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Original Paper

High Phenolic Extra Virgin Olive Oil Influences the Gut-brain Axis in Individuals Diagnosed with Autism Spectrum Disorder

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Abstract

Autism Spectrum Disorder (ASD) is an umbrella term that incorporates a range of neurodevelopmental disorders which affect behaviour, emotion, and communication. In recent years, the effects of dietary habits in individuals with ASD have been mentioned and a distinct correlation has been observed between ASD and gastrointestinal (GI) disorders. We used the Autism Treatment Evaluation Checklist (ATEC) to evaluate whether we can observe differences in behaviour by administering High Phenolic Extra Virgin Olive Oil (HP EVOO) in combination with a special diet to reduce inflammation and consequently improve behaviour. We hypothesise that ASD symptoms will soothe at a proportional rate relative to their severity. Individuals that followed the protocol showed a decrease in severity of their symptoms. Regarding data analysis, we calculated the p-values to evaluate the significance of our results. Our objective is to establish the beneficial influence that HP EVOO has on the gut-brain axis by soothing ASD symptoms.

Keywords

ASD, gut-brain axis, gut microbiota, HP EVOO, inflammation, nutrition

***The European Food Safety Authority (EFSA) approved health claim on olive oil polyphenols as established in Commission Regulation (EU) No. 432/2012. The claim may only be used for olive oil containing at least 5 mg of hydroxytyrosol and its derivatives (tyrosol, oleocanthal), per 20g of olive oil.**

****The HP EVOO used in this study is "Eliama Daily Value Gold".**

Introduction

Autism Spectrum Disorder (ASD) consists of various neurodevelopmental disorders in which individuals exhibit difficulties with social interaction. Gene mutations can have a pleiotropic effect on their phenotype and result in a combination of ASD with other neurodevelopmental disorders. Recent studies on zebrafish models have indicated that neuropathies could initiate at the stage of organogenesis when the brain is being formed and the circuits of the nervous system have yet to be developed. These studies on prenatal factors have shown that the individual can be more sensitive to genetic mutations at specific stages of embryo development (Kozol, 2018).

Studies have established a direct correlation between gastrointestinal (GI) and brain inflammation, and gut microbiota have an important role in this bidirectional communication (Carabotti, Scirocco, Maselli, & Severi, 2015) by regulating the absorption of bacterial metabolites into local epithelial cells. Research has suggested that cell-to-cell coupling by gap junction protein connexin 43 (Cx43) present on the plasma membrane allows direct flow of molecules towards enteric glial cells (EGCs), which are strategically placed at the interface between the enterocytes and enteric neurons, among others (Grubišić & Parpura, 2015).

Research on Cx43 knockout mice indicated that Cx43 was of crucial significance for neurodevelopment (Brown, McClain, Patel, & Gulbransen, 2015). This connection results in pathogenic bacterial metabolites ultimately affecting the central nervous system (CNS) by activating mast cells in the brain causing neuroinflammation.

Blood work and stool analysis in previous research have shown that individuals with ASD are prone to have an imbalance in their gut microbiota which brings up questions about the role of the gut-brain axis in ASD (Adams, Johansen, Powell, Quig, & Rubin, 2011). They also display high concentration of calprotectin which is an inflammatory factor that can increase permeability of the blood brain barrier (BBB) allowing neutrophiles to enter the brain (Wozniak, Kyprou, & Tsolaki, 2020).

Since a significant percentage of individuals with ASD also exhibit GI disorders, we could hypothesise that a low nutritional value diet would negatively affect normal brain activity by dysregulating the gut-brain axis. Furthermore, many international scientific protocols stress the importance of olive oils and fish oils to reduce gut inflammation and consequently brain inflammation. Instead of administering regular extra virgin olive oil (EVOO), as other protocols suggest, high phenolic extra virgin olive oil (HP EVOO) will be used to prove that it has greater efficiency on the gut-brain axis. HP EVOO is used by many global treatment protocols for children on the autism spectrum due to many of its contents contributing to healthy function of the nervous system utilising a synergistic mechanism of anti-inflammatory, antioxidizing, and immunoactivating properties. Any HP EVOO could be used as long as it has an approved health claim by the European Food Safety Authority (EFSA) with

Commission Regulation (EU) No. 432/2012, and contains at least 5 mg of hydroxytyrosol and its derivatives (tyrosol, oleocanthal), per 20 grams of olive oil. The anti-inflammatory, anti-oxidising and immunostimulating properties of HP EVOO are due to the effect of their high polyphenolic content (Cory, Passarelli, Szeto, Tamez, & Mattei, 2018). The Mediterranean diet includes olives and olive oil (Olea europaea L.) as core products, which contain polyphenols such as tyrosol, hydroxytyrosol, oleuropein, oleocanthal, a high concentration of vitamins and unsaturated fatty acids and squalene, all of which affect synergistically on health.

Hydroxytyrosol is a highly efficient antioxidant based on its oxygen radical absorbance capacity and is the only compound able to penetrate the BBB (Fan, Peng, & Li, 2023). Oleocanthal exists only in HP EVOO and is produced when the olives are crushed and thus needs to be stored in an opaque container infused with nitrogen to avoid oxidation. Oleocanthal can modify astrocyte activation, by increased endothelial cell tightness, which is of significant importance regarding the integrity of the BBB and restores its function (Al Rihani, Darakjian, & Kaddoumi, 2020). Oleocanthal also inhibits lipopolysaccharide (LPS)-mediated upregulation of proinflammatory signaling molecules, cyclooxygenase-2 (COX-2) and 5-lipoxygenase (5-LOX) which is responsible for the biosynthesis of proinflammatory leukotrienes and therefore serves as a non-steroidal anti-inflammatory drug candidate better than ibuprofen which inhibits COX-2 but not 5-LOX (Zarghi & Arfaei, 2011). There are also high contents of oleuropein which acts as a proteasome activator and detoxifies the cells from protein build-up. Tocopherol A, squalene, monounsaturated omega - 9, omega-7 and polyunsaturated fatty acids such as omega-3 and omega-6 all present in HP EVOO demonstrate beneficial effects on the CNS using a synergistic mechanism. Research shows the potential role of polyphenols in regulating neurotrophic levels of Nerve Growth Factor (NGF) and Brain-Derived Neurotrophic Factor (BDNF). This evidence establishes their role in neuroprotection (Wozniak, Kyprou, & Tsolaki, 2020).

In this research work, we strive to alleviate ASD symptoms by using HP EVOO in combination with a special diet followed by individuals diagnosed with ASD that is based on reduced consumption of gluten, casein A1, and refined sugar according to a specialist's suggestion. We anticipate the soothing of symptoms in proportion to their severity. Previous experiments suggest that individuals with more severe ASD will exhibit more noticeable progress.

Materials & Methods

The study took place in Cyprus in the spring of 2021 and was part of a thesis project at the University of Cyprus. The study consisted of 6 participants, aged 4 to 11, all diagnosed with ASD of various severity. The ratio of males to females was 5:1. Alongside ASD most of them were also diagnosed with other related disorders. They were divided into a study population and a control population (Table 1).

Table 1. Demographic Table of Participants

STUDY POPULATION				CONTROL POPULATION			
PARTICIPANTS	**AGE (yrs)**	**SEX (M/F)**	**DIAGNOSIS**	**PARTICIPANTS**	**AGE (yrs)**	**SEX (M/F)**	**DIAGNOSIS**
1S	9	M	Mild ASD, sleep disorders	1C	11	M	Mild ASD, food allergies/intolerances
2S	7	M	Moderate ASD, ADHD	2C	9	M	Moderate ASD
3S	4	M	Severe ASD	3C	4	F	Severe ASD, Epilepsy

The children are noted with a number from 1-3, with larger numbers indicating higher severity, and then either the letter "S" (study) or "C" (control). For each child their age, sex, and diagnosis are provided. ADHD: Attention Deficit Hyperactivity Disorder.

The HP EVOO we used was "Eliama Daily Value Gold" which is approved by EFSA health claim and has all certifications. It is produced by early harvest of "koroneiki" olives. This is because even though ripe olives produce twice as much juice than unripe olives, they lack the phenolic concentration that unripe olives have.

The Autism Treatment Evaluation Checklist (ATEC) is a questionnaire that evaluates the severity of autism in children. Instead of being medically evaluated on a regular basis, the ATEC served to facilitate the evaluation process by giving the responsibility to the children's caretakers in open units or parents/guardians to fill in. It is important to note that the ATEC is not a diagnostic neuropsychological tool but should be used merely as an observation method to keep track of any immediate behavioural differences.

Table 2. Corresponding Scores to each Answer of each Subcategory of the ATEC

SCALE I	**SCALE II**	**SCALE III**	**SCALE IV**
not true **[N] = 2**	not descriptive **[N] = 0**	not descriptive **[N] = 2**	not a problem **[N] = 0**
somewhat true **[S] = 1**	somewhat descriptive **[S] = 1**	somewhat descriptive **[S] = 1**	minor problem **[MI] = 1**
very true **[V] = 0**	very descriptive **[V] = 2**	very descriptive **[V] = 0**	moderate problem **[MO] = 2**
-	-	-	serious problem **[S] = 3**

The ATEC gives a score from 0 – 180 with higher scores indicating greater severity of symptoms. The questions are divided into 4 scales: Scale I (speech), Scale II (sociability), Scale III (sensory/cognitive awareness), and Scale IV (health/physical/behaviour). Each answer is scored by the corresponding

mark displayed in the table.

The liver is relatively immature in small children and thus they exhibit a lower antioxidant capacity (Rupérez, Mesa, Anguita-Ruiz A, et al., 2020). Children with ASD not only exhibit a lower antioxidant capacity, but glutathione (GSH), an antioxidant, has a lower concentration due to its oxidation into glutathione disulfide (GSSG) which can have deleterious effects (Bjørklund, Doşa, Maes, Dadar, Frye, Peana, & Chirumbolo, 2021). For this reason, participants in the study population, were given a specific dosage of HP EVOO daily, decided by a medical specialist based on age and weight specifications that was increased weekly. ATECs for each individual were filled out at baseline and after a follow-up of 6 weeks to compare test scores.

To analyse the data we considered the P values for each subcategory of both groups. We used the "t-Test: Paired Two Sample for Means' tool on Microsoft Excel and took the "P (T<=t) one-tail" value with a confidence interval of 90% to evaluate the significance of our results for each subcategory.

Table 3. Dosages per Week

	AGE (yrs)	WEIGHT (kg)	WEEK I	WEEK II	WEEK III	WEEK IV	WEEK V	WEEK VI
1S	9	40	2.5 (am)	2.5 (am) + 2.5 (pm)	5 (am) + 2.5 (pm)	5 (am) + 2.5 (pm)	5 (am) + 5 (pm)	7.5 (am) + 7.5 (pm)
2S	7	32	1.5 (am)	1.5 (am) + 1.5 (pm)	2.5 (am) + 1.5 (pm)	2.5 (am) + 1.5 (pm)	3 (am) + 3 (pm)	5 (am) + 5 (pm)
3S	4	21	0.5 (am)	0.5 (am) + 0.5 (pm)	1 (am) + 1 (pm)	1 (am) + 1 (pm)	1.5 (am) + 1.5 (pm)	2.5 (am) + 2.5 (pm)

Dosages recommended by medical specialist for each participant of the study population which increase weekly. Dosages are measured in millilitres (mL), (am): morning and (pm): afternoon.

Results

An ATEC was filled out for each participant at baseline before HP EVOO was administered to the study population. The scores are displayed analytically for each subcategory in Table 4.

Table 4. Baseline ATEC Scores

	Scale I **Speech/Language/Communication** **Range: 0-28**	**Scale II** **Sociability** **Range: 0-40**	**Scale III** **Sensory/Cognitive Awareness** **Range: 0-36**	**Scale IV** **Health/Physical/Behaviour** **Range: 0-75**	**TOTAL** **Range: 0-180**
Study Population					
1S	1	16	11	31	59
2S	9	14	23	25	71
3S	18	31	26	36	111
Control Population					
1C	3	19	10	22	54
2C	4	15	19	27	65
3C	22	24	29	45	120

The ATECs were filled in for all participants before any intervention had begun.

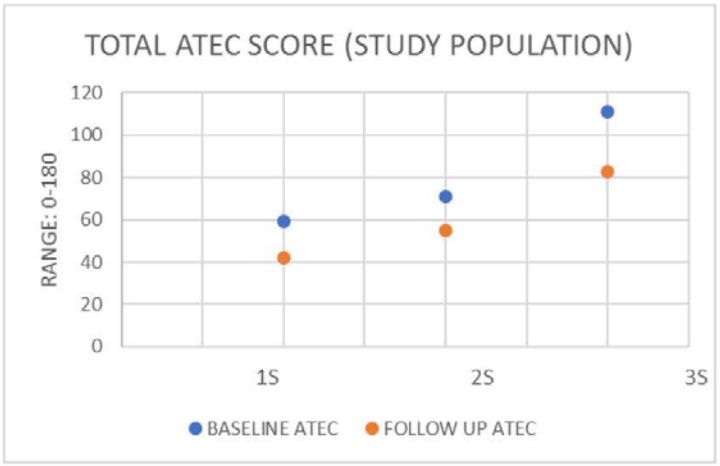
Another ATEC was filled in for each participant as a follow up 6 weeks after the first administration of HP EVOO. The control population carried on with the same dietary habits they had before the study, which consisted of *regular* olive oil in their meals, so they remained under the same circumstances as baseline with no HP EVOO administered. Scores for each subcategory are displayed analytically in Table 5.

Table 5. Follow up ATEC Scores

	Scale I **Speech/Language/Communication** **Range: 0-28**	**Scale II** **Sociability** **Range: 0-40**	**Scale III** **Sensory/Cognitive Awareness** **Range: 0-36**	**Scale IV** **Health/Physical/Behaviour** **Range: 0-75**	**TOTAL** **Range: 0-180**
Study Population					
1S	0	10	8	24	42
2S	8	9	17	21	55
3S	15	22	19	27	83
Control Population					
1C	0	10	2	14	26
2C	4	12	16	29	61
3C	25	35	28	39	127

The ATECs were filled in for all participants after 6 weeks of intervention.

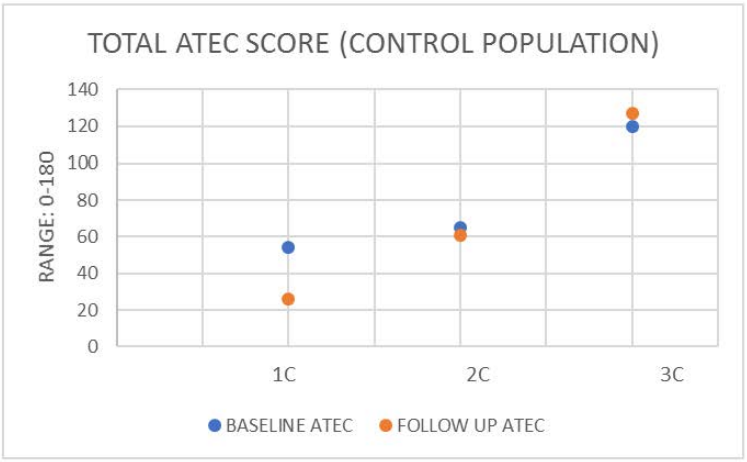
In order to visualise the difference in scores before and after intervention, we constructed a graph displaying the total ATEC scores for each participant. A statistical error was observed with the information provided by participant 1C's final ATEC. This observation can be explained by the fact that participant 1C had been consuming small amounts of black seed (cumin) oil and other natural oils which are also high in antioxidants. 1S exhibited an improvement even though they were in isolation due to COVID-19 during the intervention. 2C showed a stable clinical profile. 2S who also has ADHD, showed an improvement in symptoms. 3C, who was also diagnosed with epilepsy, 3C held a relatively stable score and was the only participant that is female. 3S has severe ASD and showed an impressive drop from a score of 111 to 83 (Figure 1 & 2).



	BASELINE ATEC	FOLLOW UP ATEC
1S	59	42
2S	71	55
3S	111	83

Figure 1. Total ATEC Score (Study Population)

The total ATEC score for all participants of the study population decreased.



	BASELINE ATEC	FOLLOW UP ATEC
1C	54	26
2C	65	61
3C	120	127

Figure 2. Total ATEC Score (Control Population)

For participants 1C and 2C the total score was decreased whereas 3C showed an increase.

We suppose a null hypothesis, that ASD symptoms will not alleviate in the time span of the study. The t-Tests executed with the given data from the study population provided us with p-values for each subcategory to rule out the possibility that the results occurred by chance. For subcategories II, III, and the TOTAL ATEC scores of the study population, we had statistically significant results that supported our alternative hypothesis, that ASD symptoms indeed alleviated in the time span of the study. Scales I and IV provided us with p-values of P=.15 and P=.12, respectively, which are considerably close to α=0.1 and therefore, even if they provide us with a statistical error, can be interpreted as “marginally significant” (Figure 3).

Table 6. Statistical Analysis

STUDY POPULATION				**CONTROL POPULATION**			
SCALE	**P-VALUE**	**P>.1 or P<.1**	**H_0**	**SCALE**	**P-VALUE**	**P>.1 or P<.1**	**H_0**
I	(*P*=.15)	*P*>.1	supported	**I**	(*P*=.25)	*P*>.1	supported
II	(*P*=.09)	*P*<.1	rejected	**II**	(*P*=.33)	*P*>.1	supported
III	(*P*=.02)	*P*<.1	rejected	**III**	(*P*=.15)	*P*>.1	supported
IV	(*P*=.12)	*P*>.1	supported	**IV**	(*P*=.35)	*P*>.1	supported
TOTAL	(*P*=.08)	*P*<.1	rejected	**TOTAL**	(*P*=.42)	*P*>.1	supported

P values and null hypothesis (H_0) for each subcategory of the study and control populations.

Discussion

The ATEC score of each subcategory, for individuals in the study population, decreased significantly and it is important to stress that these differences were observed in a mere span of 6 weeks, even if there was only one week's worth of the optimal dosage administration. This is because the first 5 weeks are an "adapting" phase and that the "intervening" phase starts with the corresponding dosage to each child on the sixth week.

The fact that 1C was administered substances other than HP EVOO during the observation would mean that they should not have been part of the control population, but this fact only serves to prove the argument that oils with high antioxidant concentration are beneficial for mental health development. Also, we cannot rule out the parent's bias while filling out the ATEC.

Since 3C was the only individual who was female, they aren't considered an ideal participant due to the major differences dividing males with ASD and females with ASD. However due to the fact that 3C was a part of the control group and because their symptoms were the severest, their data was used to observe the expected stability in their clinical profile. It is important to stress the decline of 3S's ASD symptoms because it justifies the argument that individuals with more severe ASD are more strongly influenced by the benefits of this protocol when it is followed strictly. Children on the severe autism spectrum usually face significant challenges with communication and social skills. They are also the most likely to remain non-verbal or unable to communicate effectively and may therefore require augmentative and alternative means of communication.

In future replications of this pilot study more parameters, a larger population, longer duration and more ATEC results, rather than only two measurements, could be studied to confirm the findings of this brief research report (Mahapatra, S., Khokhlovich, E., Martinez, S. et al., 2020). Use of different neuropsychological tools used for other conditions such as multiple sclerosis (MS) but adapted to ASD such as the Frontal Assessment Battery (FAB), the Brief International Cognitive Assessment for Multiple Sclerosis (BICAMS) the Mental Health Inventory (MHI), and the Beck Depression Inventory (BDI) would provide more clarity and smoother results (Chatzikostopoulos, Tsolaki, Wozniak,

Basgiouraki, Saoulidis, Michmizos, & Koutsouraki, 2022).

Administration of HP EVOO is a non-invasive method of trying to sooth the body from inflammation since it lets the gut microbiota reduce inflammation without the risk of side effects that psychotropic medication could potentially provide. In order to revamp the protocol, along with administering the individuals with HP EVOO, they were subject to a gluten-free, casein-A1-free, refined sugar-free diet. The reason behind the dietary adjustment is because consumption of gluten, casein A1 and refined sugars can further stimulate growth of opportunistic pathogenic microorganisms in the gut which in turn can cause "leaky gut" (Camilleri, 2010) "Leaky gut" increases intestinal permeability of the epithelium, increases cytokine production, and subsequently allows the entrance of endotoxins into the central nervous system (CNS) through the BBB (Silva, Bernardi, and Frozza, 2020). Consumption of food with low-nutritional value also increases the chance of obesity occurring in individuals with ASD alongside their behavioural symptoms and given their specific limited preferences and difficult behaviours it is obviously difficult to intervene strictly in one's diet without the help of specialists (Doreswamy, Bashir, Guarecuco, Lahori, Baig, Narra, Patel, & Heindl, 2020).

Short-chain fatty acids (SCFAs) are catabolic products of foods high in starch and fibre which interact with G protein-coupled receptors of the epithelium and immune cells (Sivaprakasam, Prasad, & Singh, 2016), stimulate the vagus nerve, translocate through systemic circulation to the CNS and are capable of increasing neurogenesis, improving cognitive development, reducing inflammatory signaling and improving BBB integrity (Breit, Kupferberg, Rogler, & Hasler, 2018). However, in individuals with ASD, when there is an abundance of SCFAs being digested, due to dysbiosis, the liver lacks the capability of metabolising at a quicker rate and the excess SCFAs, especially propionic acid, can remain in the bloodstream and can cause neuroinflammation (Thomas, Meeking, Mepham, Tichenoff, Possmayer, Liu, & MacFabe, 2012). Furthermore, another very important factor to consider is oxidative stress that can cause weakening of the BBB thus activating an inflammatory response. Hydroxytyrosol can prevent this stress because it is able to absorb oxygen radicals giving a positive impact on brain function when it penetrates the BBB.

Conclusion

Our results show that consumption of substances with anti-inflammatory, antioxidant, immunostimulating properties directly benefit mental health and soothe symptoms of neurodevelopmental disorders. Participants of the study population all showed progress in their symptoms whereas the control population, who continued following their usual dietary habits which consisted of normal olive oil, showed a stable clinical profile. It's safe to say that the beneficial effects of HP EVOO were thanks to the high phenolic content. The guardians themselves have given their feedback, confirming the benefit of supplying themselves with HP EVOO. Despite the limitations we faced, this protocol could potentially revolutionise therapies since it works with a different, intrinsic

approach than conventional treatments. This pilot study could be the base of innovative research as a new approach in dealing with ASD and if continued routinely, at the maximum dosage given at 6 weeks, there is no doubt that this method will improve symptoms especially if the intervention occurs at a young age. Of course, we need more information in the future to confirm the influence of HP EVOO. The key to success is nutrition which plays an important role in the regulation of the gut microbiome to prevent neuroinflammation. In conclusion, we should keep in mind the quote of Hippocrates, the father of medicine: "*Let food be thy medicine and medicine be thy food*".

Limitations

It is important to consider the toll that the COVID-19 pandemic took on the outcome of this study due to inevitable lockdowns and isolations which made communication and progression of the study slower and led to compromised results. The general situation caused drastic psychological effects to everyone let alone to children with ASD who are sensitive to adapting and require special attention and care. Another factor to consider is that the limited time frame gave us less time to observe the children in the "intervening" phase and that there were no donations for the purchase of materials. Nevertheless, the vast majority of the data gathered in this pilot study has proved promising.

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Permission was granted from all parties.

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All sources and certifications are at disposal.

Conflicts of interest: None

References

Adams, J. B., Johansen, L. J., Powell, L. D., Quig, D., & Rubin, R. A. (2011). Gastrointestinal flora and gastrointestinal status in children with autism—Comparisons to typical children and correlation with autism severity. *BMC gastroenterology*, *11*, 22. https://doi.org/10.1186/1471-230X-11-22

Al Rihani, S. B., Darakjian, L. I., & Kaddoumi, A. (2019). Oleocanthal-Rich Extra-Virgin Olive Oil Restores the Blood-Brain Barrier Function through NLRP3 Inflammasome Inhibition Simultaneously with Autophagy Induction in TgSwDI Mice. *ACS chemical neuroscience*, *10*(8), 3543-3554. https://doi.org/10.1021/acschemneuro.9b00175

Bjørklund, Doşa, Maes, Dadar, Frye, Peana, & Chirumbolo. (2021). The impact of glutathione metabolism in autism spectrum disorder. *Pharmacological Research*, *166*, 105437. https://doi.org/10.1016/j.phrs.2021.105437

Breit, S., Kupferberg, A., Rogler, G., & Hasler, G. (2018). Vagus Nerve as Modulator of the Brain-Gut Axis in Psychiatric and Inflammatory Disorders. *Frontiers in psychiatry*, *9*, 44. https://doi.org/10.3389/fpsyt.2018.00044

Brown, I. A., McClain, J. L., Patel, B. A., & Gulbransen, B. D. (2015). Activation of Enteric Glia Contributes to Enteric Neuron Death in Inflammation. The FASEB Journal, 29. Retrieved from https://www.semanticscholar.org/paper/Activation-of-Enteric-Glia-Contributes-to-Enteric-Brown-McClain/b5f98eeb497033897b0823305d30d51f3202ef04

Camilleri M. (2019). Leaky gut: Mechanisms, measurement and clinical implications in humans. *Gut*, *68*(8), 1516-1526. https://doi.org/10.1136/gutjnl-2019-318427

Carabotti, M., Scirocco, A., Maselli, M. A., & Severi, C. (2015). The gut-brain axis: Interactions between enteric microbiota, central and enteric nervous systems. *Annals of gastroenterology*, *28*(2), 203-209. https://pubmed.ncbi.nlm.nih.gov/25830558/

Chatzikostopoulos, T., Tsolaki, M., Wozniak, G., Basgiouraki, E., Saoulidis, I., Michmizos, D., & Koutsouraki, E. (2022). The Effects of Early-Harvest Extra Virgin Olive Oil on Cognition and Mental Health of Primary (PPMS) or Secondary (SPMS) Progressive Multiple Sclerosis Patients The Effects of Early-Harvest Extra Virgin Olive Oil on Cognition and Mental Health of Primary (PPMS) or Secondary (SPMS) Progressive Multiple Sclerosis Patients. *Global Journal of Medical Research*. Retrieved from https://www.researchgate.net/publication/369570598_The_Effects_of_Early-Harvest_Extra_Virgin_Olive_Oil_on_Cognition_and_Mental_Health_of_Primary_PPMS_or_Secondary_SPMS_Progressive_Multiple_Sclerosis_Patients_The_Effects_of_Early-Harvest_Extra_Virgin_O

Cory, H., Passarelli, S., Szeto, J., Tamez, M., & Mattei, J. (2018). The Role of Polyphenols in Human Health and Food Systems: A Mini-Review. *Frontiers in nutrition*, *5*, 87. https://doi.org/10.3389/fnut.2018.00087

Doreswamy, S., Bashir, A., Guarecuco, J. E., Lahori, S., Baig, A., Narra, L. R., Patel, P., & Heindl, S. E. (2020). Effects of Diet, Nutrition, and Exercise in Children with Autism and Autism Spectrum Disorder: A Literature Review. *Cureus*, *12*(12), e12222. https://doi.org/10.7759/cureus.12222

Fan, L., Peng, Y., & Li, X. (2023). Brain regional pharmacokinetics of hydroxytyrosol and its molecular mechanism against depression assessed by multi-omics approaches. *Phytomedicine : international journal of phytotherapy and phytopharmacology*, *112*, 154712. https://doi.org/10.1016/j.phymed.2023.154712

Grubišić, V., & Parpura, V. (2015). The second brain in autism spectrum disorder: could connexin 43 expressed in enteric glial cells play a role? *Frontiers in cellular neuroscience*, *9*, 242. https://doi.org/10.3389/fncel.2015.00242

Kozol R. A. (2018). Prenatal Neuropathologies in Autism Spectrum Disorder and Intellectual Disability: The Gestation of a Comprehensive Zebrafish Model. *Journal of developmental biology*, *6*(4), 29. https://doi.org/10.3390/jdb6040029

Mahapatra, S., Khokhlovich, E., Martinez, S., Kannel, B., Edelson, S. M., & Vyshedskiy, A. (2020). Longitudinal Epidemiological Study of Autism Subgroups Using Autism Treatment Evaluation Checklist (ATEC) Score. *Journal of autism and developmental disorders*, *50*(5), 1497-1508. https://doi.org/10.1007/s10803-018-3699-2

Rupérez, A. I., Mesa, M. D., Anguita-Ruiz, A., González-Gil, E. M., Vázquez-Cobela, R., Moreno, L. A., Gil, Á., Gil-Campos, M., Leis, R., Bueno, G., & Aguilera, C. M. (2020). Antioxidants and Oxidative Stress in Children: Influence of Puberty and Metabolically Unhealthy Status. *Antioxidants (Basel, Switzerland)*, *9*(7), 618. https://doi.org/10.3390/antiox9070618

Silva, Y. P., Bernardi, A., & Frozza, R. L. (2020). The Role of Short-Chain Fatty Acids from Gut Microbiota in Gut-Brain Communication. *Frontiers in Endocrinology*, *11*. Retrieved from https://www.frontiersin.org/articles/10.3389/fendo.2020.00025

Sivaprakasam, S., Prasad, P. D., & Singh, N. (2016). Benefits of short-chain fatty acids and their receptors in inflammation and carcinogenesis. *Pharmacology & therapeutics*, *164*, 144-151. https://doi.org/10.1016/j.pharmthera.2016.04.007

Thomas, R. H., Meeking, M. M., Mepham, J. R., Tichenoff, L., Possmayer, F., Liu, S., & MacFabe, D. F. (2012). The enteric bacterial metabolite propionic acid alters brain and plasma phospholipid molecular species: further development of a rodent model of autism spectrum disorders. *Journal of neuroinflammation*, *9*, 153. https://doi.org/10.1186/1742-2094-9-153

Wozniak, G., Kyprou, M., & Tsolaki, M. (2020). *The Effect of Cretan High Phenolic Olive Oil on Fecal Calprotectin Levels in the Course of Multiple Sclerosis*, *12*. 65-79. Retrieved from https://www.researchgate.net/publication/369571347_The_Effect_of_Cretan_High_Phenolic_Olive_Oil_on_Fecal_Calprotectin_Levels_in_the_Course_of_Multiple_Sclerosis

Zarghi, A., & Arfaei, S. (2011). Selective COX-2 Inhibitors: A Review of Their Structure-Activity Relationships. *Iranian journal of pharmaceutical research: IJPR*, *10*(4), 655-683. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3813081/