

Aluminum Vaccine Adjuvants: Are they Safe?

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Abstract: Aluminum is an experimentally demonstrated neurotoxin and the most commonly used vaccine adjuvant. Despite almost 90 years of widespread use of aluminum adjuvants, medical science's understanding about their mechanisms of action is still remarkably poor. There is also a concerning scarcity of data on toxicology and pharmacokinetics of these compounds. In spite of this, the notion that aluminum in vaccines is safe appears to be widely accepted. Experimental research, however, clearly shows that aluminum adjuvants have a potential to induce serious immunological disorders in humans. In particular, aluminum in adjuvant form carries a risk for autoimmunity, long-term brain inflammation and associated neurological complications and may thus have profound and widespread adverse health consequences. In our opinion, the possibility that vaccine benefits may have been overrated and the risk of potential adverse effects underestimated, has not been rigorously evaluated in the medical and scientific community. We hope that the present paper will provide a framework for a much needed and long overdue assessment of this highly contentious medical issue.

Keywords: Aluminum adjuvants, adjuvant safety, autoimmunity, autism, Gulf War Syndrome, multiple sclerosis, macrophagic myofasciitis, neurotoxicity, seizures, Th2 immune response, vaccines.

INTRODUCTION

Aluminum is the most commonly used vaccine adjuvant and until recently the only one licensed for use in the U.S. [1-4]. In its absence, antigenic components of most vaccines (with the exception of live attenuated vaccines), fail to launch an adequate immune response [1, 5, 6]. Paradoxically, despite almost 90 years of widespread use of aluminum adjuvants [3] their precise mechanism of action remains poorly understood [1, 2]. Furthermore, a growing number of studies have linked the use of aluminum adjuvants to serious autoimmune outcomes in humans [5-8]. That concerns about aluminum adjuvant safety are indeed warranted is evident from the summary conclusions of the *Aluminum in Vaccines* workshop held in Puerto Rico in 2000 [2]. The written consensus amongst the participants of the workshop was listed under the rubric of "pervasive uncertainty", a term used to denote what remained unknown regarding potential aluminum toxicity from adjuvants. The specific areas of concern were: "1) toxicology and pharmacokinetics, specifically the processing of aluminum by infants and children, 2) mechanisms by which aluminum adjuvants interact with the immune system and 3) the necessity of adjuvants in booster doses." In the concluding paragraphs of the summary, the report nevertheless claimed that "the use of salts of aluminum as adjuvants in vaccines has proven to be safe and effective" [2]. In light of the items of "pervasive uncertainty", this statement remains questionable. Given that multiple aluminum-adjuvanted vaccines are often given to very young children (i.e., 2 to 6 months of age), in a single day at individual vaccination sessions [9, 10], concerns for potential impacts of total adjuvant-derived aluminum body burden may be significant [11, 12]. These issues warrant serious consideration since, to the best of our knowledge, no adequate studies have been conducted to assess the safety of simultaneous administration of different vaccines to young children. Another issue of concern is the lack of any toxicological evaluation about concomitant administration of aluminum with other known toxic compounds which are routine constituents of commercial vaccine preparations, e.g., formaldehyde, formalin, mercury, phenoxyethanol, phenol, sodium borate, polysorbate 80, glutaraldehyde [13, 14]. In spite of all this, aluminum adjuvants are generally regarded as safe [2, 13],

and some researchers have even recommended that no further research efforts should be spent on this topic despite "a lack of good-quality evidence"[15].

In the following paper we aim to provide an overview of what is currently known about aluminum adjuvants, their modes of action and mechanisms of potential toxicity. We first present well established evidence that implicates aluminum in a variety of neurological disorders. We then elaborate on the unresolved controversy about aluminum adjuvant safety.

ALUMINUM TOXICITY IN ANIMALS AND HUMANS

Aluminum is a well demonstrated toxin in biological systems [16] whose more specific impacts on the nervous system have been widely documented (Table 1). As early as 1911, Dr. William Gies had summarized data from 7 years worth of experimental testing in humans and animals on the effects of oral consumption of aluminium salts, then used primarily in baking powders, food preservation, and dye manufacturing [17]. The outcome of these studies led Gies to conclude that: "the use in food of aluminum or any other aluminum compound is a dangerous practice." Gies' concerns have since been borne out by experimental studies showing that oral exposure to aluminum that is at levels "typically" consumed in an average "Western diet" over an extended period of time, produce strikingly similar outcomes in rodents to those induced by intracerebral injection of aluminum salts (Table 1) with the exception of seizures and fatalities [18, 19]. Animals intoxicated with dietary aluminum routinely show impaired performance in learning and memory tasks, impaired concentration, and behavioural changes including confusion and repetitive behaviours [18, 19]. Consistent with these observations, according to the most recent and elaborate toxicological report for aluminum prepared by the Agency for Toxic Substances and Disease Registry (ATSDR): "There is a rather extensive database on the oral toxicity of aluminum in animals. These studies clearly identify the nervous system as the most sensitive target of aluminum toxicity"[16].

In humans, aluminum toxicity has been solidly linked to dialysis-associated encephalopathy syndrome, also known as dialysis dementia (Table 1). This syndrome occurs in patients with renal failure subjected to chronic dialysis treatment and is caused by accumulation of intravenously administered aluminum from the dialysis fluid (which is derived from aluminum-treated tap water [20]). Dialysis dementia is associated with abnormally high levels of plasma and brain aluminum and is generally fatal within 3 to 7

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Table 1. Neurodevelopmental Toxicity of Aluminum Compounds in Various Species

Aluminum source/compound	Dose & duration	Route	Species	Neurodevelopmental adverse effects
Standard infant feeding solution	~20 µg/kg/day, >10 days	Intravenous (parenteral)	Human, premature infants	Reduced developmental attainment at the corrected post-term age of 18 months, as evidenced by significantly lower Bayley Mental Development Index (BMDI) scores (mean loss of one point on the BMDI/day of full intravenous feeding, after adjustment for potentially confounding factors) compared to infants fed with Al-depleted solutions [32]
Al-containing antacids	Chronic	Oral	Human infants	Craniosynostosis (premature ossification of the skull and obliteration of the sutures) [33]
Al-containing dialysis fluid (derived from Al-sulphate treated tap water)	1 ppm, chronic (2-5 years)	Intravenous	Human, kidney failure patients (15-61 years old at the start of the dialysis treatment)	Speech impairments (stuttering, dysarthria, dyspraxia, motor aphasia), movement disorders (twitches, tremors, myoclonic jerks, seizures, motor apraxia), cognitive impairments and behavioural changes (progressive dementia, paranoia, confusion, psychosis), death [21]
Al-sulphate (present as flocculant in potable water supplies, accidentally released in high amounts)	500-3000 x the acceptable limit under European Union legislation (0.200 mg/L), chronic (15 years)	Oral	Human adult (female, 44 years old)	Sporadic early-onset β amyloid angiopathy (Alzheimer's-related disease), difficulty in finding words, progressive dementia, visual hallucinations headache, anxiety, cerebral ischaemia, death [34]
Various dietary	Chronic	Oral	Elderly human subjects	Impaired visuo-motor coordination, poor long-term memory, and increased sensitivity to flicker (correlated with high Al-serum levels [35])
Al-oxide fumes, occupational exposure	0.13-1.95 mg/m ³ , chronic	Inhalation	Human, adults (mean age 39 years)	Headache, emotional irritability, concentration difficulty, insomnia, mood lability [36]
Various: Al-chloride, Al-phosphate, Al-powder slurry	Single sub-lethal dose	Intracerebral injection	Cats, rabbits	Decline in memory, impaired learning responses, deterioration in psychomotor control, epileptic seizures and death, neurofibrillary degeneration (resembling Alzheimer's disease neurofibrillary tangles [37-42])
Al-hydroxide	2 injections, 2 weeks apart	Subcutaneous injection (behind the neck)	Mice, 3-month old	Motor neuron degeneration and apoptosis, motor function deficits, decrease in strength, cognitive deficits and decreased performance in learning tasks, decrements in spatial memory, activation of microglia [43, 44]
Al-containing food pellets	0.5-1.7 mg/kg/day (typical human), chronic (22-32 months)	Oral	Rats, 6-month old at the start of treatment	Cognitive deterioration and impaired performance in learning tasks, impaired concentration, behavioural changes including confusion and repetitive behaviour [45]
Al-lactate	500-1000 ppm, chronic (during gestation and lactation)	Oral	Mice dams	Hind limb paralysis, seizures and death (dams), lower neurobehavioral development and altered performance on a neurobehavioural test battery in pups (foot splay, forelimb and hind limb grip strengths [46])

months following the sudden overt manifestation of clinical symptoms in patients who had been on dialysis treatment for 3 to 7 years [21, 22] (unless treated with chelating agent such as desferrioxamine (DFO) or reverse osmosis to remove aluminum salts from the water used to prepare the dialysis fluid [20-23]). Symptoms appear suddenly and worsen either during or immediately after a dialysis session [21, 22, 24-26]. The first symptom to appear is a speech abnormality, then tremors, impaired psychomotor control, memory losses, impaired concentration, behavioural changes, epileptic seizures, coma and death [20-22, 24-26]. Although frequent ingestion of aluminum-containing medicines was also thought to be a contributing factor in dialysis dementia [26], it should be noted that there were no incidences of this syndrome prior to introduction of aluminum salts in water supplies [21, 27]. Furthermore, symptomatic patients rapidly improved when efforts were made to remove aluminum from the dialysis fluid, despite the fact they still ingested large amounts of

aluminum-containing phosphate binding gels [21]. In addition to dialysis dementia, a host of neurodegenerative complications and diseases such as Alzheimer's [11, 28], Parkinson's disease [29], amyotrophic lateral sclerosis (ALS) [29], multiple sclerosis [30], Gulf War Syndrome (GWS) [5, 6], autism [31], and epilepsy [12] may also be related to aluminum exposure. While it is likely that these diseases are of multifactorial etiologies, aluminum certainly has the potential to serve as a toxic co-factor.

ALUMINUM EXPOSURE FROM VACCINES: BODY BURDENS AND RISKS

During the course of the last 30 years, the number of officially scheduled vaccines deemed necessary for children in the U.S. has increased sharply, from 10 in the 1980s to 32 in the late 2000s, 18 of which contain aluminum adjuvants [11]. The issue of vaccine safety thus becomes even more pertinent given that, to the best of

our knowledge, no adequate clinical studies have been conducted to establish the safety of concomitant administration of two experimentally-established neurotoxins, aluminum and mercury, the latter in the form of ethyl mercury (thimerosal) in infants and children. Since these molecules negatively affect many of the same biochemical processes and enzymes implicated in the etiology of autism, the potential for a synergistic toxic action is plausible [31, 47]. Additionally, for the purpose of evaluating safety and efficacy, vaccine clinical trials often use an aluminium-containing placebo, either containing the same or greater amount of aluminum as the test vaccine [48-51]. Without exception, these trials report a comparable rate of adverse reactions between the placebo and the vaccine group (for example, 63.7% vs 65.3% of systemic events and 1.7% vs 1.8% of serious adverse events respectively [51]). According to the U.S. Food and Drug Administration (FDA), a placebo is “an inactive pill, liquid, or powder that has no treatment value” [52]. The well-established neurotoxic properties of aluminium (Table 1) therefore suggest that aluminum cannot constitute as a valid placebo.

In 1965, Klatzo *et al.* [38] demonstrated that aluminum phosphate, the primary constituent of Holt’s adjuvant, induced degeneration and neurofibrillary tangle-like histological changes in neurons (a hallmark feature of Alzheimer’s disease), when injected intracerebrally into rabbits. The aluminum-injected animals also suffered from convulsions [38]. While direct application of aluminum adjuvants to the central nervous system (CNS) is unquestionably neurotoxic [37, 38, 40, 42], little is known about aluminum transport into and out of the CNS, its toxicokinetics, and the impact on different neuronal subpopulations following subcutaneous or intramuscular injections. The reason for this is that under current regulatory policies, evaluation of pharmacokinetic properties is not required for vaccines [53]. This issue is of special concern in context to worldwide mass immunization practices involving children whose nervous systems are undergoing rapid development. Furthermore, an immature developing blood brain barrier (BBB) is more permeable to toxic substances than that of an

adult [16, 54]. In addition, there are critical periods in neurodevelopment that occur within first few years of postnatal life during which exposure to neurotoxic insults may induce CNS damage [16, 47, 55]. In that respect, it is worth noting that any potential CNS damage caused by aluminum in children may not be evident until a later stage of development [16].

Bishop *et al.* [32] have shown that, parenteral exposure to as little as 20 µg/kg bw of aluminum for >10 days may result in long-term detrimental outcomes in neurologic development in preterm infants. In 2004, the U.S. Food and Drug Administration (FDA) set a limit for aluminum from parenteral sources for individuals with impaired kidney function and premature neonates at no greater than 4 to 5 µg/kg bw/day, stating that levels above those have been associated with CNS and bone toxicity [56]. In addition, according to the FDA, tissue loading may occur at even lower levels of administration [56]. What the upper limit for “safe” aluminum exposure might be for healthy neonates is not known.

In spite of these above data, newborns, infants and children up to 6 months of age in the U.S. and other developed countries receive 14.7 to 49 times more than the FDA safety limits for aluminum from parenteral sources from vaccines through mandatory immunization programs (Table 2). Specifically, 2-month old children in U.K., U.S., Canada and Australia routinely receive as much as 220 to 245 µg/kg bw of aluminum per vaccination session (Table 2), a burden equivalent to 34 standard adult-dose injections of hepatitis B vaccine (Table 3). Similarly, newborns at birth receive 73.5 µg Al/kg bw/day from a single hepatitis B vaccine, which is a dose equivalent to 10 standard adult-dose injections of hepatitis B vaccine in a single day (Table 3). Whether such doses of aluminum are safe even for adults is not known. However, detrimental effects associated with multiple vaccinations over a short period of time in U.S. and other Coalition military personnel who developed GWS in an aftermath of only six anthrax vaccine inoculations [5, 6], may suggest that adults in some circumstances are also vulnerable to deleterious CNS effects of adjuvant-aluminum. Notably, these inoculations were not given in a

Table 2. Estimated total aluminum body burden (µg/kg bw/day) per vaccination session in various developed countries. Vaccine schedules were obtained from the following sources: U.K. (U.K. Department of Health [10]), U.S. (Centers for Disease Control and Prevention [9]), Canada (Public Health Agency of Canada [57]) and Australia (Australian Government Department of Health and Aging [58]). Aluminum content of vaccines was according to Offit and Jew [3]

	Birth	1 mo	2 mo	3 mo	4 mo	5 mo	6 mo
U.K.	73.5	62.5	245	184	193	0	0
U.S.	73.5	0	245	0	171.1	0	161.2
Canada	73.5	0	220	0	193	0	111.8
Australia	73.5	0	220	0	193	0	144.7

FDA safety limit for Al from parenteral sources: 5 µg/kg bw/day.

Table 3. Comparison of aluminum body burden from vaccines in children and adults. Note that the closest an adult can get to the aluminum body burden from vaccines that compares to that of a child is in special circumstances, such as Gulf War deployed military personnel. Each anthrax vaccine administered to Gulf War veterans contained 1200 µg Al/mL (600 µg Al/dose) [59]. Currently licensed hepatitis B vaccines Enderix-B and Recombivax contain 250 (pediatric) and 500 µg Al/dose (adult) [3]. Age-specific weights were sourced from Haddad and Krishnan [60]

	An infant receiving 1 HepB injection (250 µg/dose) at birth	A 2-month old receiving the full U.S. scheduled set of injections	An adult receiving 6 anthrax injections over 18 months	An adult receiving 73.5 µg/kg bw/visit from HepB at 500 µg/dose	An adult receiving 245 µg/kg bw/visit from HepB at 500 µg/dose
Total Al (µg)	250	1225	3600	5145	17,150
Bw (kg)	3.4	5	70	70	70
Total Al µg/kg bw/day	73.5	245	51.4	73.5	245
# of Al-adjuvanted HepB at 500 µg/dose	NA	NA	NA	10	34

single day but were spread out over several weeks and up to 18 months (Table 3).

In a recent review, Offit and Jew [3], in addressing concerns about potential aluminum adjuvant toxicity, cited as evidence an uncontrolled feeding study by Golub *et al.* [61], which used aluminum lactate as the form of treatment. The reviewers stated that: "No adverse reactions were observed when mice were fed quantities of aluminum as high as 62 mg/kg/day" [3], when in fact 20% of the mice showed significantly lower motor activity [61]. Moreover, Golub *et al.* [61] emphasized that: "The clear cut influence of dietary Al on motor activity suggests the value of further testing of Al fed animals in areas of sensory-motor competence as well as cognitive and social functioning". Also often unrecognized by researchers [3, 13] is the fact that different aluminum compounds may vary in their toxic potential or that the extent of toxicity of a particular compound depends on a specific route of administration, duration of exposure, and species studied. For example, while feeding aluminum hydroxide at 66.5, 133, and 266 mg Al/kg/day to mice does not appear to cause neurodevelopmental damage [62, 63], parenteral administration of aluminum chloride at 40 mg/kg/day causes maternal deaths in rats, as well as embryo lethality, growth retardation and fetal abnormalities [64]. The latter effects were also shown to occur at lower doses (20 mg/kg/day [64]). The authors of the former study that used higher doses of aluminum hydroxide concluded that this form of aluminum is very poorly absorbed and thus does not reach the fetus at levels which might pose a developmental hazard [63]. A rigorous survey of the primary literature further shows that evidence for pre, perinatal and postnatal aluminum neurotoxicity is well established [65-71], even at very low doses of aluminum. For example, Gonda *et al.* [72] have shown that parenteral exposure during gestation days 7 to 15 to as little as 2.5, 5 and 10 mg/kg/day of aluminum lactate results in diminished performance and lengthened latency in avoidance response in rat pups. The evidence for potential aluminum toxicity in early life is thus more firmly established than suggested by some researchers [3, 13, 15].

Finally, it should be fairly obvious that parenterally administered aluminum bears more relevance to vaccine exposure than dietary aluminum. In this context it is worth noting that unlike dietary aluminum of which only ~0.25 % is absorbed into systemic circulation [73], aluminum from vaccines may be absorbed at nearly 100% efficiency [74]. It is also important to note that ionic aluminum will not have the same toxicokinetic properties as aluminum bound to an antigen. While ionic aluminum may be excreted *via* the kidneys, the sizes of most antigen-aluminum complexes (24-83 kDa [59, 75, 76]), are higher than the molecular weight cut-off of the glomerulus (~18 kDa [12]), likely precluding efficient excretion of these compounds. Indeed, effective excretion would in fact obviate the basic reason that adjuvants are used at all. For all these reasons, vaccine-derived aluminum has a much greater potential to induce neurological damage than that obtained through diet, even in those with effective renal function. In addition, adjuvant-aluminum can gain access to the CNS as demonstrated by Redhead *et al.* [77], who showed that intraperitoneal injection of aluminum adsorbed vaccines in mice caused a transient rise in brain tissue aluminum levels peaking around the second and third day after injection.

ALUMINUM TOXICOKINETICS: DEVELOPING BRAIN, A SINK FOR ADJUVANT-ALUMINUM?

Experiments by Levy *et al.* [78] in which antibodies were raised against an immunogen prepared from aluminum and bovine serum albumin (BSA) suggested that aluminum on its own may act as an antigen. These results raise questions concerning the possibility that vaccination with aluminum adjuvants may increase an individual's susceptibility to subsequent exposure to aluminum. Given the

ubiquity of bioavailable aluminum compounds (food, water, cosmetics, pharmaceuticals [16]), such issues warrant further investigation. The existing data available on the pharmacokinetics of aluminum adjuvants suggest that these compounds may access systemic circulation and cross the blood brain barrier. Flarend *et al.* [79] estimated aluminum absorption in adult female rabbits following intramuscular injection of two forms of ²⁶Al labeled adjuvants, aluminum hydroxide and aluminum phosphate. The results showed that both were rapidly absorbed, appearing in the blood as early as one hour after injection [79]. Blood levels of aluminum remained elevated for 28 days post-injection in both cases and subsequent tissue analysis revealed elevated levels of aluminum in kidney, spleen, liver, heart, lymph nodes and, notably, brain [79]. In Flarend *et al.*'s [79] study the level of aluminum in the brain was lower compared to the other organs, however the study by Yumoto *et al.* [80] indicated that such a pattern of tissue distribution may be age-dependent. Following a single subcutaneous injection of ²⁶Al on gestation day 15, these investigators showed that 0.2% of the ²⁶Al injected into a pregnant rat had been transplacentally transferred to the fetuses. Notably, the amount of the radiolabeled aluminum in the fetal brain was 30% higher than in the liver, while in the dams, brain aluminum levels were only 1% of the levels found in the liver [80]. The possibility that the fetal brain may act as a sink for aluminum may be of concern since under certain circumstances, vaccination of pregnant women with a number of aluminum-adjuvanted vaccines (tetanus, hepatitis A and B, meningococcal and pneumococcal is recommended [3, 81]) under the current U.S. immunization guidelines [82].

ADVERSE EFFECTS ASSOCIATED WITH ALUMINUM ADJUVANTS

A recently described syndrome termed macrophagic myofasciitis (MMF) has been specifically attributed to aluminum adjuvants in recipients of hepatitis A and B and tetanus toxoid (Td) vaccines [83]. MMF patients were found to suffer from diffuse arthromyalgias, chronic fatigue, muscle weakness and in some cases, multiple sclerosis [83]. Muscle biopsies show extensive infiltration by granular periodic acid-Schiff's reagent-positive macrophages and lymphocytes and inconspicuous muscle-fibre damage [2, 7, 83-85]. While most MMF patients appeared to have a normal white blood count, laboratory analysis showed evidence of increased inflammation and the presence of serum auto-antibodies. The former was indicated by significant increases in the levels of inflammatory cytokines interleukin (IL)-1 receptor antagonist and IL-6 [2]. Electron microscopy and microanalytical analysis showed that the appearance of MMF lesions was due to long-term persistence of aluminum adjuvants at the site of injections and concomitant ongoing local immune reactions [8, 83]. Aluminum was shown to persist at the site of injection from several months up to 8 years following vaccination [83, 85]. MMF lesions were subsequently also reproduced in rats by injection of aluminum adjuvants [86].

Aluminum adjuvants are exceptionally potent stimulators of the immune system and their specific action is to shift the immune response towards a Th2 profile. In that respect, Dr. Gherardi who first described MMF noted: "It is plausible that persistent systemic immune activation that fails to switch off represents the pathophysiologic basis of chronic fatigue syndrome associated with macrophagic myofasciitis, similarly to what happens in patients with post-infectious chronic fatigue and possibly idiopathic chronic fatigue syndrome" [8]. The symptoms of MMF are similar to those of GWS, a multisystem disorder which has been linked to multiple vaccinations administered over a short period of time (Table 3 [6, 8]). As with autism and MMF, GWS patients also show Th2 predominance and a significant risk factor in causing this syndrome may be aluminum hydroxide adjuvant from the anthrax vaccine.

Injections of aluminum hydroxide at levels comparable to those administered to Gulf War veterans, were shown to cause significant motor neuron degeneration as well as impairments in motor function and decrements in spatial memory capacity in young CD-1 male mice [43, 44].

Of even graver concern is that persistent Th2 stimulation, due to repeated administration of aluminum-adjuvanted vaccines, may have profound long-term adverse effects on the developing immune system in children. A newborn infant has an undeveloped immune system which is limited in function [87] and requires a series of challenges to bring it to full capacity. Prior introduction of mandatory vaccines, these challenges were largely in the forms of relatively minor childhood diseases such as mumps and measles. Vaccinations targeted at stimulating antibody production by the humoral immune system (Th2) located in the bone marrow, bypass the cellular immune system (Th1) on mucosal surfaces (respiratory and gastrointestinal tract), leaving the latter unchallenged during the critical period of development. Since Th1 progenitors will not differentiate into Th1 cells in the absence of Th1-cytokines [88] (due chronic stimulation of the Th2 pathway), the end result of a prolonged Th2 shift may be permanently stunted cellular (Th1) immunity. Ironically, Th1 immunity is inherently far more efficient in clearing viral pathogens than Th2 immunity [6, 88, 89], which further raises a question about the general efficacy of aluminum-adjuvanted vaccines in fighting viral infections. Notably, a similar mechanism by which acute, subacute or chronic stress selectively suppress cellular (Th1) immunity but boosts humoral (Th2) immunity, is thought to be responsible for the onset and/or course of many infectious, autoimmune/inflammatory, allergic and neoplastic diseases [89]. For example, research indicates that by inducing a Th2 shift, stress hormones may increase susceptibility to acute respiratory infections caused by flu viruses and enhance disease progression in human immunodeficiency virus (HIV)-positive individuals [89]. Furthermore, severe acute stress associated with high adrenaline output leads to histamine release from Th2 type immune cells (mast cells), which may either initiate new or exacerbate existing allergic reactions [89]. Finally, high histamine levels have been observed in various cancer tissues, suggesting that stress hormone dependent amplification of Th2 responses can increase the susceptibility to tumorigenesis [89]. Taken together, these observations potentially explain why naturally acquired immunity against common childhood diseases may protect against certain aggressive types of tumors in humans [90], asthma and other allergies [91, 92], as well as neurodegenerative disorders such as Parkinson's [93].

Although most autoimmune diseases are Th1-related, others such as lupus-like syndromes (Table 4), are mediated by Th2 cytokines IL-10 [89] and IL-4 [95]. It is thought that vaccine adjuvants may trigger autoimmunity through a bystander effect, by activating dormant autoreactive T-cells in predisposing individuals [96]. Notably, the repertoire of adverse reactions and syndromes associated with aluminum-adjuvanted vaccines (Table 4), appears

to fit the spectrum of diseases stemming from immune dysfunction [5, 6]. In addition, fatalities have been reported among individuals who were vaccinated against with the anthrax vaccine. These included deaths from sudden cardiac arrest, myocardial infarction with polyarteritis nodosa, aplastic anemia, CNS lymphoma and suicide [59]. Since the anthrax vaccine contains a higher dose of aluminum than most other aluminium-adjuvanted vaccines (0.6 mg/dose vs 0.5 mg/dose Engerix-B [59, 94]), combined with another potent adjuvant and Th2 stimulant, squalene [6], the potential for synergistic adverse actions by these two adjuvants in humans cannot be discounted.

Fatal outcomes have also been reported following administration of pediatric aluminum-adjuvanted hexavalent vaccines, one of which (Hexavac) was subsequently withdrawn from use, apparently due to its poor effectiveness [97]. Zinka *et al.* [98] reported six cases of sudden infant death that occurred within 48 hours after vaccination with hexavalent vaccines. The post-mortem analysis of six children aged 4 to 17 months (five of whom were vaccinated with Hexavac and one with Infanrix Hexa), revealed abnormal pathologic findings particularly affecting the nervous system [98]. The overall pathological abnormalities included acute congestion, defective BBB, infiltration of the leptomeninges by macrophages and lymphocytes, perivascular lymphocytic infiltration, diffuse infiltration of the pons, mesencephalon and cortex by T-lymphocytes, microglia in the hippocampus and pons, and in one case, necrosis in the cerebellum [98]. Increased serum mast-cell tryptase and numbers of eosinophilic granulocytes were also found indicating that an anaphylactic reaction developed subsequent to vaccination [98]. As shown in Table 4, anaphylaxis appears to be a common side effect associated with aluminum-adjuvanted vaccines. According to Zinka *et al.* [98], there was a 13-fold increase in infant death following introduction of hexavalent vaccines into immunization practice [97]. Although there is no conclusive proof that these deaths were directly caused by vaccination, the authors felt it was "important to inform vaccinating physicians and pediatricians as well as parents about such possibly fatal complications after application of hexavalent vaccines" [98]. Finally, the neuropathological findings by Zinka *et al.* [98] are consistent with neurotoxic properties of aluminum adjuvants. For example, as shown by our group as well others, aluminum is a BBB neurotoxin [54, 99] that has a propensity to activate brain microglia and increase the production of inflammatory cytokines thereby instigating and/or exacerbating inflammation and excitotoxicity in the brain [31, 43, 44, 100-104].

Permanent activation of brain inflammatory responses has long been recognized as a factor in etiology of many neurodegenerative diseases [105] including Alzheimer's disease [106, 107], autism [31, 108-110], multiple sclerosis [30] and dialysis dementia [111]. Notably, all of these diseases have been previously linked to aluminum exposure [12, 21, 28, 30, 31, 107, 111]. Aluminum potentiates inflammatory responses in the brain by multiple mechanisms, such as activation of microglia [31, 44, 100, 101, 107,

Table 4. Engerix-B and BioThrax (Anthrax Vaccine) Common Post-Licensure Adverse Effects [59, 94]

Blood and Lymphatic System Disorders	Idiopathic thrombocytopenia
Immune System Disorders	Anaphylaxis and/or other generalized hypersensitivity reactions, inflammatory arthritis/arthritis, fever, and dermatologic reactions such as erythema, systemic lupus erythematosus
Nervous System Disorders	Encephalitis, multiple sclerosis, Guillain-Barré syndrome, transverse myelitis, facial palsy, seizures, syncope
Eye Disorders	Visual disturbances
Cardiac Disorders	Cardiac arrhythmias
Respiratory, Thoracic and Mediastinal Disorders	Asthma
Skin and Subcutaneous Tissue Disorders	Angioedema, erythema
Musculoskeletal and Connective Tissue Disorders	Arthritis, myalgia, muscle weakness

112] and induction of pro-inflammatory gene expression [107]. Regarding the latter, aluminum at nanomolar to low micromolar concentrations augments specific neuroinflammatory and pro-apoptotic signalling cascades, strikingly similar to those observed in Alzheimer's disease brains [104], by driving expression from a subset of stress-inducible promoters in cultured human primary brain cells [113-115]. For example, out of 8 induced genes up-regulated in cultured human neurons by 100 nm aluminum, 7 showed expression patterns similar to those observed in Alzheimer's disease, including hypoxia inducible factor (HIF)-1 and nuclear factor (NF)- κ B-responsive amyloid β -protein precursor (A β PP), IL-1 β precursor, NF- κ B subunits, cytosolic phospholipase A₂ (cPLA₂), cyclooxygenase (COX)-2 and DAXX, a regulatory protein known to induce apoptosis and repress transcription [114]. Both HIF-1 and NF- κ B are up-regulated in Alzheimer's disease where they fuel the pro-inflammatory cycle which leads to further exacerbation of oxidative stress and inflammation, culminating in neuronal death [105, 116]. Taken together, these results underscore the potential of physiologically relevant levels of aluminum to drive genotoxic mechanisms characteristic of neurodegenerative disease processes [115].

CONCLUSIONS

Aluminum in various forms can be toxic to the nervous system. The widespread presence in the human environment may underlie a number of CNS disorders. The continued use of aluminum adjuvants in various vaccines for children as well as the general public may be of significant concern. In particular, aluminum presented in this form carries a risk for autoimmunity, long-term brain inflammation and associated neurological complications and may thus have profound and widespread adverse health consequences. The widely accepted notion of aluminum adjuvant safety does not appear to be firmly established in the scientific literature and, as such, this absence may have led to an erroneous conclusions regarding the significance of these compounds in the etiologies of many common neurological disorders. Furthermore, the continued use of aluminum-containing placebos in vaccine clinical trials may have led to an underestimation of the true rate of adverse outcomes associated with aluminum-adjuvanted vaccines. In our opinion, a comprehensive evaluation of the overall impact of aluminum on human health is overdue. Such an evaluation should include studies designed to determine the short and long-term impacts of dietary aluminum as well as the potential impacts in different age groups of exposure to adjuvant aluminum alone and in combination with other potentially toxic vaccine constituents (e.g., formaldehyde, formalin, mercury, phenoxyethanol, phenol, sodium borate, polysorbate 80, glutaraldehyde). For the latter, until vaccine safety can be comprehensively demonstrated by controlled independent long-term studies that examine the impact on the nervous system in detail, many of those already vaccinated as well as those currently receiving injections may be at risk for health complications that exceed the potential benefits that vaccine prophylaxis may provide. The issue of aluminum adjuvanted vaccine safety is especially pertinent in light of the legislation which might mandate vaccination regimes for civilian populations (e.g., the Biodefense and Pandemic Vaccine and Drug Development Act of 2005). Whether the risk of protection from a dreaded disease outweighs the risk of toxicity from its presumed prophylactic agent is a question that demands far more rigorous scrutiny than has been provided to date.

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