and were recruited via e-mail listservs, various autism organization websites (e.g. message boards), and by word-of-mouth. There were no exclusion criteria. Participants were parents or primary caregivers of children with a clinical diagnosis of an ASD. Implied consent was given by all study participants; all procedures were reviewed and approved by The Pennsylvania State University Institutional Review Board. Four hundred and forty-eight participants attempted the survey (described below). Eighty-six percent (N = 387) of the participants provided complete survey responses and their data were included in the following analyses.

Diagnoses among the study sample of participants’ ASD children were as follows: 49.4% autism (N = 191), 16.8% high-functioning autism (N = 65), 15.8% Asperger syndrome (N = 61), 4.9% PDD (N = 19), 28.4% pervasive developmental disorder-not otherwise specified (PDD-NOS) (N = 110), 0.3% Rett syndrome (N = 1), and 0.3% childhood disintegrative disorder (N = 1). Of the ASD children whose parents were surveyed, 82% were male (N = 315) and 18% were female (N = 69). Three parents chose not to provide the gender of their children. The majority (88.9%) of ASD children in the survey sample were described by their parent or primary caregiver as Caucasian (N = 330), 1.6% as African American (N = 6), 1.9% as Asian (N = 7), 3.2% as Hispanic (N = 12), 0.3% as Native Hawaiian/other Pacific Islander (N = 1), and 4% as mixed racial, bi-racial, or ‘other’ (N = 15).

**Procedures**

Eligible parents/primary caregivers had the opportunity to complete an online survey for all of their children who were diagnosed with an ASD. If desired, participants were able to leave and return to the survey from the same computer over the course of the day. Data were collected over a 5-month period of time in 2008.

**Instrument**

A 90-item online questionnaire was developed and administered via Survey Monkey, a widely used service for online research that offers a high level of encryption for data safety. The survey included questions regarding various demographics about the participants’ autistic child(ren), the child(ren)’s diagnoses, parental familiarity with the GFCF dietary treatment, parental belief in the GFCF diet, parental implementation of the GFCF diet, parental report of effectiveness of the GFCF diet, observable changes in various autism-related symptoms and behaviors, as well as parental perception of the sustainability of the GFCF diet as a long-term treatment option.

**Assessment of diet implementation**

Measures of diet implementation included variables regarding diet compliance and length of diet implementation. Diet compliance was assessed using the following measures: diet strictness and frequency of diet errors (during times when child is under parental care and during times when child is outside of parental care). Length of diet implementation was represented by the length of the longest diet implementation trial.
Assessment of diet effectiveness

Measures of diet effectiveness included: the number of improved characteristic ASD behaviors (i.e. decreases in self-stimulatory behaviors, hyperactivity, sensory seeking behaviors, temper tantrums, the lining up of objects, and echolalia), physiological symptoms (i.e. decreases in bodily rash, red ring around the anus, constipation, diarrhea, and seizures), and social behaviors (i.e. increases in social responsiveness, eye contact, engagement, attention span, requesting behavior, commenting on (or naming) objects, pointing, language production, sign language production (number of signs), and imaginative play) that are typically delayed or abnormal in children with ASD.

Data analyses

Following data collection, data were cleaned and checked for completeness using Excel. Separate, one-way analyses of variances were used to examine group differences. Tukey’s B post hoc analyses were conducted to further investigate statistically significant group differences when appropriate. All tests were two-tailed, and statistical significance was accepted at alpha <0.05.

Results

Degree of diet compliance

Gluten and/or casein elimination

Participants were grouped into two categories based on their compliance during implementation of the GF and/or CF diet. Specifically, parents (N = 293) were categorized into two groups: (1) those who completely eliminated all food containing gluten and/or casein from their child’s diet (N = 223) and (2) those who did not completely eliminate all foods containing gluten and/or casein (i.e. some foods containing gluten and/or casein were not eliminated from the child’s diet) (N = 70).

Parents who eliminated all GF and/or CF foods reported that a greater number of their children’s ASD behaviors, physiological symptoms, and social behaviors improved after starting the diet compared to children whose parents did not eliminate all GF and/or CF foods (F(1,291) = 12.92, P < 0.05, F(1,290) = 4.36, P < 0.05, and F(1,291) = 19.66, P < 0.05, respectively) (see Fig. 1).

Diet errors in parental implementation

Participants were grouped into four categories based on the number of times their child broke the diet during their care. Specifically, parents (N = 271) were categorized into four groups: (1) those who never broke the diet (N = 77), (2) those who broke the diet two or fewer times per year (N = 62), (3) those who broke the diet about every other month (N = 40), and (4) those who broke the diet at least once a month (N = 92).

Frequency of diet errors during parental care influenced ASD behaviors, but not digestive symptoms or social behaviors (see Fig. 2) (F(3,267) = 4.45, P < 0.05, F(3,266) = 1.55, n.s., and F(3,267) = 1.60, n.s., respectively). Specifically, parents who broke the diet at least once per month and those who broke the diet about every other month reported fewer ASD behavior improvements than did those parents who broke the diet two or fewer times a year (Tukey’s B, P < 0.05). Unexpectedly, parents who reported never breaking the diet did not report an improvement in ASD behavior.
Diet errors outside parental care

Participants were grouped into four categories based on the number of times their child broke the diet outside their care. Specifically, parents (N = 261) were categorized into four groups: (1) those who reported that the diet was never broken outside their care (N = 63), (2) those who reported that the diet was broken two or fewer times per year outside their care (N = 23), (3) those who reported that the diet was broken about every other month outside their care (N = 82), and (4) those who reported that the diet was broken at least once a month outside their care (N = 93).

The effectiveness of the diet was also influenced by the frequency of diet errors outside of parental care, but not in the expected direction. Frequency of diet errors outside of parental care influenced ASD behaviors and physiological symptoms, but not social behaviors (F(3,257) = 2.66, P < 0.05, F(3,256) = 2.75, P < 0.05, F(3,257) = 2.34, n.s., respectively). Specifically, ASD behaviors and physiological symptoms, but not social behaviors, improved among children whose parents reported that the diet was broken two or fewer times a year compared to children whose parents reported that the diet was never broken outside of their care (Tukey’s B, P < 0.05) (see Fig. 3).

Length of diet implementation

Parents also were categorized into four groups based on length of diet implementation: (1) parents who implemented the diet for less than 6 months (N = 77), (2) parents who implemented the diet for more than 6 months but less than 1 year (N = 42), (3) parents who implemented the diet for 1 year (N = 51), and (4) parents who implemented the diet for 2 or more years (N = 96). Parents who implemented the diet for 6 months or less reported that the diet was less effective in reducing their child’s ASD behaviors (F(3,262) = 12.08, P < 0.01), physiological symptoms (F(3,261) = 9.88, P < 0.01), and social behaviors (F(3,262) = 6.78, P < 0.01), compared to all other groups (i.e. parents who implemented the diet for more than six months) (see Fig. 4).

Physical symptoms and reported diet effectiveness

In addition to parental reports of diet effectiveness, parents also reported the presence of several physical symptoms (i.e. GI symptoms, allergies, food sensitivities). GI symptoms included diarrhea and constipation, and food allergy symptoms included skin problems, red cheeks, red ears, rash, and/or red ring around mouth and/or anus, hives, dark circles under the eyes, sneezing, stuffy nose, and itchy, watery, red eyes. Of the 387 children represented in the study pool, 92.7% (N = 280/302 respondents) had GI symptoms (specifically, constipation and/or diarrhea), 93.4% (N = 312/334 respondents) had allergy symptoms, 68.3% (N = 149/218 respondents) had at least one diagnosed food allergy, and 61.3% (N = 204/333 respondents) had at least one suspected food sensitivity.

The diet was effective in improving ASD behaviors (F(1,300) = 9.77, P < 0.05), physiological symptoms (F(1,300) = 21.44, P < 0.05), and social behaviors (F(1,300) = 8.54, P < 0.05), for those children with GI symptoms (specifically, constipation and diarrhea), compared to those children with no GI symptoms (see Fig. 5). Similarly, the diet improved ASD behaviors...
F(1,332) = 8.53, \( P < 0.05 \), physiological symptoms \( F(1,330) = 18.49, \ P < 0.05 \), and social behaviors \( F(1,332) = 7.94, \ P < 0.05 \), for those children with allergy symptoms, compared to those children with no allergy symptoms (see Fig. 5).

Further, the diet was more effective in improving physiological symptoms \( F(1,214) = 10.35, \ P < 0.05 \) and social behaviors \( F(1,216) = 4.64, \ P < 0.05 \), but not ASD behaviors \( F(1,216) = 3.30, \text{n.s.} \) for those children with diagnosed food allergies, compared to those children with no allergy symptoms (see Fig. 5). For children with suspected food sensitivities, the diet improved ASD behaviors \( F(1,331) = 18.91, \ P < 0.01 \), physiological symptoms \( F(1,330) = 31.57, \ P < 0.01 \), and social behaviors \( F(1,331) = 22.05, \ P < 0.01 \), compared to those children with no suspected food sensitivities (see Fig. 6).

**Discussion**

Research suggests that the GFCF dietary intervention may positively affect some children diagnosed with ASD; however, the subsets of children which could potentially benefit from the dietary intervention remain undetermined. The current study analyzed parental report survey data in order to begin to examine which ASD subpopulation and diet implementation factors are associated with greater diet responsiveness. In this study, analysis of parental report data revealed significant group improvements in ASD behaviors, physiological symptoms, and social behaviors related to several ASD subpopulation and diet implementation factors.

Parental report of strict elimination of all foods containing gluten and/or casein was associated with an improvement of ASD behavior, physiological symptoms, and social behaviors, compared to partial elimination interventions. Moreover, parents who reported that diet errors occurred during their care about every other month or at least once a month reported fewer improvements in their child’s ASD behaviors, compared to those parents whose child broke the diet two or fewer times per year during their care. Similarly, diet compliance outside the home correlated with improved ASD behaviors and physiological symptoms among children whose parents reported that the diet was broken two or fewer times per year. Unexpectedly, those parents who reported that the diet was never broken outside of their care reported the fewest number of improved ASD behaviors and physiological symptoms, suggesting that (1) parents may be unaware of diet errors occurring outside of their care and that (2) self-report of these errors may be underestimated. Data from the other diet error outside of parental care groups (i.e. diet was broken two or fewer times per year, about every other month, or at least once per month outside parental care) suggest that diet efficacy improves with decreasing number of diet errors. Together, these findings support the notion that strict diet elimination and compliance may be important factors to consider in optimizing the efficacy of the GFCF dietary intervention for children with ASD. Experimental studies that implement the diet and manipulate the number of diet errors are needed to examine this phenomenon, and it will be important that researchers include ASD subpopulation best responders in these studies in order to minimize potential confounding and mediating factors. Additionally, it is important for researchers
to determine whether parents who report never breaking the diet indeed never broke the diet. This could be confirmed using blood tests (e.g. serum IgA level) in addition to food logs. This compliance check may help to explain why fewer improvements in ASD behaviors were noted by parents who reported that their child never broke the diet during their care (i.e. following strict dietary compliance within the home). Potential explanations include frequent dietary errors outside of parental care (perhaps unknowingly), incomplete gluten and/or casein elimination within the home, or ASD subpopulation factors, such as no history of physiological or allergic symptoms, which correlate with poorer response to the diet intervention. It is important that future studies integrate diet errors inside and outside of the home into one measure of diet error severity.

With regard to length of diet implementation, past randomized trials investigating the GFCF diet’s effectiveness for children with ASD have included 12-month dietary intervention trials, while home trials appear to vary in length. Our analyses revealed that parents who implemented the diet for 6 months or less reported fewer improvements in their child’s ASD behaviors, physiological symptoms, and social behaviors, compared to parents who implemented the diet for more than 6 months. This finding suggests that length of trial implementation may be an important factor in determining whether or not the diet is effective on an individual basis, and within-subject, repeated-measures experimental studies are necessary to determine if shorter diet trials correspond to fewer outcome improvements, compared to longer trials. A determination of adequate trial length is important so that physicians can appropriately advise parents and determine if the diet is effective for each individual child with ASD, taking into account subpopulation factors (e.g. physiological and allergy symptoms) similar to those reported in the present study.

Previous research suggests that there are developmental, behavioral, and somatic factors that might be used to subclass children with ASDs and, further, that some children with ASD may respond more positively to the GFCF dietary intervention, compared to other children. For children with GI symptoms (specifically, constipation and diarrhea), parents reported a greater improvement in ASD behaviors, physiological symptoms, and social behaviors, compared to those children with no GI symptoms. This finding suggests that children predisposed to GI abnormalities might particularly benefit more from a GFCF dietary intervention. Children with allergy symptoms seemed to respond similarly with greater improvements in ASD symptoms, physiological symptoms, and social behaviors, compared to those children with no allergy symptoms. The prevalence of GI and allergy symptoms among our participant pool, along with the associated improved diet efficacy among this subset, in particular, reflects the demonstration by Jyonouchi et al. that 91.8% of the children with autism in their study had a positive response to a gluten-free, casein-free, soy-free diet. Parents of children with diagnosed food allergies also reported greater improvements in their child’s physiological and social behaviors, compared to those of children without food allergy diagnoses. This finding supports the conclusions of Jyonouchi regarding food allergies and ASD. Interestingly, parents who suspected that their child had undiagnosed food sensitivities reported greater improvements in ASD behaviors, physiological symptoms, and social behaviors, compared to those who did not suspect their child had any food sensitivities. Together, these findings suggest that ASD subpopulation factors, such as food and other allergies and food sensitivities, might help investigators to identify potential best responders. Based on these findings, the potential value of the gluten-free, casein-free dietary intervention for treating the subset of ASD children with GI and/or allergy symptoms as well as food allergies and/or sensitivities must be experimentally investigated in future studies.

In all, support was generated for the hypotheses that the GFCF diet would be more effective for those children with digestive issues (namely, constipation and diarrhea), compared to those with no digestive symptoms, and for those children with food allergies, compared to those with no food allergies. The present results suggest that the GFCF diet could be more effective for the subset of ASD children with a history of GI and/or allergy symptoms, and/or a history of food allergies/suspected food sensitivities. As this study is the first to suggest that children with physiological (i.e. GI) and allergy symptoms might benefit most from a GFCF diet, more rigorous research studies are needed in order to more carefully articulate the clinical significance of the GFCF dietary intervention for these children, specifically.

It is critical that future studies investigate the intricacies of subpopulation and diet implementation factors in order to identify those children with ASD who might benefit most from the GFCF dietary intervention. One limitation of this study is that it is a retrospective analysis of parental report regarding the effectiveness of the GFCF diet and, as such, the interpretations of its results are somewhat limited. Specifically, the findings reported may be highly sensitive to parental perceptions (i.e. those parents who started a GFCF diet prior to their child’s diagnosis may also have reported that the diet was more effective, compared to other parents), and thus, their high effectiveness ratings may be explained by a placebo effect. It will be important for future studies to
confirm pre-diet symptoms and diagnoses as well as post-diet symptom improvement with clinical reports in order to eliminate this inherent bias.

Despite the retrospective nature of this survey, the findings remain important because they provide support for a growing handful of studies that suggest that the GFCF diet may positively affect developmental outcomes for some children diagnosed with ASD. Additionally, these findings suggest that the diet may also reduce physiological symptoms (i.e. bodily rash, red ring around the anus, constipation, diarrhea, seizures). Further, to date, parental report information regarding GFCF diet intervention compliance and pre-diet digestive and allergy symptoms as they relate to diet intervention effectiveness has not been scientifically analyzed to our knowledge. Thus, in light of recent experimental studies supporting the potential efficacy of the GFCF diet in improving developmental outcomes for some children diagnosed with ASD, it was imperative that this parental report information be collected and analyzed in a systematic fashion, so that more rigorous experimental studies can be designed to investigate the subpopulation and implementation factors that play a role in optimizing diet efficacy.

As with all studies investigating the effectiveness of the gluten-free, casein-free dietary intervention for children with ASD, it should be noted that children with ASD use various other therapies in conjunction with the diet. These additional therapies must be considered as confounding factors when interpreting the results of the present study and in the analyses of future studies.

Conclusion
These findings provide additional support for the use of this diet in treatment of some children with ASDs, and substantiate the importance of further investigations into the various, nuanced biological and diet implementation factors that interact to optimize and attenuate the diet’s effectiveness for treating these children. From this point forward, it is crucial that the importance of diet compliance, including complete elimination of gluten and casein, is emphasized by those medical practitioners guiding parents through proper implementation of the GFCF intervention for children with ASDs. As an extension of this investigation, it is critical that future researchers identify those patient and implementation characteristics that will ultimately maximize the effectiveness of the diet, such that those individuals with ASD who will benefit most may effectively use it. The priority of future research should be to define the immunological and GI diagnoses and symptoms that best predict those individuals who will be most responsive to the GFCF diet. If these factors are identified, parents and medical practitioners will have the knowledge required to determine, on a case-by-case basis, whether or not the GFCF diet would be a worthwhile treatment endeavor. Future experimental studies are imperative in order to confirm this study’s findings and to further delineate ASD subpopulations that will best respond to the GFCF dietary intervention.

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References