Biol Trace Elem Res (2013) 151:181–186
DOI 10.1007/s12011-012-9553-z

Levels of Heavy Metals and Essential Minerals in Hair Samples of Children with Autism in Oman: a Case–Control Study

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Received: 29 September 2012 / Accepted: 12 November 2012 / Published online: 28 November 2012

Abstract Toxic levels of heavy metals and low levels of essential minerals have been suggested to play a critical role in the pathogenesis of autism spectrum disorders (ASD). This study documents the levels of heavy metals and essential minerals in hair samples of children with ASD in Muscat, the urbanized capital of Oman, Muscat. The study included 27 children with ASD and 27 matched non-ASD controls. Parental interviews were held and dietary intake questionnaires completed in conjunction with the collection of hair samples. Analysis of heavy metals and essential minerals was carried out by inductively coupled plasma mass spectrometry. Chi-square analysis and non-parametric Fisher's exact tests were used to assess statistical significance. Children with ASD had significantly higher levels of all 11 analyzed heavy metals in their hair samples ($P<0.05$), ranging from 150 to 365 % of control levels. ASD children also had significantly higher levels of essential minerals sulfur, sodium, magnesium, potassium, zinc, and iron, but lower levels of calcium and copper in their hair samples. This study corroborates data from previous studies in different parts of the world indicating the presence of elevated levels of heavy metals and selective depletion of essential minerals in the hair of children with ASD.

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Keywords Autism · Heavy metals · Essential minerals · Hair · Oman · Case–control

Background

Autism spectrum disorders (ASDs) are a group of neurodevelopmental disorders characterized by impaired psychosocial and adaptive skills functioning that has a direct bearing on social interaction, communication skills, and patterns of activity [1]. Prevalence rates of ASDs range from 1 to 20 per 1,000 people worldwide [2, 3]. Although partially attributed to improved diagnosis accuracy and clinical awareness [4], the magnitude of children diagnosed with autism has dramatically increased in recent years [5–7]. With the increasing number of children with ASD in many parts of the world, the interplay between nurture and ecological and biological factors in ASD pathogenesis appears to be important [8, 9].

Few studies have explored the levels of heavy metals and essential minerals among children diagnosed with ASD in Arabian Gulf countries [10, 11]. Preliminary surveys have suggested that this region has a significant number of children with ASD; the rate has been shown to range from 29/10,000 in the United Arab Emirates [12] to 1.4/10,000 in Oman [13].

Despite its global burden, attempts to pinpoint possible pathophysiological mechanisms for ASD have led to contradictory conclusions. Among numerous factors linked to the development of ASD is the contribution of environmental toxins, acting as neurotoxins to the central nervous system and adversely affecting prenatal development [14, 15]. Various birth defects and fetal developmental abnormalities have been linked to environmental toxins [16–18] , and studies from different parts of the world have documented possible adverse roles of heavy metals in the development and expression of ASD [19–22]. Essential minerals are crucially important for the proper functioning of biological systems [23], and dysregulation of their levels may contribute to the development of ASD [24, 25]. In support of such a view, a study in Venezuela [26] documented an inverse relation between either an "excess or a deficiency" of essential minerals, which paralleled variations in ASD severity. Levels of the trace elements copper, zinc, magnesium, and selenium in hair and nail samples of ASD patients from South India strongly correlated with the severity of the disease [27]. Studies from other parts of the world generated similar results [28–30]. Given the aforementioned link between heavy metal toxicity, disturbance of essential minerals, and ASD, the present study explored whether there are variations in hair levels of heavy metals and essential minerals content in Omani children diagnosed with ASD and those without. The present study tested the hypothesis that children with ASD will show variations and deregulated levels of heavy metals and essential minerals when compared to non-ASD controls.

Methods

The study was conducted over the period from December 2009 to August 2010 among children and adolescents seeking consultation at the Child Psychiatry outpatient clinic of Sultan Qaboos University Hospital (SQUH). All consenting (from accompanying parent) participants (n=27), 3–14 years, who fulfilled the criteria for diagnosis of ASD according to threshold defined by *Childhood Autism Rating Scale* [31] and *Diagnostic and Statistical Manual of Mental Disorders*, *Fourth Edition*, *Text Revision* [1] were invited to participate in this study. Each participant's mother was interviewed one-on-one. Interview questions were designed to elicit information regarding the child's developmental milestones, socio-demographic background, and dietary history, in addition to other risk factors that might indicate a predisposition to autism, including pregnancy complications, preterm delivery, breastfeeding, and family history of autism. To overcome the problem of "reverse causality"— that autistic children might have changed their social or dietary habits after diagnosis—only recently diagnosed cases were included, for which the dietary intake questionnaire was administered within 10–14 days of confirming the diagnosis.

In addition to those who fulfilled criteria for diagnosis of ASD, participants with overall similarity in terms of age, gender, and ethnicity were recruited as controls. Control subjects were randomly selected from eligible outpatients at the Department of Child Health at SQUH. Inclusion criteria included those seeking consultation for trauma, routine physical examination, dental problems, and dermatological problems. Further exclusion criteria for the control group included the presence of any overt neurodevelopmental or behavioral disturbances or history of pervasive and persistent malnutrition. Pediatricians and a family physician were involved in ruling out the possible presence of any sub-clinical autistic features in the control group, and controls had no known characteristics of ASD.

The accompanying family members for both groups (children with ASD and control) were explicitly informed that their participation in this study was purely voluntary and their participation or otherwise would not hamper their care seeking at this tertiary care hospital. The study was approved by the Medical Research Ethics Committee of Sultan Qaboos University. All caregivers of participants provided their informed consent to be included in the study.

The present protocol for hair sampling has been detailed elsewhere [25]. To recapitulate, all measurements were conducted on a Perkin–Elmer mass spectrometer (Sciex Elan 6100), standardized by a regression method by means of a four-point calibration curve using commercially available stock standard solutions. The accuracy of the study calibration and the entire methodology was verified by the appropriate use of reagent blanks, independent calibration verification standard check solutions, and pooled hair specimens, pooled hair aqueous solutions, and other hair material with established reference ranges. Hair sampling has several advantages compared to blood sampling. Hair specimen has high validity in determining the state of a given mineral in the body [32]. In addition, it is a non-invasive procedure, and therefore, hair specimen can be collected more easily than blood specimen. It does not require a professional medic to collect it, and it is generally resistant to contamination compared to blood or urine specimens.

Chi-square analyses were used to evaluate the statistical significance of differences among portions of categorical data. The non-parametric Fisher's exact test (two tailed) was used instead of the Chi-square test for small sample sizes where the expected frequency was less than five in any of the two by two table cells. For measurements of heavy metals, median and inter-quartile ranges were used as measures of location and dispersion because of a relatively large variation in values. The non-parametric Mann–Whitney test was used to ascertain any significant differences between the mean values of two continuous variables. All statistical analyses were performed using the Statistical Package for Social Sciences (SPSS) software (Version 19.0, IBM, Chicago, Illinois, USA); a cut-off P value of <0.05 was the threshold of statistical significance for all tests.

Results

A total 54 children, 27 children with ASD and 27 controls, participated in this study. The male-to-female ratio was approximately 4:1 for ASD subjects and 3:1 for control subjects. The average age was not significantly different between children with ASD and controls. Selected socio-demographic characteristics of all subjects at the time of enrollment in the study are summarized in Table 1.

Biochemical analysis of heavy metal levels in hair samples revealed statistically significant differences for each of the metals analyzed ($p<0.05$), with ASD children exhibiting higher levels in their hair samples compared to non-ASD controls (Table 2). The extent of the increase varied from 50 % for boron, to 265 % for molybdenum.

When compared to controls, children with ASD had significantly higher levels of several essential minerals (sulfur, sodium, magnesium, potassium, zinc, and iron) in their hair samples (Table 3). In contrast, calcium and copper levels were significantly higher among controls ($p<0.05$), while levels of phosphorous and manganese were not significantly different. It is worth mentioning that sulfur is not an essential mineral,

Table 1 Socio-demographic characteristics of ASD cases and control group, Oman, 2010

Characteristics	Cases (*N*=27) *N* (%)	Controls (*N*=27) *N* (%)	*P* value
Gender			0.51
Male	22 (81.5)	20 (74.1)	
Female	5 (18.5)	7 (25.9)	
Mean age (year)	5.3 (1.5)	5.5 (1.4)	0.84
Mean birth weight (kg)			
2.50–3.99	23 (85.2)	21 (77.8)	0.12
<2.50	3 (11.1)	5 (18.5)	
>3.99	1 (3.7)	1 (3.7)	
Area of residence			0.31
Urban	19 (70.4)	17 (63.0)	
Rural	8 (29.6)	12 (44.4)	
Monthly family income (OR)			0.39
Less than 500	11 (40.7)	9 (33.3)	
500 to 1,000	10 (37.0)	12 (44.4)	
Greater than 1,000	6 (22.2)	6 (22.2)	
Educational level of mother			0.24
Illiterate	3 (11.1)	5 (18.5)	
Basic education	15 (55.6)	14 (51.9)	
Finished high school	9 (33.3)	8 (29.6)	
Occupation of mother			0.31
Working	10 (37.0)	8 (29.6)	
Housewife/retired	17 (63.0)	19 (70.4)	

Table 2 Comparison of levels of heavy metals in hair samples of cases with ASD and controls, Oman, 2010

Elements (unit)	Cases (*n*=27) Median (quartile)	Controls (*n*=27) Median (quartile)	*P* value
Lead (Pb)	12.2 (1.1)	6.3 (0.8)	0.03
Aluminum (Al)[a]	2.3 (0.3)	1.2 (0.2)	0.002
Silicon (Si)	90.1 (11.1)	53.3 (6.9)	0.02
Molybdenum (Mo)	8.4 (1.7)	2.3 (0.6)	0.01
Vanadium (V)	6.3 (0.7)	2.8 (0.4)	0.002
Chromium (Cr)	23.6 (2.3)	9.9 (1.6)	0.001
Cadmium (Cd)	7.4 (0.9)	3.0 (0.4)	0.001
Cobalt (Co)	14.7 (1.7)	6.1 (0.9)	0.001
Nickel (Ni)	11.6 (0.89)	5.6 (0.7)	0.003
Boron (B)[a]	2.1 (0.2)	1.4 (0.2)	0.003
Barium (Ba)[a]	8.4 (0.9)	4.8 (0.6)	0.003

Unit: microgram per gram of hair)

[a] Per 100 unit of measurement

Table 3 Comparison of levels of essential minerals in hair samples of cases with ASD and controls, Oman, 2010

Elements	Cases (*n*=27) Median (quartile)	Controls (*n*=27) Median (quartile)	*P* value
Sulfur (S)[a]	1,816.9 (45.9)	1,686.8 (50.8)	0.02
Phosphorus (P)	63.2 (6.7)	74.3 (4.8)	0.14
Sodium (Na)[a]	47.4 (3.2)	30.5 (2.2)	0.006
Magnesium (Mg)[a]	4.3 (0.7)	1.8 (0.2)	0.001
Potassium (K)[a]	16.8 (1.2)	11.6 (0.7)	0.007
Calcium (Ca)[a]	1.66 (1.1)	8.9 (0.7)	0.0001
Zinc (Zn)[a]	5.4 (0.82)	2.9 (2.2)	0.0001
Copper (Cu)	1.2 (0.1)	6.6 (0.7)	0.02
Manganese (Mn)	0.82 (0.2)	0.90 (0.1)	0.93
Iron (Fe)	91.3 (7.6)	46.3 (4.9)	0.0001

Unit: microgram per gram of hair)

[a] Per 100 unit of measurement

yet it was included in the analysis as it is an essential nutrient to humans.

Discussion

This study examined and compared the levels of essential minerals and heavy metals among Omani children with ASDs and those without. Improper levels of both of these factors have been suggested as possible contributors to autism. Our results are consistent with this suggestion, reinforcing awareness of the importance of proper nutritional support and avoidance of exposure to hazardous environmental toxins, especially during early development.

Several lines of evidence from the literature support the rationale of the present study to explore the role of heavy metals in the pathogenesis of Omani children with ASD. Indeed, recent studies have suggested that the country has a substantial number of children with ASD. Diagnosed ASD cases were estimated to be 1.4/10,000, but this may only be the tip of the iceberg, since many cases are believed to be undiagnosed and unreported [13]. Within the background of an emerging epidemiological trend for increasing ASD rates, Oman, and the region in general, may not be immune to the hazardous effects of heavy metals. Firstly, Oman has witnessed a rapid economic industrialization. The economic expansion and massive development of "free ports/zones" in the regions of countries adjacent to Oman have heralded it to be the "hub" for industrialization, manufacturing, transnational shipment as well as for a construction boom. Potentially toxic metals are generally found in chemical products, fertilizers, industrial paint, and building materials which are in increasing use. Toxic metals are an integral part of health provider services, including dental fillings, as well as vaccine adjuvants and preservatives used in various drug solutions. Children exposed to such potentially hazardous metals have been documented with unacceptably high heavy metal levels [18–22]. Extant empirical data suggests that high levels of heavy metals or their byproducts are found in certain species of seawater shellfish, fresh water fish, art supplies, processed food, fertilizers, and batteries [21, 33, 34]. Secondly, the Middle East has been a region in political turmoil over the last few decades, with several wars occurring in the region, including the First and Second Gulf Wars. There are speculations that the influx of military infrastructure and the resultant wars have exposed the region to environmental toxins [35, 36].

Within such background, the present study assessed the presence of lead, aluminum, silicon, molybdenum, vanadium, chromium, cadmium, cobalt, nickel, boron, and barium. Compared to children with non-diagnosis of ASD, the indices of these heavy metals were qualitatively higher in the hair of children with ASD. This study, unlike previous ones [21, 24, 25], has identified a wide spectrum of heavy metals to be significantly higher among children with ASD, further advancing the idea that heavy metals are strongly present in the biological system of children with ASD. It remains to be seen whether such presence is just a coincidence or an epiphenomenon, or whether it reflects an integral part of the initiating pathology or adaptation to having ASD.

Several factors might play a role in explaining the observed differences in levels of heavy metals and essential minerals. It is expected that children raised in the same environment in the Arabian Gulf have similar pattern of exposure to heavy metals and essential minerals. Therefore, the differences observed between ASD and non-ASD children might not be simply a result of excessive exposure to toxins, but it might be due to higher retention or probably higher absorption. There is increasing evidence that children with ASD show a marked inability to process and eliminate toxins from the body [27]. The accumulation of toxins leads to increased free radical activity in the body which eventually negatively affects the structure of the nervous system. Notably, detoxification and excretion of heavy metals relies upon their conjugation to glutathione, whose levels are significantly reduced in ASD subjects [37]. According to Al-Ayadhi [11], if multiple toxic exposures can be considered as a contributing factor that triggers ASD, prospective mothers could be screened for heavy metals exposure prior to conception. To our knowledge, such diagnostic evaluations have not been considered in medical practice in the region. Therefore, such a practice needs to be considered in order to mitigate the possible role of heavy metals in the pathogenesis of ASD.

Our second aim was to examine the levels of essential minerals among children with ASD and compare their level to neurotypical children. Previous studies in Oman have documented malnutrition and deficiencies in folate and vitamin B12 among children diagnosed with ASD [3, 38, 39], and these findings have sparked interest to explore the integrity of other dietary minerals that are vital for the proper cellular function. This study focused on sulfur, phosphorus, sodium, magnesium, potassium, calcium, zinc, copper, manganese, and iron, and the results showed that children with ASD displayed markedly lower levels of calcium and copper, but levels of sulfur, sodium, magnesium, potassium, zinc, and iron were higher. The present finding is generally consistent with previous reports of abnormal essential mineral levels in ASD [25, 40–43], although it has been suggested that their occurrence may be an artifact of ASD [44, 45]. Our results differ from the study by Abdullah et al. [24] which observed lower manganese levels. Their study also found no statistically significant difference between these two groups of children in phosphorus. As there is interplay between the function of calcium and phosphorus, it is possible that a reduction in calcium may have heightened presently observed phosphorus functioning [46]. The lower levels of copper and higher levels of iron we found may have particular significance for promoting oxidative stress. Since children with ASD are characterized with poor eating habits, our results may be explained by an inadequate nutritional intake which, in turn, has implications for potential dietary intervention.

Several limitations of our study must be acknowledged, and the inferences that can be taken from these findings should be made with caution. The present results remain limited by the available information. In particular, detailed data on possible sources of exposure to heavy metals in the environment were lacking. The results would have been more comprehensive if metal testing was also performed on mothers of concerned children. Furthermore, the inferences of this study were made on a relatively small sample size: only 27 children with ASD and a comparable number of controls. Data analysis might have been more robust if a larger sample size had been considered. As noted earlier, one intractable issue pertinent to these types of study is to rule out whether the presence of toxic material in the hair is a reflection of pathology or a reaction or adaptation to some inherent mechanisms that are central to ASD. The present case–control study is not equipped to shed light on such a complex issue. Finally, previous research geared towards deciphering the “etiology” of ASD is rife with claims and counterclaims that were detrimental for reaching evidence-based enlightened views on ASD causation. Despite the above-mentioned caveats, to our knowledge, this is the first study to link levels of heavy metals and essential minerals in hair samples of Omani children with autism, and our findings largely substantiate studies from other parts of the world.

Conclusions

The present study demonstrated toxic heavy metal exposure and nutritional inadequacies in children with ASD as compared to children with no cardinal symptoms of ASD. It would be premature to conclude that levels of heavy metals and essential minerals in hair samples of Omani children with autism constitute a major component in the pathophysiology of ASD. Therefore, more studies to evaluate this possibility are imperative. The strength of this study is that it substantiates the findings from previous similar studies.

Acknowledgment This study was supported by the Internal Grant Fund (IG/AGR/Food/10/01) and the Strategic Research Fund offered by His Majesty Sultan of Oman (SR/MED/FMCO/11/01), College of Medicine and Health Sciences, Sultan Qaboos University.

Competing Interests None.

References

1. American Psychiatric Association (2000) Diagnostic and statistical manual of mental disorders DSM-IV-TR, 4th edn. American Psychiatric Publication, Washington DC
2. Rutter M (2005) Incidence of autism spectrum disorders: changes over time and their meaning. Acta Paediatr 94(1):2–15
3. Kim YS, Leventhal BL, Koh YJ, Fombonne E, Laska E, Lim EC, Cheon KA, Kim SJ, Kim YK, Lee H, Song DH, Grinker RR (2011) Prevalence of autism spectrum disorders in a total population sample. Am J Psychiatry 168(9):904–912
4. Hertz-Picciotto I, Delwiche L (2009) The rise in autism and the role of age at diagnosis. Epidemiol 20(1):84–90
5. Centre for Disease Control (2007) Prevalence of autism spectrum disorders—autism and developmental disabilities monitoring network, 14 sites, United States, 2002. In: Surveillance summaries. MMWR 56(SS-1):12–28, February 9, 2007
6. Baron-Cohen S, Scott FJ, Allison C, Williams J, Bolton P, Matthews FE, Brayne C (2009) Prevalence of autism-spectrum conditions: UK school-based population study. Br J Psychiatry 194(6):500–509
7. Kogan MD, Blumberg SJ, Schieve LA, Boyle CA, Perrin JM, Ghandour RM, Singh GK, Strickland BB, Trevathan E, Van Dyck PC (2009) Prevalence of parent-reported diagnosis of autism spectrum disorder among children in the US, 2007. Pediatr 124 (5):1395–1403
8. Zaroff CM, Uhm SY (2012) Prevalence of autism spectrum disorders and influence of country of measurement and ethnicity. Soc Psychiatry Psychiatr Epidemiol 47(3):395–398
9. Elsabbagh M, Divan G, Koh YJ, Kim YS, Kauchali S, Marcín C, Montiel-Nava C, Patel V, Paula CS, Wang C, Yasamy MT, Fombonne E (2012) Global prevalence of autism and other pervasive developmental disorders. Autism Res 5(3):160–179
10. Blaurock-Busch E, Amin OR, Rabah T (2011) Heavy metals and trace elements in hair and urine of a sample of Arab children with autistic spectrum disorder. Maedica (Buchar) 6 (4):247–257
11. Al-Ayadhi LY (2005) Heavy metals and trace elements in hair samples of autistic children in central Saudi Arabia. Neurosci (Riyadh) 10(3):213–218

12. Eapen V, Mabrouk AA, Zoubeidi T, Yunis F (2007) Prevalence of pervasive developmental disorders in preschool children in the UAE. J Trop Pediatr 53:202–205
13. Al-Farsi YM, Al-Sharbati MM, Al-Farsi OA, Al-Shafaee MS, Brooks DR, Waly MI (2011) Brief report: prevalence of autistic spectrum disorders in the Sultanate of Oman. J Autism Dev Disord 41(6):821–825
14. Bernard S, Enayati A, Redwood L, Roger H, Binstock T (2001) Autism: a novel form of mercury poisoning. Med Hypotheses 56:462–471
15. Herbert MR (2010) Contributions of the environment and environmentally vulnerable physiology to autism spectrum disorders. Curr Opin Neurol 23(2):103–110
16. Geschwind SA, Stolwijk JA, Bracken M, Fitzgerald E, Stark A, Olsen C, Melius J (1992) Risk of congenital malformations associated with proximity to hazardous waste sites. Am J Epidemiol 135(11):1197–1207
17. Evans H, Laties VG, Weiss B (1975) Behavioral effects of mercury and methylmercury. Fed Proc 34:1858–1867
18. Hensen MC, Chedrese PJ (2004) Endocrine disruption by cadmium, a common environmental toxicant with paradoxical effects on reproduction. Exp Biol Med 229:383–392
19. Thompson WW, Price C, Goodson B, Shay DK, Benson P, Hinrichsen VL, Lewis E, Eriksen E, Ray P, Marcy SM, Dunn J, Jackson LA, Lieu TA, Black S, Stewart G, Weintraub ES, Davis RL, DeStefano F, Vaccine Safety Datalink Team (2007) Early thimerosal exposure and neuropsychological outcomes at 7 to 10 years. N Engl J Med 357(13):1281–1292
20. Soden SE, Lowry JA, Garrison CB, Wasserman GS (2007) 24-Hour provoked urine excretion test for heavy metals in children with autism and typically developing controls, a pilot study. Clin Toxicol (Phila) 45(5):476–481
21. Elsheshtawy E, Toba S, Kheta S et al (2011) Study of some biomarkers in hair of children with autism. Middle East Curr Psych 18:6–10
22. Abdullah MM, Ly AR, Goldberg WA, Clarke-Stewart KA, Dudgeon JV, Mull CG, Chan TJ, Kent EE, Mason AZ, Ericson JE (2012) Heavy metal in children's tooth enamel: related to autism and disruptive behaviors. J Autism Dev Disord 42(6):929–936
23. Vallee BL (1952) Trace elements in biochemistry and nutrition. Nutr Rev 10(3):65–68
24. Fido A, Dashti H, Al-Saad S (2002) Biological correlates of childhood autism: "trace elements". Trace Elem Electrolytes 19 (4):205–208
25. Fido A, Al-Saad S (2005) Toxic trace elements in the hair of children with autism. Autism 9(3):290–298
26. Semprún-Hernández N, Bohórquez-Visier AP, Henríquez AB, Bohórquez RC, Medina FH, Maury-Sintjago E, Ocando NM (2012) Copper, zinc, calcium and magnesium profiles in subjects with autistic disorder according to their functioning level. Trace Elem Electrolytes 29(1):1–5
27. Lakshmi Priya MD, Geetha A (2011) Level of trace elements (copper, zinc, magnesium and selenium) and toxic elements (lead and mercury) in the hair and nail of children with autism. Biol Trace Elem Res 142(2):148–158
28. Lubkowska A, Sobieraj W (2009) Concentrations of magnesium, calcium, iron, selenium, zinc and copper in the hair of autistic children. Trace Elem Electrolytes 26(2):72–77
29. Wecker L, Miller SB, Cochran SR (1985) Trace element concentrations in hair from autistic children. J Ment Defic Res 29(Pt 1):15–22
30. Kern JK, Jones AM (2006) Evidence of toxicity, oxidative stress, and neuronal insult in autism. J Toxicol Environ Health B Crit Rev 9(6):485–499
31. Schopler E, Reichler RJ, Renner BR (1994) The childhood autism rating scale. Western Psychological Services, Los Angeles, California
32. Filipek PA, Accardo PJ, Baranek GT, Cook EH Jr, Dawson G, Gordon B, Gravel JS, Johnson CP, Kallen RJ, Levy SE, Minshew NJ, Ozonoff S, Prizant BM, Rapin I, Rogers SJ, Stone WL, Teplin S, Tuchman RF, Volkmar FR (1999) The screening and diagnosis of autistic spectrum disorders. J Autism Dev Disord 29(6):439–484
33. Al-Yousuf MH, El-Shahawi MS, Al-Ghais SM (2000) Trace metals in liver, skin and muscle of *Lethrinus lentjan* fish species in relation to body length and sex. Sci Total Environ 256(2–3):87–94
34. De Mora S, Fowler SW, Wyse E, Azemard S (2004) Distribution of heavy metals in marine bivalves, fish and coastal sediments in the Gulf and Gulf of Oman. Mar Pollut Bull 49(5–6):410–424
35. Doyle P, Roman E, Maconochie N (1997) Birth defects among children of Persian Gulf War veterans. N Engl J Med 337 (16):1175–1176
36. Rajab KE, Mohammad AM, Mustafa F (2000) Incidence of spontaneous abortion in Bahrain before and after the Gulf War of 1991. Int J Gynaecol Obstet 68(2):139–144
37. Frustaci A, Neri M, Cesario A, Adams JB, Domenici E, Dalla Bernardina B, Bonassi S (2012) Oxidative stress-related biomarkers in autism: systematic review and meta-analyses. Free Radic Biol Med 52(10):2128–2141
38. Al-Farsi YM, Al-Sharbati MM, Waly MI, Al-Farsi OA, Al Shafaee MA, Deth RC (2011) Malnutrition among preschool-aged autistic children in Oman. Res Autism Spectr Disord 5(4):1549–1552
39. Al-Farsi YM, Waly MI, Ouhtit A, Al-Sharbati MM, Al-Shafaee MA, Al-Farsi OA, Al-Khaduri MM, Gupta I, Al-Khalili M, AL-Adawi S, Hodgson NW, Deth RC (2012) Low folate and vitamin B12 nourishment is common among Omani children newly diagnosed with autism. Nutrition (in press)
40. Adams JB, Romdalvik J, Ramanujam VM, Legator MS (2007) Mercury, lead, and zinc in baby teeth of children with autism versus controls. J Toxicol Environ Health A 70(12):1046–1051
41. Carneiro MFH, Moresco MB, Chagas GR, De Oliveira Souza VC, Rhoden CR, Barbosa F Jr (2011) Assessment of trace elements in scalp hair of a young urban population in Brazil. Biol Trace Elem Res 143(2):815–824
42. Gentile PS, Trentalange MJ, Zamichek W, Coleman M (1983) Brief report: trace elements in the hair of autistic and control children. J Autism Dev Disord 13(2):205–206
43. Jung MA, Jang HS, Park EJ, Lee HW, Choi JH (2008) Study on the mineral and heavy metal contents in the hair of preschool aged autistic children. J Korean Soc Food Sci Nutr 37(11):1422–1426
44. De Palma G, Catalani S, Franco A, Brighenti M, Apostoli P (2012) Lack of correlation between metallic elements analyzed in hair by ICP-MS and autism. J Autism Dev Disord 42(3):342–353
45. deSoto MC, Hitlan RT (2010) Sorting out the spinning of autism: heavy metals and the question of incidence. Acta Neurobiol Exp (Wars) 70(2):165–176
46. Soliman AT, Madina EH, Morsi MR (1996) Radiological, biochemical, and hormonal changes in malnourished children with rachitic manifestations. J Trop Pediatr 42(1):34–37