Nuansa Fajar Cemerlang
International Conference
OPTIMAL
Obstetricians Participating in Training for Innovative Measures to Advance Leadership
ISSN 3064-2698

IMPROVEMENT OF AUTISM SPECTRUM DISORDER (ASD) SYMPTOMS THROUGH CONSUMPTION OF FOODS HIGH IN SELENIUM

Neny Triana[1*], Suwoyo[2]
[1]*STIKes Karya Husada Kediri, East Java, Indonesia*
[2]*Poltekkes Kemenkes Malang, East Java, Indonesia*

**Corresponding author: Nenytriana979797@gmail.com*

Abstract

Autism Spectrum Disorder (ASD) is characterized by two symptoms, namely communication/social interaction disorders and restrictive/repetitive behavior. The purpose of this study was to determine the effect of consuming high-selenium foods in the form of high-selenium functional side dishes (HSFF) and selenium supplements (SS) on improving the development of communication, social interaction, sensory cognition, and physical behavior. A randomized controlled experiment design was conducted on 65 ASD children from 14 autism therapy clinics, divided into: HSFF intervention group (n = 22), SS intervention group (n = 22), and control group (n = 21). Before the intervention, all groups underwent a casein-free, gluten-free, and sugar-free (CFGFSF) diet. The HSFF intervention group consisted of side dishes and SS, while the control group was given a placebo in the form of mocaf flour for three months. The results of the intervention were assessed by evaluating improvements in ASD development at the beginning and end of the intervention. The Mann Whitney test and linear regression test were used to evaluate the effects of HSFF and SS. The results of the study in the HSFF and SS groups showed a significant increase in the development of communication, social interaction, sensory cognition, and physical behavior compared to the control group ($p<0.05$), but HS did not affect the improvement of social interaction development after CFGFSF diet compliance and research interventions were given ($p=0.139$). The provision of HSFF and SS can improve the development of ASD children.

Keywords: ASD Children, Developmental Symptoms, Improvement.

1. INTRODUCTION

Developmental problems are one of the nursing problems that nurses often face in children with autism spectrum disorders (ASD). ASD is a group of neurodevelopmental disorders. The prevalence of ASD in the world is 0.6% (Salari et al., 2022). Impaired development of social interaction and communication, repetitive behavior, and limited activity are clinical symptoms of ASD (Christensen et al., 2019) Previous studies have shown that an imbalance in selenium element homeostasis is an important risk factor for ASD (Modabbernia et al., 2017; Sealey et al., 2016). Recent studies have shown that children with ASD have very low levels of selenium (Se) in the blood (Kaczmarek et al., 2024). Selenium minerals and proteins in the body are very important for brain development. Selenium minerals are very important in managing oxidative damage in the brain, and it is suspected that an imbalance in selenium homeostasis may be associated with ASD (Raymond et al., 2014). Selenium is one of the minerals that is recommended to be consumed daily with a dose according to the age of ASD children (J B Adams, 2015).

Selenium minerals can be consumed by ASD children in the form of functional foods or supplements. Selenium supplements are a type of Complementary and Alternative Medicine (CAM) biological therapy with the principle of the methylation cycle that is expected to improve the development of Autism Spectrume Disorder (ASD) which is of interest to parents (Brondino

et al., 2015). Beef liver is one type of functional food that contains selenium (Li, 2017). The selenium content in beef liver is very easy for the body to digest, easy to obtain, and easy for ASD children to consume. The selenium content in 100g of beef liver is 42µg (Li, 2017).

A new experimental study on a mouse model made of ASD found that selenium can improve social function, repetitive behavior, and cognitive function (Wu et al., 2022). Another experimental study found that selenium can reduce ASD symptoms in ASD children (Triana et al., 2023). The effectiveness of selenium mineral administration on the development of communication, social interaction, sensory cognitive, and physical behavior in children with ASD is still unclear, although mineral administration is the type of CAM that is most in demand by parents (James B Adams et al., 2018). Adams et al. in their study found that around 85% of parents liked the administration of minerals and felt that there was a slight improvement in their children, but there is no quantitative data on how much the improvement was (James B Adams et al., 2018). This study aims to determine the effect of consuming high selenium foods in the form of high selenium functional food (HSFF) side dishes in the form of beef liver and selenium supplements (SS) on improving the development of symptoms of communication, social interaction, sensory cognitive, and physical behavior.

2. METHODOLOGY

This study used a Randomized Control Trial (RCT) design. The population and sample of 66 participants (ASD children), the sample was randomly drawn at the beginning of the study into three groups (HSFF intervention group (n = 22), selenium supplement intervention group (n = 22), and control group (n = 22), using block size randomization with a computer application by the researcher. The study sample was 65 ASD children from the beginning to the end of the study. Participants were recruited from 14 different autism clinics in East Java. The researcher did not know the sample of ASD children (blind) when the intervention was carried out. The sample of ASD children in the intervention and control groups did not know whether they were given a placebo intervention or were given food or supplements containing the mineral selenium. The sample size based on the research of Adams et al (James B Adams et al., 2018), it is estimated that a minimum of 22 participants in each group will provide 80% power to detect a difference of 20 scores on the scale, with ά 0.05, allowing an error rate of 10%. The sample was diagnosed with ASD by a pediatrician at the clinic where ASD children were treated based on the Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-5). The inclusion criteria were 2-6 years of age. The exclusion criteria were children with Bell's palsy and other neurological deficits, heart disorders, and mental disorders to prevent bias in the results of developmental assessments.

Intervention: Children with ASD and their parents in the control group and intervention group were given education about the Casein Free Gluten Free Sugar Free (CFGFSF) diet before the intervention was carried out by the researcher. The sample group was advised to follow the CFGFSF diet as a standard diet for children with ASD before the intervention. The three groups were evaluated once a week to ensure the implementation of the CFGFSF diet. Parents of participants were asked to fill in a food logbook consumed by the intervention group and the control group every day.

The HSFF intervention group was given in the form of side dishes with a dose of 50g/day. Beef liver was processed into side dishes cooked by a nutritionist to ensure that the side dishes used the CFGFSF principle. Parents of HSFF group participants were asked to write down how many side dishes were eaten. The control group was given something similar to powder that looked like a supplement, but did not contain high selenium. The SS intervention group was given selenium supplements in the form of powder with a dose of 1 x 20 µg/day. The control group was given a placebo. The packaging of selenium supplements and placebos was carried out by pharmacists from pharmacies that had been accredited by Good Pharmacy Practice (GPP) in the same packaging. The intervention was carried out for three months. Parents of participants were asked to fill out a logbook every time they completed the intervention. Observation of the logbook and clarification of the contents of the logbook to the parents of participants were carried out to measure compliance with the CFGFSF diet and its intervention. Compliance with the CFGFSF diet was measured by dividing the number of days participants consumed casein-free, gluten-free, and sugar-free foods by the number of intervention days then multiplied by 100. Compliance with the research intervention was measured by dividing the total days of the intervention. the

number of days participants consumed selenium supplements/high selenium side dishes/placebos given to each group by the number of intervention days then multiplied by 100.

Measurement and data collection: To determine the decline in development using the Autism Treatment Evaluation Checklist (ATEC) assessment of children with ASD was assessed with (ATEC). ATEC is an instrument commonly used to evaluate the effectiveness of certain therapies or interventions in the development of ASD (Mahapatra et al., 2018). For the developmental test-item analysis using the Spearman correlation test, a correlation coefficient value greater than 0.9 was found for all subscales and total, indicating that the ATEC is reliable (Freire et al., 2018). For the concurrent validity of the ATEC, a correlation coefficient of 0.8 ($p < 0.001$) was found, indicating the high validity of this tool (Freire et al., 2018). The range of scores obtained was 0-28 in communication, 0-40 in social interaction, 0-36 in sensory cognitive, and 0-75 in physical behavior. A decrease in score indicates an increase in results. Scores were collected based on ASD therapy before and after three months of CFGFSF diet education and selenium consumption in the clinic. The therapist who assessed the results was blinded after being assigned to carry out the intervention. The therapist was not informed about the intervention given to the participants so that he did not know the form of intervention carried out by the researcher.

Data analysis: Protocol analysis was used in this study. The Kolmogorov-Smirnov test was used to evaluate the distribution of numerical data. Chi-square test was used to compare baseline categorical data between groups. One-way ANOVA and Kruskal Wallis test were used to compare baseline numerical data and adherence in diet and intervention data. Mann-Whitney U test was used to see p.

3. RESULTS

This RCT design included two intervention and control groups, each containing 22 children with ASD. About 65 ASD children participated in this study until the end of the study. They were considered to have completed the trial if they have received the intervention for three months and study outcomes had been assessed. There was one ASD child who could not follow until the end of the study due to personal reasons (Figure 1).

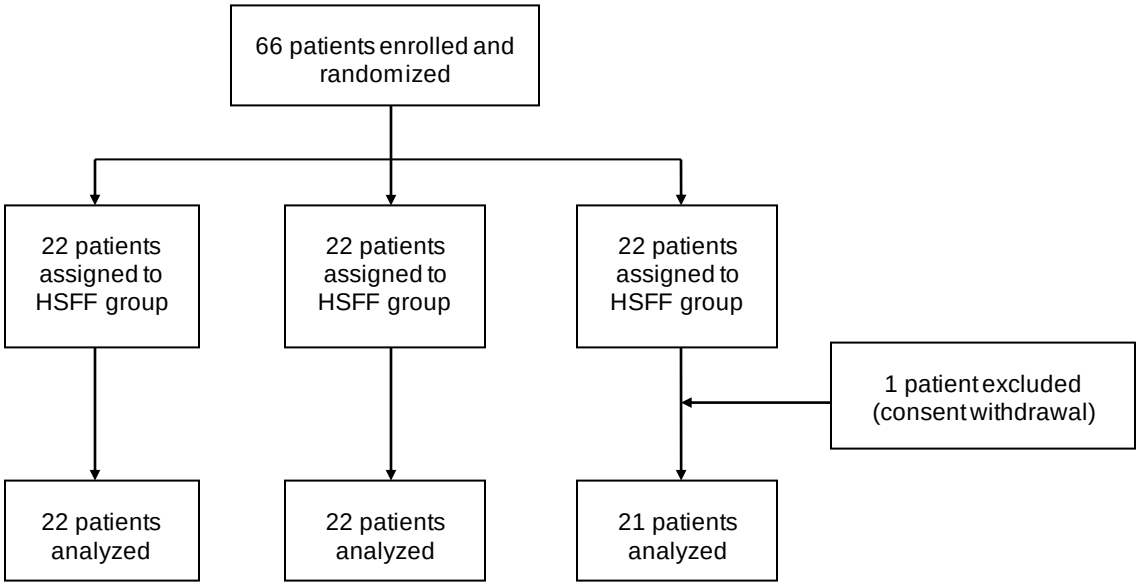

Figure 1. Consort flow diagram of patients

As Table 1 shows, no significant difference in age (p=0.729), sex (0.895), time of ASD diagnosis (p=0.865), communication (0.260), social interaction (0.581), sensory cognitive (0.281), and physical behavior (0.357).

Table 1. *Baseline characteristic of patients*

Characteristics	HSFF group	SS group	Control group	p
Age (years)	3.95 ±1.43	3.91 ± 0.97	3.71 ± 1.34	0.729
Male	19 (33.30)	19 (33.30)	19 (33.30)	0.895
Age for first ASD diagnosis (years)	2.09 ± 0.47	2.04 ± 0.54	2.03 ± 0.47	0.865
Communication	23.86 ± 5.81	20.68 ± 6.34	23.57 ± 6.98	0.260
Social interaction	24.18 ± 9.42	22.90 ± 7.38	25.52 ± 7.57	0.581
Sensory cognitive	21.13 ± 4.68	20.13 ± 7.95	18.04 ± 6.11	0.281
Physical behaviour	29.68 ± 10.76	33.54 ± 10.69	28.95 ± 12.26	0.357

HSFF = high selenium functional food, SS = selenium supplement

Table 2 shows that during the study process, the selenium supplement intervention group had lowest level of compliance with the CFGFSF diet, but the HSFF intervention group had the lowest level of compliance with the intervention given by researchers. There were significant differences in CFGFSG diet and study intervention between groups.

Table 2. *CFGFSF diet and selenium intervention compliance*

Compliance	HSFF group	SS group	Control group	p
CFGFSF diet (%)	69.15 ± 32.01	52.35 ± 24.35	75.94 ± 21.92	0.004
Intervention (%)	83.33 ± 22.34	100.00 ± 0.00	100.00 ± 0.00	0.000

HSFF = high selenium functional food, SS = selenium supplement, CFGFSF: Casein Free Gluten Free Sugar Free

Table 3 shows the change (delta) in communication, social interaction, sensory cognitive, and physical behavior scores. Both consuming HSFF and SS showed significant reduction in communication, social interaction, cognitive sensory, physical, and behaviour scores compared to no selenium administration after the intervention ($p<0.05$).

Table 3. *Delta communication, social interaction, sensory cognitive, and physical behavior*

Delta	HSFF group	SS group	Control group	P HSFF vs SS	p HSFF vs control	p SS vs control
Communication	-7.55 ± -1.02	-8.87 ±-0.79	-1.53 ± -0.29	0.044	0.000	0.000
Social interaction	-5.50 ± -0.99	-14.05 ± -1.29	-2.91 ± -0.15	0.000	0.001	0.009
Sensory cognitive	-4.95 ± -0.17	-11.73 ±-2.23	-1.62 ± -0.37	0.001	0.000	0.002
Physical behaviour	-4.95 ± -2.19	-18.54 ±-2.88	-2.00 ± -0.01	0.044	0.000	0.000

HSFF = high selenium functional food, SS = selenium supplement

Table 4 shows the multivariate analysis that consuming selenium in the form of HSFF and SS affected on the improvement of communication, sensory cognitive, and physical behavior by adjusting for the compliance level of CFGFSF diet and intervention of the study given, however consuming HSFF did not affect social interaction improvement ($p = 0.139$) while consuming SS affected social interaction improvement.

Table 4. *Multiple linier regression analysis showing effect of HSFF and SS with adjusting for compliance of CFGFSF diet and study intervention*

	B	95% CI	P
HSFF			
Communication	6.175	3.436 – 8.915	0.000
Social interaction	1.813	0.932 – 6.404	0.139
Sensory cognitive	4.949	1.895 – 9.044	0.002
Physical behaviour	4.681	1.786 – 7.575	0.002
SS			
Communication	8.247	6.633 – 9.861	0.000
Social interaction	11.356	8.472 – 14.239	0.000
Sensory cognitive	11.110	7.491 – 14.730	0.000
Physical behaviour	18.340	12.930 – 23.750	0.000

HSFF = high selenium functional food, SS = selenium supplement, CI = confidence interval

4. DISCUSSIONS

The study demonstrated that the consuming selenium in the form of HSFF and SS could significantly improve communication, social interaction, sensory cognitive, and physical behavior compared to the control group. This study has proven that selenium both in the form of functional food and supplement can improve the developmental level in ASD children. However, the improvements in ASD were higher in the group consuming SS than in the group consuming HSFF. Selenium in the form of supplement has a more powerful effect on reducing ASD severity than in the form of functional food. Following the previous study in ASD mice model, selenium supplement significantly improved social interaction, repetitive stereotype behavior, and cognitive function (Wu et al., 2022).

We found that the ASD children had better communication, social interaction, cognitive sensory, and behaviour after giving selenium for three months. This improvement may occur because the administration of selenium has fulfilled the selenium deficiency in ASD children. Giving selenium is expected to help the deficiency of the selenium needed for improving ASD symptoms (James B Adams et al., 2018; Bermingham et al., 2014). Following James B Adam et al study, a slight improvement occurred in ASD with the administration of the mineral at a weight-adjusted dose (James B Adams et al., 2018). HSFF in this study was processed beef liver which was given as much as 50g/day containing around 20µg selenium, while SS was given 20µg/day. The daily selenium requirement for ASD children is likely to be sufficient, whereas the daily requirement for children aged 2-7 years is around 15-20 µg/day (Kipp et al., 2015).

Although the levels of the selenium given to HSFF and SS groups were same, compliance with the HSFF intervention was lower than compliance to the SS intervention. This compliance may lead to differences in the rate of the improvement level. The level of compliance with the HSFF intervention was only 83%, while the compliance of ASD children to consume selenium supplements was 100%. ASD children did not consume selenium in the form of HSFF which is given in the form of side dishes every day. There was boredom in the child, because he had to eat beef liver every day even though it was in the form of a different food dish.

Besides consuming selenium, participants had a CFGFSF diet during the study and the study found that the levels of compliance to the diet after the CFGSF diet education was different between groups significantly. The compliance with the CFGFSF diet in control group was higher than in the intervention group. The previous studies found that there was little improvement by doing the diet in ASD children (James B Adams et al., 2018; Brondino et al., 2015; Marsden et al., 2019). However, after adjustment for the CFGFSF diet and intervention compliance, only consuming SS were still shown to improve communication, social interaction, sensory cognitive, and physical behavior. Consuming HSFF improved communication, sensory cognitive and physical behavior but not for social interaction in ASD children. The improvement indicated that selenium might be associated with the pathogenesis of ASD. As mentioned in the previous study, the improvement the ASD mice following selenium supplementation was associated with the fact that selenium altered monoamine neurotransmitter levels in brain tissue, reduced oxidative stress and neuroinflammation in hippocampal tissue, and relieved neural cell damage (Wu et al., 2022). In the body, selenium mainly exists in the form of selenoproteins (Kipp et al., 2015). The decreased level of selenoproteins causes irreversible oxidative stress injury in neurons (Brigelius-Flohé & Flohé, 2017). The study in mice model found that selenium supplementation increased the expression of selenoproteins, which resulted in enhanced antioxidant capacity, reduced reactive oxygen species production, and decreased oxidative stress in tissues (Wu et al., 2022). The micronutrient selenium, which acts as an antioxidant, can improve the development of communication, sensory cognitive, and physical behavior in ASD.

5. CONCLUSIONS

Selenium in the form of supplement and functional food could developmental level, however SS was more powerful in improving communication, social interaction, cognitive sensory, physical, and behavior in AD children than in the form of functional food. The results suggest that selenium supplement and selenium functional food may benefit children with ASD As the implication of the study, parents who have ASD children can give selenium in the form of supplement and functional food to improve communication, social interaction, sensory cognitive,

and physical behavior. The health worker can advise the parents to consume food containing selenium or selenium supplement to improve their ASD children.

There were several limitations of the study. First, all types of selenium containing foods that may be consumed during the study, the types of drugs prescribed by doctors that can affect the results were not controlled in the study. Second, data collection on severity level was not carried out by two therapists at each clinic. However, the data collector is a therapist that person trained in the measurement of the outcomes. Third, the study did not collect the selenium serum level before and after intervention.

ACKNOWLEDGEMENTS

The authors' deepest appreciation to all the participants who helped us in the completion of the manuscript and Kemendikbudristek who provided funding for this research.

REFERENCES

[1] Adams, J B. (2015). Vitamin/mineral supplements for children and adults with autism. *Vitam Miner*, *3*(127), 1318–2376. https://doi.org/10.4172/2376-1318.1000127.

[2] Adams, James B, Audhya, T., Geis, E., Gehn, E., Fimbres, V., Pollard, E. L., Mitchell, J., Ingram, J., Hellmers, R., Laake, D., & others. (2018). Comprehensive nutritional and dietary intervention for autism spectrum disorder—a randomized, controlled 12-month trial. *Nutrients*, *10*(3), 369. https://doi.org/10.3390/nu10030369

[3] Bermingham, E. N., Hesketh, J. E., Sinclair, B. R., Koolaard, J. P., & Roy, N. C. (2014). Selenium-enriched foods are more effective at increasing glutathione peroxidase (GPx) activity compared with selenomethionine: A meta-analysis. *Nutrients*, *6*(10), 4002–4031. https://doi.org/10.3390/nu6104002.

[4] Brigelius-Flohé, R., & Flohé, L. (2017). Selenium and redox signaling. *Archives of Biochemistry and Biophysics*, *617*, 48–59. https://doi.org/10.1016/j.abb.2016.08.003.

[5] Brondino, N., Fusar-Poli, L., Rocchetti, M., Provenzani, U., Barale, F., & Politi, P. (2015). Complementary and alternative therapies for autism spectrum disorder. *Evidence-Based Complementary and Alternative Medicine*, *2015*. https://doi.org/10.1155/2015/258589.

[6] Christensen, D. L., Maenner, M. J., Bilder, D., Constantino, J. N., Daniels, J., Durkin, M. S., Fitzgerald, R. T., Kurzius-Spencer, M., Pettygrove, S. D., Robinson, C., & others. (2019). Prevalence and characteristics of autism spectrum disorder among children aged 4 years— early autism and developmental disabilities monitoring network, seven sites, United States, 2010, 2012, and 2014. *MMWR Surveillance Summaries*, *68*(2), 1. https://doi.org/10.15585/mmwr.ss6802a1.

[7] Freire, M. H., André, A. M., & Kummer, A. M. (2018). Test-retest reliability and concurrent validity of Autism Treatment Evaluation Checklist (ATEC). *Jornal Brasileiro de Psiquiatria*, *67*, 63–64. https://doi.org/10.1590/0047-2085000000186/

[8] Kaczmarek, K. A., Dobrzyńska, M., & Drzymała-Czyż, S. (2024). Iron, magnesium, zinc and selenium-The most common elemental deficiencies in children with autism spectrum disorder. *Research in Autism Spectrum Disorders*, *110*, 102288.

[9] Kipp, A. P., Strohm, D., Brigelius-Flohé, R., Schomburg, L., Bechthold, A. ea, Leschik-Bonnet, E., Heseker, H., (DGE, G. N. S., & others. (2015). Revised reference values for selenium intake. *Journal of Trace Elements in Medicine and Biology*, *32*, 195–199. https://doi.org/10.1016/j.jtemb.2015.07.005.

[10] Li, C. (2017). The role of beef in human nutrition and health. In *Ensuring safety and quality in the prod uction of beef* (Vol. 2, p. 329). https://doi.org/10.19103/AS.2016.0009.16.

[11] Mahapatra, S., Vyshedsky, D., Martinez, S., Kannel, B., Braverman, J., E delson, S. M.,

& Vyshedskiy, A. (2018). Autism Treatment Evaluation Checklist (ATEC) norms: A “growth chart” for ATEC score changes as a function of age. *Children*, *5*(2), 25.

[12] Marsden, R. E. F., Francis, J., & Garner, I. (2019). Use of GFCF diets in children with ASD. An investigation into parents’ beliefs using the theory of planned behaviour. *Journal of Autism and Developmental Disorders*, *49*(9), 3716–3731. https://doi.org/10.1007/s10803-019-04035-8.

[13] Modabbernia, A., Velthorst, E., & Reichenberg, A. (2017). Environmental risk factors for autism: an evidence-based review of systematic reviews and meta-analyses. *Molecular Autism*, *8*(1), 1–16. https://doi.org/10.1186/s13229-017-0121-4.

[14] Raymond, L. J., Deth, R. C., & Ralston, N. V. C. (2014). Potential role of selenoenzymes and antioxidant metabolism in relation to autism etiology and pathology. *Autism Research and Treatment*, *2014*. https://doi.org/10.1155/2014/164938.

[15] Salari, N., Rasoulpoor, S., Rasoulpoor, S., Shohaimi, S., Jafarpour, S., Abdoli, N., Khaledi-Paveh, B., & Mohammadi, M. (2022). The global prevalence of autism spectrum disorder: a comprehensive systematic review and meta-analysis. *Italian Journal of Pediatrics*, *48*(1), 1– 16. https://doi.org/10.1186/s13052-022-01310-w.

[16] Sealey, L. A., Hughes, B. W., Sriskanda, A. N., Guest, J. R., Gibson, A. D., Johnson-Williams, L., Pace, D. G., & Bagasra, O. (2016). Environmental factors in the development of autism spectrum disorders. *Environment International*, *88*, 288–298. https://doi.org/10.1016/j.envint.2015.12.021.

[17] Triana, N., Sulchan, M., Mexitalia, M., & Suryani, M. (2023). Effect of Selenium Micronutrient on Inflammatory Status in Autism Spectrum Disorder Children: A Randomized Control Trial. *Evidence Based Care*, *13*(1), 44–51. https://doi.org/10.22038/ebcj.2023.67834.2775.

[18] Wu, H., Zhao, G., Liu, S., Zhang, Q., Wang, P., Cao, Y., & Wu, L. (2022). Supplementation with selenium attenuates autism-like behaviors and improves oxidative stress, inflammation and related gene expression in an autism disease model. *The Journal of Nutritional Biochemistry*, *107*, 109034. https://doi.org/10.1016/j.jnutbio.2022.109034.