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The effect of Taraxacum officinale extract on the sodium valproate-induced model of autism and assessment of oxidative stress

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ABSTRACT

Background:

A lifelong neurodevelopmental illness, autism spectrum disorder (ASD) is typified by core symptoms that include confined, repetitive patterns of behavior, interests, or hobbies, as well as persistent difficulties in social communication and social interaction. The incidence and burden of ASD on the affected person, their family, and society are both noteworthy.

Aim of the study

To evaluate the effects of Taraxacum officinale extract on the sodium valproate-induced model of autism and the level of antioxidant stress biomarkers.

Materials and Methods:

60 healthy albino mice (20 female and 40 male) were used in this study. The mice were split into 20 groups, each with one male and two females for mating. After establishing the zero-day, On the twelfth day of gestation, female mice were divided into two groups. One group was given intraperitoneal (i.p.) injections of water only. By contrast, to create an experimental model of autism, the second group was given a single intraperitoneal injection (i.p.) of sodium valproate at a dosage of 600 mg/kg. The baby mice were taken from their moms on the 40th day after birth, and they were divided into eight groups at random (four control groups and four VPA experimental groups) with each group consisting of the following: control no treatment no=10, T.O. 200 mg/kg no=10, T.O. 400 mg/kg no=10, and risperidone 1 mg/kg no=10. Following a 20-day course of therapy, all groups underwent an open field test. Following the completion of the experiment and 24 hours after the last treatment dose, mice were beheaded, with just two brain hemispheres removed. These were homogenized and ready for the Elisa procedure, which measures GSH and MDA concentrations.

Results

The finding of this study showed that the length of the tail in prenatally VPA-exposed groups decreased in comparison with Control groups. Also showed that TOE at both doses (200mg/kg and 400mg/kg) had anxiolytic activity, decreased hyperactivity, increased latency duration in the core, and showed better results than risperidone in reducing grooming and rearing in VPA- mice during the open field test. Also showed antioxidant activity by

decreasing MDA level with superior efficacy to risperidone in VPA- offspring; although *T. Officinale* at both doses decreased lipid peroxidation in control groups better than risperidone and at a dose of 400mg/kg it increased the level of GSH in VPA- offspring more than risperidone did.

Conclusion:

These results showed that prenatal exposure to sodium valproate at a dose of 600mg/kg caused a decrease in the length of the tail of mice in comparison to control offspring. TOE acts as an anxiolytic by managing symptoms of autism, such as anxiety, hyperactivity, and fear. It reduced grooming and rearing in VPA mice more effectively than risperidone. TOE had antioxidant activity by decreasing the levels of MDA and GSH in control and VPA offspring.

Keywords : autism spectrum disorder (ASD), length of tail, oxidative stress, GSH, and MDA.

INTRODUCTION

Autism spectrum disorder (ASD) is a neurodevelopmental illness characterized by repetitive behavior, social communication difficulties, and deficiencies in verbal and nonverbal communication. It affects two to three percent of the population and is characterized by limited, repetitive patterns of behavior, interests, and activities [1]. The etiology of autism spectrum disorder (ASD) is complex and has been related to genetic variables as well as environmental factors such as chemicals, infections, and drugs that alter epigenetic modifications [2]. ASD is linked to elevated ROS levels and decreased antioxidant capacity, which can lead to pro-inflammatory conditions and oxidative stress. The immune system's dysfunction can also cause changes in cell metabolism, including brain cell metabolism, promoting oxidative stress and chronic inflammatory diseases. Therefore, it is expected that individuals with ASD will have a strong relationship between the immune system, inflammation, and oxidative stress [3]. Reactive oxygen species (ROS) are produced during the reduction-oxidation reaction, a crucial mechanism in cell biology. These can be intentionally produced to kill pathogens or as intermediates in enzymatic reactions, or accidentally through drug metabolism, electron leakage, exposure to chemicals, pollutants, and radiation [4]. ROS levels are typically low due to enzyme systems. Still, excessive production can disrupt the body's antioxidant defense, leading to oxidative stress, damage to cellular lipids, proteins, DNA, neuroinflammation, impaired astrocyte-neuron communication, and ASD [5,6]. Mitochondria have various functions such as apoptosis regulation, calcium signaling, and energy metabolism. They are also the main source of ROS through the electron transport chain. Oxidative stress can lead to mitochondrial dysfunction, causing a vicious cycle of ROS production. The number of mitochondria varies by cell type and energy requirements, with high-energy-demanding cells like brain cells containing more mitochondria. The brain is particularly vulnerable to oxidative stress due to its high energy consumption [4,7]. Reduced GSH, a tripeptide with antioxidant properties, is crucial for maintaining intracellular redox pathways and eliminating harmful substances. ASD is linked to lower concentrations of reduced GSH, partly due to oxidation to glutathione disulfide (GSSG). GSSG reactions with protein sulfhydryl (SH) groups can cause proteotoxic stress and other abnormalities in the brain and blood. GSH metabolism changes, affecting redox-independent systems, are also linked to ASD pathophysiology. GSH modulates glutamate ionotropic receptors, protecting the brain against glutamate excitotoxicity. Additionally, glutamate has a role as a glutathione precursor so the interaction between glutathione and glutamate in brain diseases can vary from synergism to antagonism [8,9]. MDA, a byproduct of lipid peroxidation, is a sign of oxidative stress, causing tissue damage and causing damage

to various cell types. As a highly neurotoxic molecule, MDA remains in the body longer than reactive oxygen species, potentially affecting the pathophysiology of ASD. It damages brain tissue, induces necrosis and apoptosis, and damages cortical neurons by depolarizing the mitochondrial membrane, cross-linking proteins, and inducing calcium overload and influx. This increased cellular accumulation could be a factor in the development of neurodegenerative illnesses like autism spectrum disorder [10,11]. Pregnant use of valproic acid (VPA) during pregnancy increases the risk of somatic abnormalities, neurodevelopmental delays, and a 7–10× increase in the frequency of ASD in offspring. Pregnant rodents exposed to VPA exhibit birth malformations, neurodevelopmental impairments, and cognitive/social abnormalities similar to children with ASD. Theories of VPA neurotoxicity include increased free radical production, disruptions in cell migration and proliferation, modifications to inflammatory and immunologic markers, also VPA acting as an inhibitor of histone deacetylase altering the expression of several genes [12]. There is currently insufficient data to support the routine use of new medication to treat specific symptoms of ASD or control its fundamental social symptoms. The only drugs approved by the FDA to treat ASD symptoms are aripiprazole and risperidone [13].

This study utilized the *Taraxacum officinale* (TO) or dandelion, a plant belonging to the Asteraceae family, a subfamily of the Cichorioideae. Despite its European origin, it is found globally and has therapeutic properties. The plant is tolerant of soil and used in the food industry, but dosages should not exceed 4g or 12g per day. It is used in herbal medicine and is known for its antioxidant, hepatoprotective, anti-inflammatory, anti-obesity, and antidepressant properties, which are beneficial for treating various conditions [14,15]. Examples of antioxidant compounds in TO include chicoric acid (present in all plant parts), chlorogenic acid (flower), caffeic acid (flower and root), hydroxycinnamic acids (fruit), and luteolin (above-ground plant part) [16]. Flavonoids, important polyphenolic chemicals with antioxidant properties, help maintain B-cell function by interacting with reactive oxygen species (ROS) and free radicals. They also aid in energy homeostasis, shielding biomolecules from potential oxidative damage, which could lead to various diseases [17]. Previous studies showed that Dandelions' leaves and petals contain phenolic fractions that significantly reduce lipid peroxidation, carbonylation of proteins, and thiol group oxidation in human plasma proteins. These effects were observed in H2O2 and H2O2/Fe-induced oxidation. After using 85% phenolic fractions at a concentration of 0.5 mg/mL, plasma lipid peroxidation inhibition was around 50% [18]. Another study found that TO administration significantly reduced MDA and ROS levels in diabetic animals, while increasing the activities of superoxide dismutase, GPX, catalase, and glutathione reductase. TOE also ameliorated mitochondrial dysfunction, such as decreased enzyme activity and increased ROS production. These findings indicate the antioxidant activity of TO.[19].

The open field test (OFT) is a widely used animal model for studying adaptation behavior in novel environments, particularly concerning anxiety and impulsive movement. It provides data on emotional responses to anxiety, aiding in the discovery of new therapeutic targets and anxiolytic substances [20].

Materials and Methods

60 healthy albino mice, 20 male and 40 female, weighing 25–40 g and 12 weeks old, were used in this investigation. They were obtained from the Ministry of Science and Technology in Baghdad, Iraq. The animals were housed in the animal house of the College of Medicine/University of Babylon. They were kept in standard plastic cages with free access to food pellets and water, and they were kept at a temperature of 25±5 °C, 60±5% humidity, and

a 12-hour light/dark cycle. This study was conducted from July 10, 2023, to February 14, 2024, at the college.

Once the mice have been acclimated for two weeks make 20 groups of mice, one male and two females for mating. The vaginal plug, which sticks to the cervix and vaginal canal for 24 to 48 hours after ejaculation, is a sign that mating has occurred after the mice have mated for the night. Find out if the plug is there. Grab the base of the tail to elevate the back legs of the female mouse momentarily. Determine the copulatory plug that is close to the vaginal entrance using a toothpick. To take a vaginal swab sample, gently extend the lips of the vaginal opening with a cotton-tipped swab. Then, to identify sperm, lay the specimen on a sanitized glass microscope slide and examine it under a microscope. Females who had sperm in their vaginal swap were confirmed to be pregnant at day zero [21,22].

Two groups of female mice were formed on the twelfth day of gestation. Intraperitoneally (i.p.) injections of normal saline were given to the control group. On the other hand, to create an experimental model of autism, the experimental group was given a single intraperitoneal injection (i.p.) of sodium valproate at a dose of 600 mg/kg [23].

A complete portion of *Taraxacum officinale* (TO) was purchased from the local market in Baghdad to prepare it. An electric grinder was used to crush a dried plant into a powder, and 30g of the powder was extracted using 300 ml of 70% ethanol in a soxhlet device, ensuring a 1:10 solid mass to solvent ratio. The resulting crude extracts were further concentrated by evaporating the ethanol in the microwave at 40 °C. There was a residue that was dark brown. The leftover material was preserved in airtight receptacles and placed in a deep freezer. About 2g of extract was produced from 30 g of powdered plant [24].

On the 40th postnatal day, the offspring mice were separated from their mothers and placed into 8 groups (4 control groups and 4 VPA groups), each group with 10 animals:

Control groups

Group 1: was given oral saline for 20 days.

Group 2: was given oral *Taraxacum officinale* 200 mg/kg for 20 days.

Group 3: was given 400 mg/kg of *Taraxacum officinale* orally for 20 days.

Group 4: took risperidone orally for 20 days at a dose of 1 mg/kg [25].

VPA groups

Group 5: took oral saline for 20 days

Group 6: was given oral Taraxacum officinale 200 mg/kg for 20 days.

Group 7: was given oral Taraxacum officinale 400 mg/kg for 20 days.

Group 8: taken orally for 20 days at a dose of 1 mg/kg risperidone [25].

After a day off from the last therapeutic dosage, the open field test was carried out. The open field was made by using a 100 cm x 100 cm wooden square surrounded by a 40 cm high wall, and its square floor was divided into 100 equal squares by thin white lines. A test mouse was

placed in an open field box and given five minutes to roam freely in each quadrant. The tracking camera recorded the mouse's movement. After each trial, 70% ethanol was used to remove any traces of the previous mouse's scent. Crossings measured the number of square crossings, while rearings measured the number of erect positions the mouse adopted. The time spent in the central region of the open field box was measured by adding up visits to the center. Grooming measured the total number of licking the tail, ventrum, and wiping the face.

Following the completion of the open field test and leaving 24 hours after the last treatment dosage, Mice were beheaded and had their heads severed from their bodies. Making an incision in the midsagittal scalp, removing the skin, and cutting into the back of the skull with sharp scissors exposed the skull. The mouse's brain was visible once the skull was peeled apart using forceps. After the brain was carefully extracted from the skull, the cerebellum and olfactory bulb were cut off, leaving just two hemispheres of the brain intact. We used 10% (w/v) phosphate-buffered saline PBS to wash both hemispheres. After that, the brain was quickly chilled by being placed on ice in an Eppendorf tube. following which it was frozen at -20°C and put in the refrigerator. Weighing 0.1 g of frozen brain, it was placed in an Eppendorf tube containing PBS. After that, the brain tissue was homogenized with a homogenizer and spun for 30 seconds at a temperature between 2 and 8 °C. The Eppendorf tubes were then placed in a centrifuge and spun for 5 minutes at a temperature between 2 and 8 °C at 1300 rpm in order to extract the supernatants. Next, carefully remove the supernatant with a micropipette so that it may be biochemically tested to measure the concentrations of MDA and GSH at wavelengths of 535 nm and 412 nm, respectively, using a spectrophotometer.

Statistical analysis

Data entry, processing, and analysis were performed using the 26th edition of the Statistical Package for the Social Sciences (SPSS). One-way analysis of variance (ANOVA) and post hoc least significant difference testing were used to evaluate the study's findings. Independent samples t-test was used to compare the mean length of the tail between VPA and control groups. The results were shown as the standard deviation (LSD) and mean. P-values of ($p=< 0.05$) and ($p=< 0.001$) were used to classify differences as statistically significant and extremely significant, respectively.

Ethical approval

This study was approved by the committee of publication ethics at the College of Medicine/ University of Babylon, Iraq. A local ethics committee examined and approved the study protocol, subject information, and permission form, as indicated by document number 4-20, on 8/8/2023 to get this approval.

Results and Discussion

Results

1. Open field test

A. Line crossing

The number of line crossings was extremely significantly elevated ($p=<0.001$) in the VPA-Saline group in comparison with other groups and the number of line crossings was extremely significantly reduced ($p=<0.001$) in both high and low-dose VPA-*T. officinale* and VPA-

Risperidone groups as compared with the VPA-Saline group. The number of line crossings significantly declined (p= 0.015) in the Control-Risperidone group in contrast to the Control-Saline group. All these details are mentioned in **Figure 1**.

B. Latency time on the central zone

Latency time on the central zone was extremely significantly declined (p=<0.001)in the VPA-Saline group in comparison with other groups while Latency time on the central zone was extremely significantly raised (p=<0.001) in in both high and low dose VPA-*T. officinale* and VPA-Risperidone groups as compared with the VPA-Saline group. Latency time on the central zone was significantly elevated (p= 0.047) in low-dose Control- *T. officinale* as compared to the Control-Saline group. all these details are mentioned in **Figure 2**.

C. Grooming

The number of grooming was extremely significantly elevated (p=<0.001) in the VPA-Saline group as compared with other groups and was extremely significantly decreased (p<=0.001) in VPA-Risperidone and both high and low-dose VPA-*T. officinale.*The number of grooming was significantly declined in high dose VPA-*T. officinale* (p= 0.01) and (p= 0.027) in contrast to low dose VPA-*T. officinale* and VPA-Risperidone groups respectively. Every information described above was included in **Figure 3**.

D. Rearing

The number of rearing was extremely significantly reduced (p=<0.001) in the Control-Risperidone group as compared with Control-Saline and low-dose Control-*T. officinale* groups. The number of rearing was extremely significantly elevated (p=<0.001) in the VPA-Saline group as compared with other groups while the number of rearing was extremely significantly reduced (p=<0.001) in both high and low-dose VPA-*T. officinale* and VPA-Risperidone groups as compared with the VPA-Saline group. The number of rearing significantly declined (p= 0.037) in the high-dose VPA-*T.officinale* group as compared to the VPA-Risperidone group. Every information described above is included in **Figure 4**.

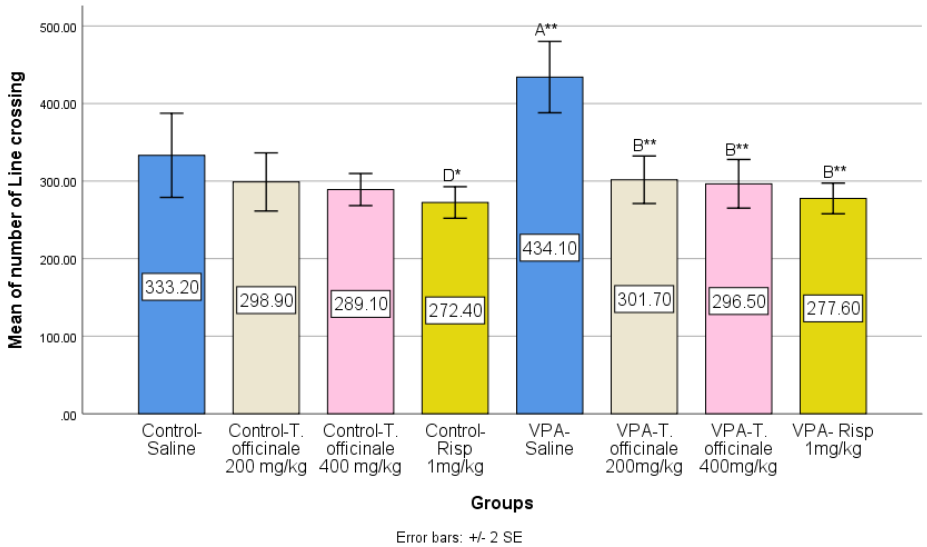

Figure 1: Effect of *Taraxacum officinale* and Risperidone on the number of line crossing in the open field test. *p= <0.05 (significant), **p= <0.001 (extremely significant). A: as compared with other groups, B: as compared to the VPA-saline group, and D: in comparison with the control saline group.

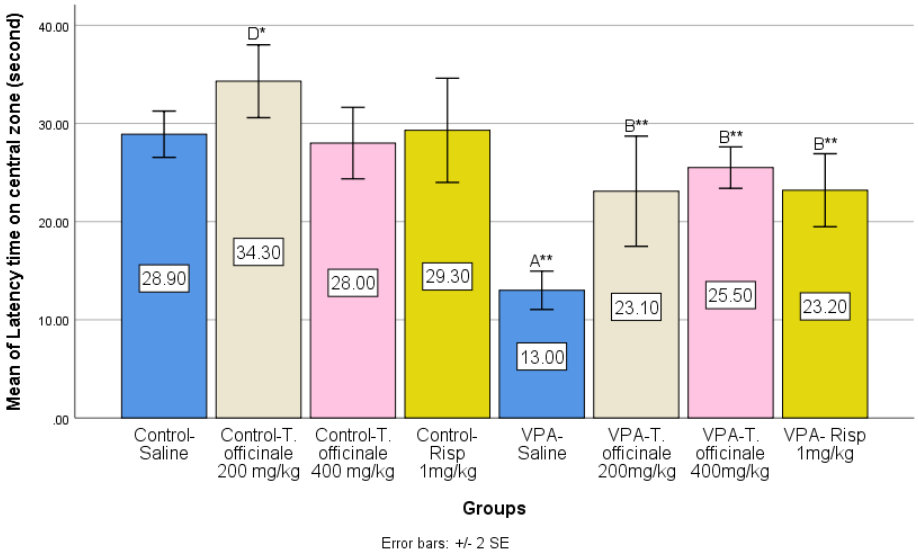

Figure 2: Effect of *Taraxacum officinale* and Risperidone on the latency time on the central zone in the open field test. *p= <0.05 (significant), **p=<0.001 (extremely significant). A: as compared with other groups, B: as compared to the VPA-saline group, and D: as compared to the Control-Saline group.

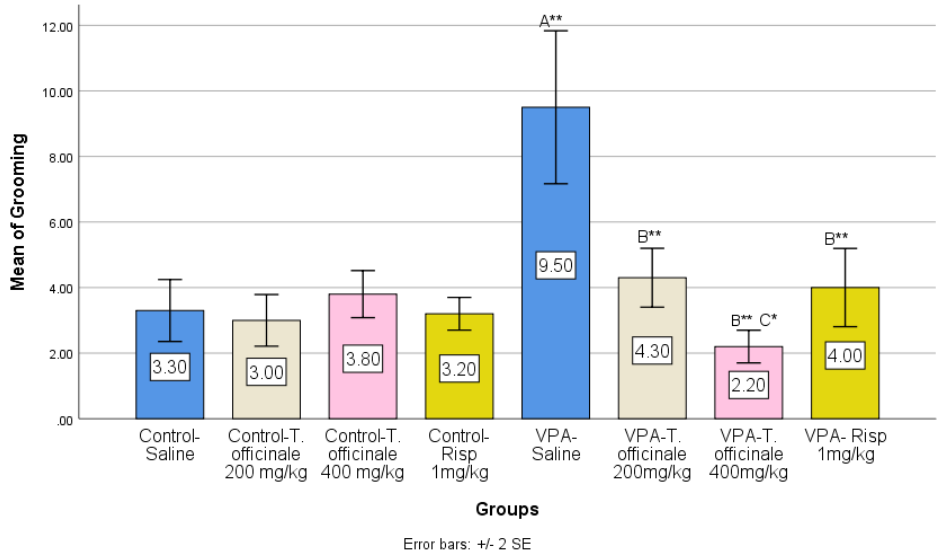

Figure 3: Effect of *Taraxacum officinale* and Risperidone on number of grooming in open field test. *p<=0.05 (significant), **p=<0.001 (extremely significant). A: as compared with other groups, B: as compared to the VPA-saline group, and C: in comparison with the VPA-risperidone group.

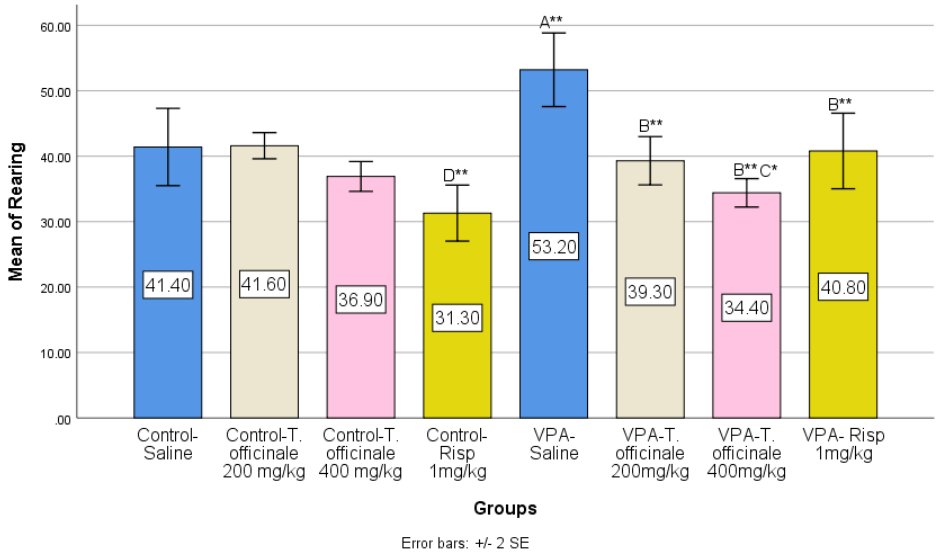

Figure 4: Effect of *Taraxacum officinale* and Risperidone on the number of rearing in an open field test. *p=<0.05 (significant), **p=<0.001 (extremely significant). A: as compared with other groups, B: as compared to the VPA-saline group, D: in comparison with the control saline group and C: in comparison with the VPA-risperidone group.

2. Oxidative stress assessment

A. Malondialdehyde (MDA)

The concentration of malondialdehyde was extremely significantly raised (p=<0.001) in the VPA-Saline group in contrast to other groups. The concentration of malondialdehyde was extremely significantly declined (p=<0.001) in both low and high-dose VPA-*T.officinale* and VPA-Risperidone groups. The level of malondialdehyde was significantly elevated (p= 0.003) in high-dose VPA-*T. officinale* group as compared with the VPA-Risperidone group while the level of malondialdehyde was significantly declined (p= 0.031) in high dose Control-T. officinale group as opposed to the Control-Saline group. All of the above details are described in **Figure 5**.

B. Reduced glutathione (GSH)

The level of reduced glutathione was extremely significantly reduced (p=<0.001) in the VPA-Saline group as compared with other groups while the level of reduced glutathione was extremely significantly raised in VPA-Risperidone and both low and high VPA-*T.officinale* groups. The concentration of reduced glutathione in high-dose VPA-*T. officinale* group was significantly elevated (p= 0.002) and extremely significantly elevated (p=<0.001) in comparison with VPA-Risperidone and low dose VPA-*T.officinale* groups respectively. The level of reduced glutathione significantly declined (p= 0.027) in the Control-Risperidone group as opposed to the Control-Saline group. all this information is described in **Figure 6**.

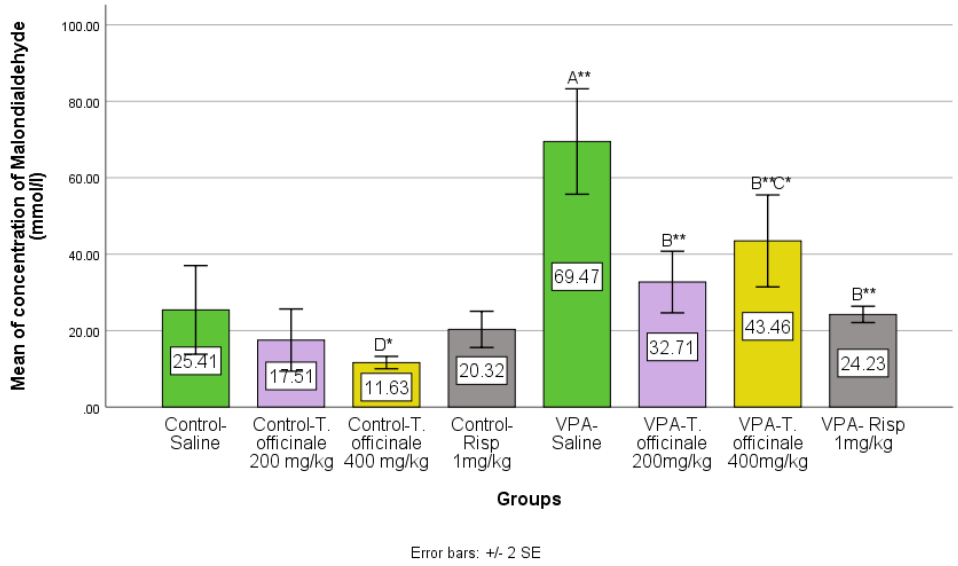

Figure 5: Effect of *Taraxacum officinale* and Risperidone on the concentration of malondialdehyde. *p=<0.05 (significant), **p=<0.001 (extremely significant). A: as compared with other groups, B: as compared to the VPA-saline group, D: in comparison with the control saline group and C: in comparison with the VPA-risperidone group.

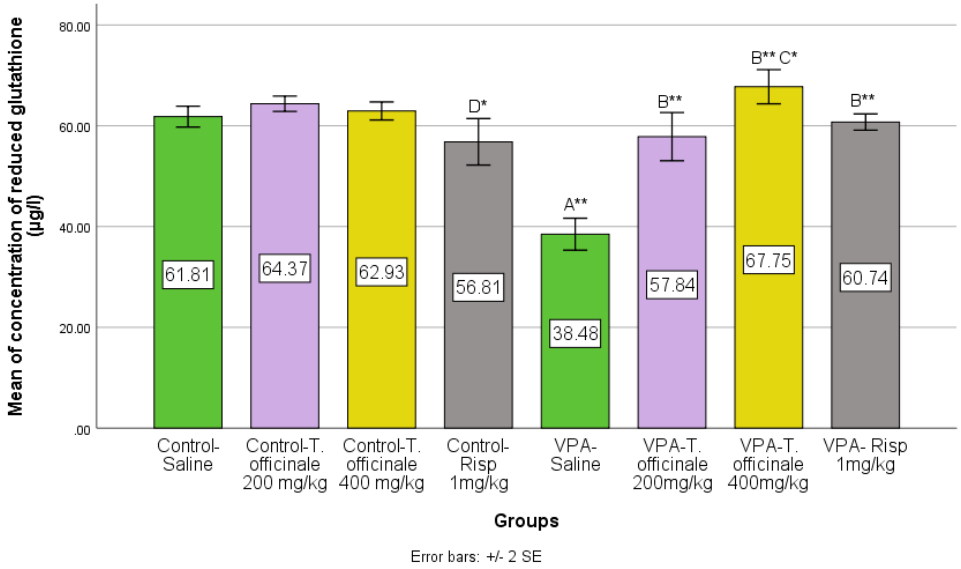

Figure 6: Effect of *Taraxacum officinale* and Risperidone on the concentration of reduced glutathione. *p<=0.05 (significant), **p=<0.001 (extremely significant). A: as compared with other groups, B: as compared to the VPA-saline group, D: in comparison with the control saline group and C: in comparison with the VPA-risperidone group.

3. Length of tail

The length of the tail was extremely significantly decreased (p=<0.001) in VPA groups as compared with Control groups as described in **Figure 7**.

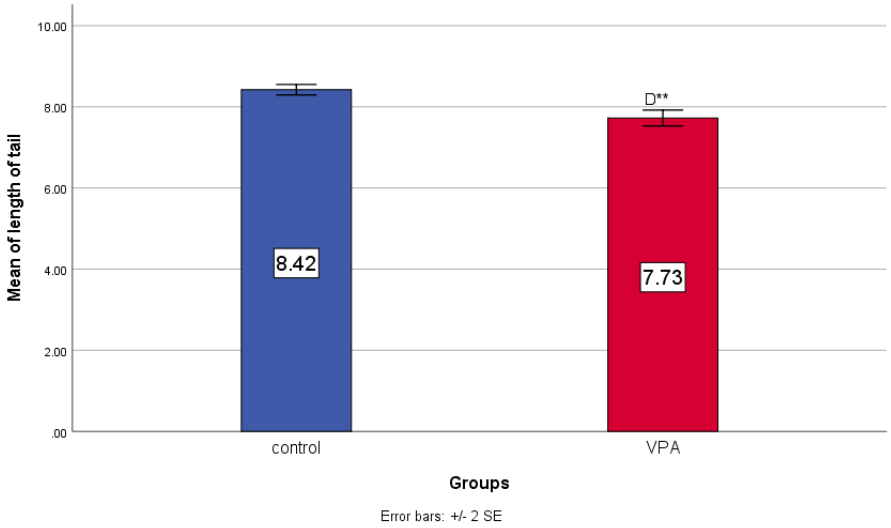

Figure 3.1: Effect of sodium valproate administration for pregnant mice on length of tail for offspring. P**=<0.001 (extremely significant). D: as compared to control.

Discussion

An open-field test is frequently used to evaluate overall motor activity, investigate behavior, and examine behavior related to anxiety. The study's findings demonstrated that, in comparison to the control group, offspring who were exposed to VPA during pregnancy traveled farther in an open field test, had higher mean values for grooming and rearing, and had lower mean values for latency time in the central zone. This outcome demonstrated a high degree of anxiousness and hyperactivity and concurred with the reports of earlier studies that show in the VPA model, the amygdala—a brain area involved in processing emotions like anxiety—plays a crucial role in moderating these recurrent behaviors. It has been demonstrated that amygdala stimulation causes rodents to become more anxious and more prone to compulsive behaviors [26,27]. The high and low dose VPA- *T. officinale* groups when compared with the VPA-Saline group show an extremely significant reduction in line crossing, rearing, and grooming while latency time in the central zone extremely increased. On the other hand in comparison with the VPA-Risperidone group, the high dose VPA- *T. officinale* group showed a significant reduction in mean of rearing and grooming. The low dose Control-*T. officinale* showed a substantial increase in latency time in the central zone compared to other groups. There is no previous research on the effect of *T. officinale* on open-field tests, the positive impact of T. officinale on open-field tests may be attributed to the modulation of neurotransmitters such as dopamine and through overexpression of Brain-Derived Neurotrophic Factor (BDNF) in the hippocampus leading to lowering anxiety and stereotyped behavior according to findings on other neurologic diseases [28,29]. The VPA-Risperidone group when compared with the VPA-Saline group showed an extremely significant reduction in rearing, grooming, and line crossing, while latency time in the central zone extremely increased. On the other hand, the control-risperidone group when compared to the Control-saline group showed a significant decline in a number of line crossings and an extremely significant reduction in the mean value of rearing. The risperidone's beneficial effects in open field tests result from antagonizing dopamine D2 receptors reducing hyperactivity and repetitive behaviors, while antagonizing the serotonin 5-HT2A receptor enhances cognitive inflexibility, a type of higher-order repetitive behavior. this outcome is consistent with earlier research [26,27].

In this study, oxidative stress was evaluated by measuring the levels of lipid peroxidation (MDA; an end product of lipid peroxidation) and the antioxidant enzyme activity such as GSH which is a major cellular radical scavenger. The finding of this research showed that the VPA-Saline group in comparison to the control group had an extremely significant rise in the level of lipid peroxidation (MDA) and an extremely significant decline in the level of reduced glutathione (GSH) indicating oxidative stress, these outcomes were in line with the previous research [30]. After administration of risperidone and *T. Officicinale* therapy to the control and VPA groups for 21 days, the level of MDA extremely significantly declined in VPA-*T. Offiicinale* and VPA-risperidone groups as compared to the VPA-Saline group, and the level of lipid peroxidation significantly reduced in the VPA-Risperidone group when compared with VPA-T. Officinale groups. These findings indicate that both risperidone and *T. Officinale* had powerful antioxidant effects with superior efficacy for risperidone. In regard to a control groups, the level of MDA significantly declined in contrast to the Control-Saline group [31,32].

A positive effect of *T. Officinale* on the level of MDA indicates it had great antioxidant activity and was mediated by different mechanisms such as direct ROS-scavenging, indirect ROS-scavenging (i.e., metal ion chelation), decreased lipid peroxidation, and increasing antioxidant capacity. The most important scavenging compounds are phytophenols (flavonoids and caffeoyl derivatives) and triterpenes (taraxasterol). This is in line with earlier research on other diseases [32,33,34]. Also, it has demonstrated that NADPH oxidase which generates reactive oxygen species (ROS) is suppressed by flavonoids [35]. In this study, the low-dose VPA- *T. Officinale* 200mg/kg group showed a better reduction in the level of MDA than the high-dose VPA-T. Officinale 400mg/kg group, this effect may be attributed to phytosterols that present in *T. Officinale* which are susceptible to oxidation and can change from antioxidants to pro-oxidants and vice versa due to their structural resemblance to cholesterol and by lowering the dose of *T. Officinale* from 400mg to 200mg giving better result, these finding in line with previous research [34].

Also, it was found that the level of GSH in VPA-*T. Officinale* and VPA-Risperidone groups were extremely significantly elevated as compared with the VPA-Saline group. The high dose VPA-*T. Officinale* 400mg/kg group showed a greater increase in the level of GSH when compared with the low dose VPA- *T. Officinale* and VPA-Risperidone groups which indicated that the effect of *T. Officinale* on reduced glutathione levels evolved in proportion to the *T. Officinale* dose; as the dose raised, so GSH levels were raised. The study's findings concurred with those of previous investigations [36,34]. The level of GSH in the Control-risperidone group significantly declined in contrast to the Control-Saline group, this may be attributed to the toxicity of risperidone in the control group, it has been demonstrated to cause oxidative stress by raising the generation of reactive oxygen species (ROS), which causes the breakdown of lysosomal membranes, mitochondrial collapse, and GSH depletion, these results in line with previous research on other cells other than brain [37]. To properly understand the intricate connection between risperidone and oxidative stress in the brain, more research is required.

The results of this study demonstrated that, in comparison to Control groups, the length of the tail was considerably ($p=>0.001$) shorter in VPA groups. This result is consistent with previous research [13 Anna Maria Tartaglione]. These anomalies are a result of neural tube defects (NTDs), and since VPA is known to create NTDs, VPA's direct teratogenic effect

may be what causes these anomalies rather than epigenetic changes. These findings were consistent with previous studies [12,38]. It showed that the teratogenic effects of VPA are only seen in the first generation, suggesting that skeletal issues are not inherited by subsequent generations.

Conclusions

These findings demonstrated that mice's tail length decreased when exposed to 600 mg/kg of sodium valproate during pregnancy compared to control mice. TOE exhibited anxiolytic activity, decreased hyperactivity, and enhanced latency duration in the core. It demonstrated superior outcomes than risperidone in lowering grooming and rearing in VPA mice during the open field test. TOE had antioxidant activity by decreasing MDA level with superior efficacy to risperidone in VPA- offspring, and at a dose of 400mg/kg it increased the level of GSH in VPA- offspring more than risperidone did. Lastly, T. Officinale at both doses decreased lipid peroxidation in control groups better than risperidone.

References

1. Yanjie Yu , Ashmita Chaulagain , Sindre Andre Pedersen , Stian Lydersen , Bennett L. Leventhal , Peter Szatmari , Branko Aleksic , Norio Ozaki and Norbert Skokauskas. 2020. Pharmacotherapy of restricted/repetitive behavior in autism spectrum disorder: a systematic review and meta-analysis. BMC Psychiatry; (2020) 20:121.
2. Mohammed Moutaz Nakha, Petrilla Jayaprakash, Salahdein Aburuz, Bassem Sadek and Amal Akour. 2023. Canagliflozin Ameliorates Oxidative Stress and Autistic-like Features in Valproic-Acid-Induced Autism in Rats: Comparison with Aripiprazole Action. Pharmaceuticals; 2023, 16, 769.
3. Luca Pangrazzi, Luigi Balasco and Yuri Bozzi. 2020. Oxidative Stress and Immune System Dysfunction in Autism Spectrum Disorders. International Journal of Molecular Sciences. (2020) 21: 3288.
4. Xukun Liu, Jing Lin, Huajie Zhang, Naseer Ullah Khan, Jun Zhang, Xiaoxiao Tang, Xueshan Cao and Liming Shen. 2022. Oxidative Stress in Autism Spectrum Disorder—Current Progress of Mechanisms and Biomarkers. Frontiers in Psychiatry; (2022) 13:813304.
5. Charlotte Lauridsen. 2019. From oxidative stress to inflammation: redox balance and immune system. Poultry Science (2019) 98:4240–4246.
6. Geir Bjørklund, NagwaA.Meguid, Mona A. El-Bana, Alexey A. Tinkov, Khaled Saad8, Maryam Dadar, Maha Hemimi, Anatoly V. Skalny, Božena Hosnedlová, Rene Kizek, Joško Osredkar, Mauricio A. Urbina, Teja Fabjan, Amira A. El-Houfey, Joanna Kałużna-Czaplińska, Paulina Gątarek, and Salvatore Chirumbolo. 2020. Oxidative Stress in Autism Spectrum Disorder. Molecular Neurobiology. (2020) 57, 2314–2332.
7. Fiona Hollis, Alexandros K Kanellopoulos, and Claudia Bagni. 2017. Mitochondrial dysfunction in Autism Spectrum Disorder: clinical features and perspectives. Current Opinion in Neurobiology. (2017) 45:178–187.
8. Geir Bjørklund, Richard E. Frye, Monica Daniela Dosa, Massimiliano Peana h, Michael Maes, Maryam Dadar, Salvatore Chirumbolo. 2021. The impact of

glutathione metabolism in autism spectrum disorder. Pharmacological Research 166 (2021) 105437.
9. Geir Bjørklunda, Alexey A. Tinkov, Božena Hosnedlová, Rene Kizek, Olga P. Ajsuvakova, Salvatore Chirumbolo, Margarita G. Skalnaya, Massimiliano Peana, Maryam Dadar, Afaf El-Ansary, Hanan Qasem, James B. Adams, Jan Aaseth, Anatoly V. Skalny. 2020. The role of glutathione redox imbalance in autism spectrum disorder: A review. Free Radical Biology and Medicine; 160: 149_162.
10. Mehmet Akif Camkurt, Ebru Fındıklı, Filiz İzci, Ergül Belge Kurutaş and Taha Can Tuman. 2016. Evaluation of Malondialdehyde, Superoxide Dismutase and Catalase Activity and Their Diagnostic Value in Drug Naïve, First Episode, Non-Smoker Major Depression Patients and Healthy Controls. Psychiatry Research; 238 (2016) 81-85.
11. Oula Nasrallah and Samar Alzeer. 2022. Measuring Some Oxidative Stress Biomarkers in Autistic Syrian Children and Their Siblings: A Case-Control Study. Biomarker Insights; 17 (2022) 1–8.
12. Anna Maria Tartaglione, Chiara Cipriani, Flavia Chiarotti, Benedetta Perrone, Emanuela Balestrieri, Claudia Matteucci, Paola Sinibaldi-Vallebona, Gemma Calamandrei and Laura Ricceri. Early Behavioral Alterations and Increased Expression of Endogenous Retroviruses Are Inherited Across Generations in Mice Prenatally Exposed to Valproic Acid. Molecular Neurobiology; 2019, 56, 3736–3750.
13. Maria Jimena Salcedo-Arellano, AnaMariaCabal-Herrera, Ruchi Harendra Punatar, Courtney Jessica Clark, Christopher Allen Romney and Randi J Hagerman. 2021. Overlapping Molecular Pathways Leading to Autism Spectrum Disorders, Fragile X Syndrome, and Targeted Treatments. Neurotherapeutics; (2021) 18:265–283.
14. Beata Olas. 2022. New Perspectives on the Effect of Dandelion, Its Food Products and Other Preparations on the Cardiovascular System and Its Diseases. Nutrients; 2022, 14, 1350.
15. Shan Huang, Ning Meng, Zhiming Liu, Li Guo, Linsha Dong, Bin Li ID and Qiang Ye. 2018. Neuroprotective Effects of Taraxacum officinale Wigg. Extract on Glutamate-Induced Oxidative Stress in HT22Cells via HO-1/Nrf2 Pathways. Nutrients; 2018, 10, 926.
16. Małgorzata Kania-Dobrowolska and Justyna Baraniak. 2022. Dandelion (Taraxacum officinale L.) as a Source of Biologically Active Compounds Supporting the Therapy of Co-Existing Diseases in Metabolic Syndrome. Foods; 2022, 11, 2858.
17. Zain Ul Aabideen, Muhammad Waseem Mumtaz, Muhammad Tayyab Akhtar Hamid Mukhtar, Syed Ali Raza, Tooba Touqeer, and Nazamid Saari. 2020. Anti-Obesity Attributes; UHPLC-QTOF-MS/MS-Based Metabolite Profiling and Molecular Docking Insights of Taraxacum officinale. Molecules; 2020, 25, 4935.
18. Dariusz Je drejek, Bogdan Kontek, Bernadetta Lis, Anna Stochmal, and Beata Olas. 2017. Evaluation of antioxidant activity of phenolic fractions from the leaves and petals of dandelion in human plasma treated with H2O2 and H2O2/Fe. Chemico-Biological Interactions; 262 (2017) 29-37.

19. Sunday Aderemi Adelakun, Aniah Julius Akomaye, Olusegun Dare Omotoso, and Olukayode Abimbola Arowosegbe. 2024. Anti-hepatopathy and anti-nephropathy activities of Taraxacum officinale in a rat model of Streptozotocin diabetes-induced hepatorenal toxicity and dyslipidemia via attenuation of oxidative stress, inflammation, apoptosis, electrolyte imbalances, and mitochondrial dysfunction. Aspects of Molecular Medicine; 3 (2024) 100034.
20. Petra Horka, Veronika Langova, Jan Hubeny, Karel Vales, Ivana Chrtkova and Jiri Horacek. 2024. Open field test for the assessment of anxiety-like behavior in Gnathonemus petersii fish. Frontiers in Behavioral Neuroscience; 17 (2024) 1280608.
21. Michael Lough-Stevens, Caleb R. Ghione, Matthew Urness, Adelaide Hobbs, Colleen M. Sweeney and Matthew D. Dean. Male-derived copulatory plugs enhance implantation success in female Mus musculus. Biology of Reproduction 2021; 104(3): 684–694.
22. Katarine Fereshetyan , Vergine Chavushyan , Margarita Danielyan , KonstantinYenkoyan. 2021. Assessment of behavioral, morphological and electrophysiological changes in prenatal and postnatal valproate induced rat models of autism spectrum disorder—Scientific Reports; (2021) 11:23471.
23. Ui-Jin Kim, NamgueHong and andJin-ChulAhn. Photobiomodulation Attenuated Cognitive Dysfunction and Neuroinflammation in a Prenatal Valproic Acid-Induced Autism Spectrum Disorder Mouse Mode. International Journal of Molecular Sciences 2022; 23: 16099.
24. Nithiyaa Perumal, Meenakshii Nallappan, Shamarina Shohaimi, Nur Kartinee Kassim, Thiam Tsui Tee and Yew Hoong Cheah. Synergistic antidiabetic activity of Taraxacum officinale (L.) Weber ex F.H. Wigg and Momordica charantia L. polyherbal combination. Biomedicine & Pharmacotherapy 2022; 145: 112401.
25. Fatemeh Piri, Mahmoud Elahdadi Salmani, and Hamid Sepehri. Improvement of autistic-like behaviors in adult rats prenatally exposed to valproic acid through early suppression of orexin receptor. Annals of Medicine & Surgery 2024; 86: 166–171.
26. Viktor Román, Nika Adham, Andrew G. Foley, Lynsey Hanratty, Bence Farkas, Balázs Lendvai and Béla Kiss. 2021. Cariprazine alleviates core behavioral deficits in the prenatal valproic acid exposure model of autism spectrum disorder. Psychopharmacology; (2021) 238:2381–2392.
27. Tanya Gandhi and Charles C. Lee. 2021. Neural Mechanisms Underlying Repetitive Behaviors in Rodent Models of Autism Spectrum Disorders. Frontiers in Cellular Neuroscience; (2021) 14:592710.
28. Cunyou Gao, Suli Kong, Benyu Guo, Xuejun Liang, Huifeng Duan and Donghe Li. 2019. Antidepressive Effects of Taraxacum Officinale in a Mouse Model of Depression Are Due to Inhibition of Corticosterone Levels and Modulation of Mitogen-Activated Protein Kinase Phosphatase-1 (Mkp-1) and Brain-Derived Neurotrophic Factor (Bdnf) Expression. Medical Science Monitor; (2019) 25: 389-394.
29. Tatiana Ilchibaeva , Anton Tsybko, Marina Lipnitskaya, Dmitry Eremin, Kseniya Milutinovich, Vladimir Naumenko and Nina Popova. 2023. Brain-Derived Neurotrophic Factor (BDNF) in Mechanisms of Autistic-like Behavior in BTBR

Mice: Crosstalk with the Dopaminergic Brain System. Biomedicines; 2023, 11, 1482.
30. Esraa M. Elnahas, Sally A. Abuelezz, Magda I. Mohamad, Mai M. Nabil, Sahar M. Abdelraouf, Nevine Bahaa, Ghada A.M. Hassan, Eman A. Ibrahim, Asmaa I. Ahmed and Sawsan Aboul Fotouh. 2020. Validation of prenatal versus postnatal valproic acid rat models of autism: A behavioral and neurobiological study. Progress in Neuro-Psychopharmacology and Biological Psychiatry; (2020) 108:110185.
31. Md. Mamun Al-Amin, Md. Faiyad Rahman Choudhury, Al Saad Chowdhury, Tahsinur Rahman Chowdhury, Preeti Jain, Mohsin Kazi, Musaed Alkholief, Sultan M. Alshehri and Hasan Mahmud Reza. Pretreatment With Risperidone Ameliorates Systemic LPS-Induced Oxidative Stress in the Cortex and Hippocampus. Frontiers in Neuroscience; 2018, 12:384.
32. Alexandra Epure, Alina E. Pârvu, Laurian Vlase, Daniela Benedec, Ovidiu Oniga, Ana-Maria Vlase, Irina Ielciu, Anca Toiu, Daniela Hanganu and Ilioara Oniga. 2023. New Approaches on the Anti-Inflammatory and Cardioprotective Properties of Taraxacum officinale Tincture. Pharmaceuticals; (2023) 16, 358.
33. Michał Majewski, Bernadetta Lis, Jerzy Juskiewicz, Katarzyna Ognik, Dariusz Jedrejek, Anna Stochmal and Beata Olas. 2021. The composition and vascular/antioxidant properties of Taraxacum officinale flower water syrup in a normal-fat diet using an obese rat model. Journal of Ethnopharmacology; 265 (2021) 113393.
34. Iulia Olimpia Pfingstgraf, Marian Taulescu, Raluca Maria Pop, Remus Orăsan, Laurian Vlase, Ana Uifalean , Doina Todea, Teodora Alexescu, Corina Toma and Alina Elena Pârvu. 2021. Protective Effects of Taraxacum officinale L. (Dandelion) Root Extract in Experimental Acute on Chronic Liver Failure. Antioxidants; (2021) 10, 504.
35. Olukayode O. Aremu, Adebola O. Oyedeji, Opeoluwa O. Oyedeji, Benedicta N. Nkeh-Chungag and Constance R. Sewani Rusike. 2019. In Vitro and In Vivo Antioxidant Properties of Taraxacum o cinale in N-Nitro-l-Arginine Methyl Ester (L-NAME)-Induced Hypertensive Rats. Antioxidants; 2019, 8, 309.
36. Fatima Zahra Kamal, Radu Lefter , Cosmin-Teodor Mihai, Hanane Farah, Alin Ciobica, AhmadAli, Iulian Radu, Ioannis Mavroudis and Abdellah Ech-Chahad. 2022. Chemical Composition, Antioxidant and Antiproliferative Activities of Taraxacum officinale Essential Oil. Molecules; 2022, 27, 6477.
37. Jaiyeola Abiola Kajero, Soraya Seedat, Jude Ohaeri, Abidemi Akindele and Oluwagbemiga Aina. 2022. Effects of cannabidiol on vacuous chewing movements, plasma glucose and oxidative stress indices in rats administered high dose risperidone. Scientific Reports; 2022, 12:19718.
38. Chang Soon Choi, Edson Luck Gonzales, Ki-Chan Kim, Sung Min Yang, Ji-Woon Kim, Darine Froy Mabunga, Jae Hoon Cheong, Seol-Heui Han, Geon Ho Bahn, and Chan Young Shin. The transgenerational inheritance of autism-like phenotypes in mice exposed to valproic acid during pregnancy. Scientific Reports; 2016, 6:36250.