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VIA ELECTRONIC FILING

May 6, 2021

Division of Dockets Management
Department of Health and Human Services
Food and Drug Administration
Commissioner Stephen M. Hahn, M.D.
5630 Fishers Lane, Rm. 1061
Rockville, MD 20852

UNITED STATES DEPARTMENT OF HEALTH AND HUMAN SERVICES AND THE FOOD AND DRUG ADMINISTRATION

PETITION FOR ADMINISTRATIVE ACTION TO ENSURE ACCURATELY REPORTED AND CONSISTENT LEVELS OF ALUMINUM IN ALL VACCINES	**:**	**Docket No. _____**

CITIZEN PETITION

This petition for administrative action is submitted on behalf of the Informed Consent Action Network and a large number of its members, including parents deciding whether to vaccinate their child/children, ("**Petitioner**") pursuant to 21 C.F.R. § 10.30 and related and relevant provisions of law (including the Federal Food, Drug, and Cosmetic Act or the Public Health Service Act) to request that the Commissioner of Food and Drugs (the "**Commissioner**") take the actions listed below to assure accurately reported and consistent levels of aluminum in Adacel, Boostrix, Engerix-B, Havrix, Infanrix, Infanrix hexa, Kinrix, Pediarix, Pedvax-HIB, Pentacel, Prevnar-13, Synflorix, and Vaqta (the "**Subject Vaccines**").

A. ACTION REQUESTED

1. The Food & Drug Administration ("**FDA**") forthwith publicly release documentation sufficient to establish that the aluminum content in each Subject Vaccine is consistent with amount provided in its labeling.[1]

2. If the FDA is unable to forthwith comply with the foregoing request, the FDA forthwith pause distribution of each Subject Vaccine until it has confirmed and publicly released documentation sufficient to establish that the aluminum content in each Subject Vaccine is consistent with the amount provided in its labeling.

B. STATEMENT OF GROUNDS

3. On April 15, 2021, Dr. Christopher Exley along with four other researchers have published a study after reviewing the aluminum content of thirteen childhood vaccines. Dr. Exley has authored over 200 published peer reviewed articles regarding aluminum, has been a Professor of Bioinorganic Chemistry at Keele University for the last 29 years, and has otherwise spent almost his entire 37-year career studying aluminum and its biological effects.

4. This study found that only three vaccines of the thirteen tested contained the amount of aluminum indicated on its labeling approved by the FDA. Six vaccines (Pentacel, Havrix, Adacel, Pedvax, Prevnar 13, and Vaqta) contained a statistically significant greater quantity while four vaccines (Infanrix, Kinrix, Pediarix, and Synflorix) contained a statistically significant lower quantity. A copy of this peer-reviewed study with these findings are appended hereto as Exhibit A and is available at https://www.sciencedirect.com/science/article/pii/S0946672X21000523.

5. These deviations from each product's labeling render the product adulterated and misbranded and violates various federal statutes and regulations, and therefore requires immediate action from the FDA to either provide proof the study's results are incorrect or otherwise cease distribution of these vaccines until this issue has been corrected. *See, e.g.*, 21 U.S.C. § 351; 21 U.S.C. § 352; 21 C.F.R. § 56; 21 C.F.R. § 57.

6. The finding in this study is extremely concerning because doses with less than the approved amount of aluminum adjuvant will not have the same efficacy, and doses with more than the approved amount of aluminum adjuvant raise safety concerns. Indeed, aluminum adjuvant is

[1] The term "labeling" as used herein shall include all documentation from the manufacturer and the FDA with regard to a given product, including its package insert, product label, patient information sheet, and approval documents, and any other documents that list its ingredients.

a known cytotoxic and neurotoxic substance used to induce autoimmunity in lab animals, and which numerous peer-reviewed publications implicate in various autoimmune conditions.[2]

7. The FDA must ensure that vaccines in current use and those that will be on the market in the future are accurately labeled. Vaccine recipients and their caregivers must be able to rely on the FDA-approved labeling for these products, especially considering that they are given to babies and children.

8. Petitioner and its constituent members, and the parents seeking to decide whether to vaccinate their children, are entitled to know if the aluminum adjuvant content in the FDA approved childhood vaccines they are being asked to inject their children with are not adulterated or mislabeled, and otherwise contain the amount of aluminum adjuvant actually listed on their label.

C. ENVIRONMENTAL IMPACT

9. The undersigned hereby states that the relief requested in this petition will have no environmental impact and therefore an environmental assessment is not required under 21 C.F.R. Sections 25.30 and 25.31.

D. ECONOMIC IMPACT

10. Economic impact information will be submitted upon request of the commissioner.

E. CERTIFICATION

11. The undersigned certifies that, to the best knowledge and belief of the undersigned, this petition includes all information and views on which the petition relies, and that it includes representative data and information known to the petitioner which are unfavorable to the petition.

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[2] https://www.simonandschuster.com/books/Imagine-You-Are-An-Aluminum-Atom/Christopher-Exley/9781510762534; https://www.wiley.com/en-us/Vaccines+and+Autoimmunity-p-9781118663431; https://www.ncbi.nlm.nih.gov/pubmed/25923134; http://icandecide.org/white-papers/ICAN-Aluminum Adjuvant-Autism.pdf; Macrophages phagocytize (ingest) aluminum adjuvant (**AA**): https://www.ncbi.nlm.nih.gov/pubmed/15297065; https://www.ncbi.nlm.nih.gov/pubmed/18496530. Macrophages transport material into the brain: https://www.ncbi.nlm.nih.gov/pubmed/27213597; https://www.ncbi.nlm.nih.gov/pubmed/21348773; https://www.ncbi.nlm.nih.gov/pubmed/27115998; https://www.ncbi.nlm.nih.gov/pubmed/27213597. AA transport to brain: https://www.ncbi.nlm.nih.gov/pubmed/26384437; https://www.ncbi.nlm.nih.gov/pubmed/27908630; https://www.ncbi.nlm.nih.gov/pubmed/23557144. AA causes neuro impairment: https://www.ncbi.nlm.nih.gov/pubmed/27908630; https://www.ncbi.nlm.nih.gov/pubmed/19740540; https://www.ncbi.nlm.nih.gov/pubmed/23932735. Macrophages infiltrate the brain : https://www.ncbi.nlm.nih.gov/pubmed/16401547; https://www.ncbi.nlm.nih.gov/pubmed/15546155; https://www.ncbi.nlm.nih.gov/pubmed/28167942; https://www.ncbi.nlm.nih.gov/pubmed/24951035.

12. The Petitioner therefore respectfully urges that this request be granted forthwith.

Respectfully submitted,

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Exhibit A

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The measurement and full statistical analysis including Bayesian methods of the aluminium content of infant vaccines

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ABSTRACT

Background: Aluminium salts are the most common adjuvants in infant vaccines. The aluminium content of a vaccine is provided by the manufacturer and is indicated on the patient information leaflet. There is no independent verification, for example by the European Medicines Agency, of the aluminium content of infant vaccines.

Methods: We have measured the aluminium content of thirteen infant vaccines using microwave-assisted acid and peroxide digestion followed by transversely heated graphite furnace atomic absorption spectrometry. Our data are compared with manufacturer's data using full statistical analyses including Bayesian methods.

Results: We found that only three vaccines contained the amount of aluminium indicated by the manufacturer. Six vaccines contained a statistically significant ($P < 0.05$) greater quantity while four vaccines contained a statistically significant ($P < 0.05$) lower quantity. The range of content for any single vaccine varied considerably, for example, from 0.172 to 0.602 mg/vaccine for Havrix.

Conclusions: The data have raised specific questions about the significance of the aluminium content of vaccines and identified areas of extremely limited information. Since aluminium is a known toxin in humans and specifically a neurotoxin, its content in vaccines should be accurate and independently monitored to ensure both efficacy and safety.

1. Introduction

Aluminium salts are the adjuvants of choice in the majority of inactivated paediatric vaccines. Their presence at injection sites following vaccination results in the activation of humoral immunity. This pathway is characterised by the differentiation of naive $CD4^+$ T cells into Th2 effector subsets and the subsequent enhancement of antigen-specific antibody titres [1–5]. T cell priming post-vaccination occurs following the cross-presentation of antigen-MHC complexes by immunocompetent phagocytes and occurs exclusively within draining lymph nodes [6] at locations often distant from the injection site. Aluminium adjuvants have been shown to facilitate this process by; i) actively increasing levels of antigen recognition and uptake by antigen-presenting cells [7–9], ii) acting in a protective capacity to prevent antigen degradation within intracellular compartments [9,10] and (iii) amplifying and sustaining antigen-MHC II expression by antigen-presenting cells [9,11,12]. Other suggested mechanisms underlying the immunostimulatory effects of aluminium adjuvants include the secretion of pro-inflammatory cytokines by activated antigen-presenting cells [13–17] and the formation of an 'antigen depot' at the vaccine injection site [18,19]. While the latter is regarded by some as superfluous to the mechanism of action of aluminium adjuvants, recent evidence has demonstrated that extracellular traps formed by neutrophils following immunization make a significant contribution to their immunological activity *in vivo* [20]. However, the modus operandi of aluminium adjuvants remains to be fully elucidated [21,22].

Three aluminium salts are currently used as adjuvants in human vaccines. Two of these, aluminium oxyhydroxide (available commercially as Alhydrogel®) and aluminium hydroxyphosphate (available commercially as AdjuPhos®) have been widely studied while the third, aluminium hydroxyphosphate sulphate, is proprietary to Merck and has not been available for independent scrutiny [22,23]. The type and amount of aluminium adjuvant used in paediatric vaccines is made available through patient information leaflets, see the manufacturer's information summarised in Table 1. The information given is, at best, vague. Descriptions of aluminium salts are often inaccurate, for

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example, the use of the term aluminium hydroxide when the form of aluminium included is aluminium oxyhydroxide. Quantitative data describing the content of aluminium in a vaccine is often presented in a number of different formats. For example, all do present these data as total aluminium but some additionally present the data in respect of the weight of aluminium salt. This practice is confusing for anyone reading the patient information leaflet. The manufacturers' stated content of aluminium in vaccines listed in Table 1 varies from 0.125 (Prevnar 13) to 0.85 (PEDIARIX) mg per 0.5 mL dose of vaccine though little or no information is available as to why the content is so varied. Why does one vaccine require more aluminium adjuvant to be effective than another? There exists a non-regulatory limit of 1.25 mg aluminium per dose of vaccine based upon maximum titres of antibodies produced. However, even this may be exceeded under certain circumstances. The European Medicines Agency (EMA) and the Food and Drug Administration (FDA) are charged with the responsibility of verifying the information provided by vaccine manufacturers in their patient information leaflets. When questioned repeatedly on this subject (for example, EMA request reference ASK-56123) neither organisation was able to confirm that they routinely measure the aluminium content of vaccines. They indicated that they relied upon data provided to them by manufacturers, though no such data were forthcoming following requests including freedom of information act requests to the FDA (For example, FOIA Requests 2019-11150 to 2019-11156 & 2019-11158). When the EMA was asked, which analytical methods were used by either their organisation or vaccine manufacturers to measure the aluminium content of vaccines, they replied that this was proprietary information and could not be provided (EMA request reference ASK-56707). It would appear that the aluminium content of a vaccine is only measured by the manufacturer, using an unspecified method, and that these data are not made publicly available.

One piece of important information about the aluminium content of a vaccine is that it is clearly critical. Why else would there be such a wide range of contents used across the paediatric vaccine schedule. Since those charged with ensuring that the information provided by vaccine manufacturers is correct are seemingly choosing to neglect this responsibility, herein we have measured the aluminium content of thirteen paediatric vaccines. We find that the measured content of aluminium is only similar to that given by the manufacturer in three out of thirteen vaccines.

2. Materials and methods

2.1. Vaccines

Whole vaccines were provided under license by a state registered paediatrician. All vaccines were in their original packaging and remained pristine and refrigerated at 4 °C until use.

2.2. Digestion of vaccines

Each whole vaccine was added to an acid-washed, dried and labelled 20 mL PFA Teflon MARSXpress digestion vessel closed with a venting plug and screw cap (CEM Technology, UK). The idea being that the whole vaccine was 'injected' into the digestion vessel in the identical manner as it would be used in human vaccination. In each case, it was assumed, though it was unlikely to be the case, that the whole vaccine volume of 0.5 mL was transferred to the digestion tube. To each whole vaccine, 1 mL of concentrated HNO_3 (Analar, 15.8 M Fisher Scientific, UK) and 1 mL 30 % w/v H_2O_2 (Aristar, BDH, UK) were added and the mixture subjected to microwave-assisted digestion (MARS6 CEM, One Touch Technology, UK). The resulting digests were further diluted by the addition of 2.5 mL of pure water (conductivity <0.067μS/cm) and stored appropriately for subsequent analyses. Method blanks were prepared where 0.5 mL of pure water was substituted for the whole vaccine. Full information pertaining to this method of sample digestion including microwave parameters are available elsewhere [24].

Table 1
Information relating to each of the thirteen infant vaccines taken directly from their patient information leaflets respectively.

Trade name of vaccine	Pharmaceutical company [Country of manufacture]	Paediatric dose (mL)	Manufacturer's stated aluminium content per paediatric dose (mg)	Infant age range	Manufacturer's description of vaccine
PEDIARIX	GlaxoSmithKline (GSK) [Belgium]	0.5	"not more than **0.85 mg** aluminium"	from 2 to 6 months	Vaccine for the active immunization against diphtheria, tetanus, pertussis, hepatitis B virus infection and poliomyelitis.
Pentacel	Sanofi Pasteur [Canada]	0.5	**0.33 mg** of aluminium as 1.5 mg aluminium phosphate	from 6 weeks to 4 years	Vaccine for the active immunization against diphtheria, tetanus, pertussis, poliomyelitis and disease caused by *Haemophilus influenzae* type b.
ENGERIX-B	GlaxoSmithKline (GSK) [Belgium]	0.5	**0.25 mg** of aluminium as aluminium hydroxide	from 1 to 6 months	Vaccine for immunization against infection caused by hepatitis B virus.
Prevnar 13	Pfizer [United States]	0.5	**0.125 mg** of aluminium (as aluminium phosphate)	from 6 weeks to 5 years	Vaccine for active immunization against disease caused by *Streptococcus pneumoniae*.
PedvaxHIB	Merck & Co., Inc [United States]	0.5	**0.225 mg** of aluminium as amorphous aluminium hydroxyphosphate sulphate	from 6 to 11 months	Vaccine for immunization against infection caused by *Haemophilus influenzae* type b.
KINRIX	GlaxoSmithKline (GSK) [Belgium]	0.5	"not more than **0.6 mg** aluminium by assay"	from 4 up to 6 years	Vaccine for immunization against diphtheria, tetanus, pertussis and poliomyelitis.
INFANRIX	GlaxoSmithKline (GSK) [Belgium]	0.5	"not more than **0.625 mg** aluminium by assay"	from 6 weeks up to 6 years	Vaccine for active immunization against diphtheria, tetanus and pertussis.
BOOSTRIX	GlaxoSmithKline (GSK) [Belgium]	0.5	"not more than **0.39 mg** aluminium by assay"	from 10 years	Vaccine for active immunization against tetanus, diphtheria and pertussis.
VAQTA	Merck & Co., Inc [United States]	0.5	**0.225 mg** of aluminium as amorphous aluminium hydroxyphosphate sulphate	from 12 months (up to 18 years)	Vaccine for immunization against disease caused by hepatitis A virus
Adacel	Sanofi Pasteur [Canada]	0.5	1.5 mg aluminium phosphate (**0.33 mg** aluminium)	from 10 years	Vaccine for active immunization against tetanus, diphtheria and pertussis.
HAVRIX	GlaxoSmithKline (GSK) [Belgium]	0.5	**0.25 mg** of aluminium as aluminium hydroxide	from 12 to 24 months	Vaccine for immunisation against disease caused by hepatitis A virus.
Infanrix hexa	GlaxoSmithKline (GSK) [Belgium]	0.5	**0.82 mg** of aluminium (as aluminium salts)	from 2 to 18 months	Vaccine for immunization against diphtheria, tetanus, pertussis, hepatitis B, poliomyelitis and *Haemophilus influenzae* type b.
Synflorix	GlaxoSmithKline (GSK) [Belgium]	0.5	**0.5 mg** of aluminium as aluminium phosphate	from 6 weeks up to 5 years	Vaccine for immunization against infections caused by *Streptococcus pneumoniae*.

2.3. Determination of aluminium

The total aluminium content of each vaccine digest and each method blank was measured by transversely heated graphite furnace atomic absorption spectrometry (TH GFAAS) using a fully established method including matrix-matched standards and commensurate quality assurance criteria [24]. Method blank data equating to 54 ngAl/5 mL digest were subtracted from each vaccine sample. Data are presented as mgAl/0.5 mL vaccine volume.

2.4. Statistics

Data on the measured concentrations of aluminium, stratified by brand, were tested for normality. Depending on whether the data were normally distributed or not, means and medians were calculated and tested two- and one-sided against the manufacturer's content of aluminium (see Table 1) using one-sample *t*-test and Wilcoxon signed rank test respectively.

Differences in aluminium content between lots were analysed for those vaccines with two lots and sufficient sample numbers.

To determine the probability, expressed as a percentage, that the measured content of aluminium in a vaccine was less than, the same as or greater than the amount given by the vaccine manufacturer, Bayesian methodology was used [25].

The hypotheses tested were as follows:

H0. Declared content of aluminium is equal to or less than the measured concentrations of aluminium. (The difference in means is zero.)

The alternative hypothesis is that the difference in means is not zero:

Ha1. Measured concentration of aluminium is not the same.

Ha2. Measured concentration of aluminium is greater.

Ha3. Measured concentration of aluminium is less.

Tests were repeated one-sided in both directions. For two-sided tests, a p-value < 0.05 was considered as statistically significant and for one-sided tests a p-value < 0.025.

Analyses were performed using R-Studio version 1.1.1093 [26] including packages ggplot2 [27], doBy [28], BEST [29], BayesianFirstAid [30] and bayesWilcoxTest [31]. The last three packages provide Bayesian alternatives to the classical hypothesis tests in R. Bayesian methods were used to calculate the percentage probability that measured aluminium concentrations were randomly the same or greater/less than amounts stated by the manufacturer. Bayes factors can complement p-values by providing additional information for hypothesis testing by quantifying the relative evidence for both alternative and null hypotheses. Moreover, the magnitude of this evidence can be presented as percentages [25,32–36].

The following code was used to produce random data sets of aluminium values for each vaccine with the stated manufacturers mean and 10 % RSD. The latter to reflect 'expected' manufacturing error.

```
rnorm2 <- function(n,mean,sd) {mean + sd*scale(rnorm(n))}
```

The Bayesian approach was repeated with the random data sets including the 10 % variation.

3. Results

3.1. Pentacel

The data for Pentacel were normally distributed. Ten individual vaccines were investigated across two lots. The aluminium content differed significantly between lots ($P < 0.030$). The highest content of aluminium measured was 0.440 mg/vaccine. The lowest content of aluminium measured was 0.343 mg/vaccine. The mean content of ten vaccines was 0.379 mg/vaccine (Table 2). The aluminium content of Pentacel was significantly higher for both lots combined ($P < 0.001$) (Fig. 1), lot 1 only ($P = 0.005$) and lot 2 only ($P = 0.004$) than the amount stated by the manufacturer (0.330 mg/vaccine) on the patient information leaflet (Table 3).

3.2. Havrix

The data for Havrix were not normally distributed. Twenty individual vaccines were investigated across two lots. The aluminium content differed significantly between lots ($P < 0.001$). The highest content of aluminium measured was 0.602 mg/vaccine. The lowest content of aluminium measured was 0.172 mg/vaccine. The median content of twenty vaccines was 0.307 mg/vaccine (Table 2). The aluminium content of Havrix was significantly higher for both lots combined ($P = 0.003$) (Fig. 1) and lot 2 only ($P = 0.003$) than the amount stated by the manufacturer (0.250 mg/vaccine) on the patient information leaflet (Table 3).

3.3. Adacel

The data for Adacel were not normally distributed. Nine individual vaccines were investigated across two lots. The aluminium content was not significantly different between lots ($P > 0.05$).The highest content of aluminium measured was 0.445 mg/vaccine. The lowest content of aluminium measured was 0.302 mg/vaccine. The median content of nine vaccines was 0.397 mg/vaccine (Table 2). The aluminium content of Adacel was significantly higher ($P = 0.006$) (Fig. 1) than the amount stated by the manufacturer (0.330 mg/vaccine) on the patient information leaflet (Table 3).

3.4. Boostrix

The data for Boostrix were normally distributed. Twenty individual vaccines were investigated from a single lot. The highest content of aluminium measured was 0.525 mg/vaccine. The lowest content of aluminium measured was 0.345 mg/vaccine. The mean content of twenty vaccines was 0.407 mg/vaccine (Table 2). The aluminium content of Boostrix was not significantly different ($P = 0.101$) (Fig. 1) to the amount stated by the manufacturer (0.390 mg/vaccine) on the patient information leaflet (Table 3).

3.5. EngerixB

The data for EngerixB were normally distributed. Twenty individual

Table 2
Descriptive statistics for each of the thirteen infant vaccines including normal distribution (N.D.), number of replicates (N), minimum (Min.), maximum (Max.), mean/median values (mg/0.5 mL dose) and indication of variance (SD, RSD). Vaccines ordered according to their mean/median Al contents.

Vaccine	N.D.	N	Min.	Max.	Mean/ Median	SD	RSD
Prevnar 13	Y	6	0.127	0.141	0.136	0.006	0.04
EngerixB	Y	20	0.187	0.287	0.249	0.027	0.11
Pedvax	Y	20	0.192	0.334	0.287	0.040	0.14
Havrix	N	20	0.172	0.602	0.307	0.084	0.28
Vaqta	N	20	0.270	0.796	0.340	0.109	0.32
Pentacel	Y	10	0.343	0.440	0.379	0.032	0.09
Adacel	N	9	0.302	0.445	0.397	0.041	0.10
Synflorix	Y	3	0.396	0.399	0.398	0.002	0.00
Boostrix	Y	20	0.345	0.525	0.407	0.043	0.11
Kinrix	N	20	0.464	0.635	0.511	0.062	0.12
Infanrix	Y	20	0.415	0.662	0.546	0.069	0.13
Pediarix	Y	20	0.575	0.743	0.661	0.039	0.06
Infanrix Hexa	Y	6	0.766	0.851	0.806	0.028	0.04

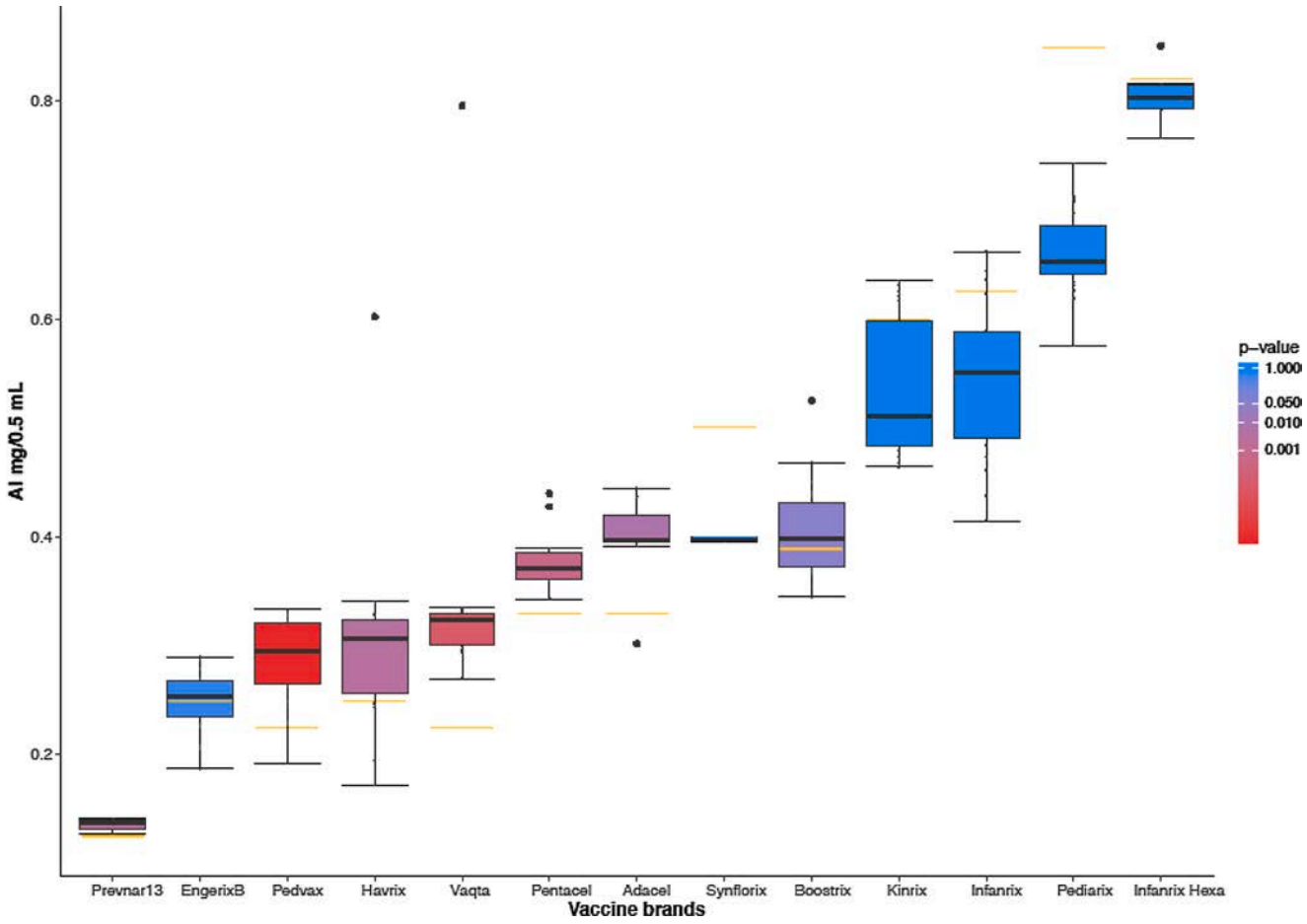

Fig. 1. Boxplots of measured aluminium concentrations (mg/0.5 mL dose) stratified per vaccine brand and compared to the manufacturers' stated amounts (yellow line). The colour gradient represents p-values of one-sided t-tests or Mann-Whitney U tests of the measured Al against the manufacturers' stated amounts. The red boxplots indicate vaccines with significantly more aluminium than that stated by the manufacturer. Boxplots in violet and blue represent data for vaccines where the measured content is either not significantly to or significantly less than the manufacturers' stated amounts. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article).

vaccines were investigated from a single lot. The highest content of aluminium measured was 0.287 mg/vaccine. The lowest content of aluminium measured was 0.187 mg/vaccine. The mean content of twenty vaccines was 0.249 mg/vaccine (Table 2). The aluminium content of EngerixB was not significantly different ($P = 0.897$) (Fig. 1) to the amount stated by the manufacturer (0.250 mg/vaccine) on the patient information leaflet (Table 3).

3.6. Infanrix

The data for Infanrix were normally distributed. Twenty individual vaccines were investigated from a single lot. The highest content of aluminium measured was 0.662 mg/vaccine. The lowest content of aluminium measured was 0.415 mg/vaccine. The mean content of twenty vaccines was 0.546 mg/vaccine (Table 2). The aluminium content of Infanrix was significantly lower ($P < 0.001$) (Fig. 1) than the amount stated by the manufacturer (0.625 mg/vaccine) on the patient information leaflet (Table 3).

3.7. Infanrix Hexa

The data for Infanrix Hexa were normally distributed. Six individual vaccines were investigated from a single lot. The highest content of aluminium measured was 0.851 mg/vaccine. The lowest content of aluminium measured was 0.766 mg/vaccine. The mean content of six vaccines was 0.806 mg/vaccine (Table 2). The aluminium content of Infanrix Hexa was not significantly different ($P = 0.268$) (Fig. 1) to the amount stated by the manufacturer (0.820 mg/vaccine) on the patient information leaflet (Table 3).

3.8. Kinrix

The data for Kinrix were not normally distributed. Twenty individual vaccines were investigated across two lots. The aluminium content was not significantly different between lots ($P > 0.05$). The highest content of aluminium measured was 0.635 mg/vaccine. The lowest content of aluminium measured was 0.464 mg/vaccine. The median content of twenty vaccines was 0.511 mg/vaccine (Table 2). The aluminium content of Kinrix was significantly less ($P = 0.001$) (Fig. 1) than the amount stated by the manufacturer (0.600 mg/vaccine) on the patient information leaflet (Table 3).

3.9. Pediarix

The data for Pediarix were normally distributed. Twenty individual vaccines were investigated across two lots. The aluminium content was not significantly different between lots ($P > 0.05$). The highest content of aluminium measured was 0.743 mg/vaccine. The lowest content of aluminium measured was 0.575 mg/vaccine. The mean content of twenty vaccines was 0.661 mg/vaccine (Table 2). The aluminium content of Pediarix was significantly lower ($P < 0.001$) (Fig. 1) than the amount stated by the manufacturer (0.850 mg/vaccine) on the patient

Table 3
Summary statistics of results including statistical test used (T test or Mann Whitney U), two- and one-sided p-values of statistical tests, difference in means (stated Al amount – measured amount) and percentage of evidence for the outcome according to Bayesian methodology. Vaccines ordered according to their mean/median Al content.

Vaccine	Test	p-value 2-sided	p-value 1-sided	Diff. in Means	Outcome (H_1)	Probability of Outcome
Prevnar 13	T	0.005	0.003	−0.01	Greater	0.993
EngerixB	T	0.897	0.551	0.00	Same or Less	0.513
Pedvax	T	< 0.001	< 0.001	−0.06	Greater	> 0.999
Havrix	MWU	0.006	0.003	−0.06	Greater	0.872
Havrix Lot 1	T	0.710	0.355	−0.01	Greater	0.661
Havrix Lot 2	MWU	0.006	0.003	−0.08	Greater	0.988
Vaqta	MWU	< 0.001	< 0.001	−0.11	Greater	0.995
Pentacel	T	0.001	< 0.001	−0.05	Greater	0.999
Pentacel Lot 1	T	0.010	0.005	−0.03	Greater	0.986
Pentacel Lot 2	T	0.008	0.004	−0.07	Greater	0.991
Adacel	MWU	0.013	0.006	−0.07	Greater	0.938
Synflorix	T	< 0.001	1.000	0.10	Same or Less	0.998
Boostrix	T	0.101	0.051	−0.02	Greater	0.924
Kinrix	MWU	0.001	0.999	0.09	Same or Less	0.878
Infanrix	T	< 0.001	1.000	0.08	Same or Less	> 0.999
Pediarix	T	< 0.001	1.000	0.19	Same or Less	> 0.999
Infanrix Hexa	T	0.268	0.866	0.01	Same or Less	0.846

information leaflet (Table 3).

3.10. Pedvax

The data for Pedvax were normally distributed. Twenty individual vaccines were investigated across two lots. The aluminium content was not significantly different between lots ($P > 0.05$). The highest content of aluminium measured was 0.334 mg/vaccine. The lowest content of aluminium measured was 0.192 mg/vaccine. The mean content of twenty vaccines was 0.287 mg/vaccine (Table 2). The aluminium content of Pedvax was significantly higher ($P < 0.001$) (Fig. 1) than the amount stated by the manufacturer (0.225 mg/vaccine) on the patient information leaflet (Table 3).

3.11. Prevnar13

The data for Prevnar13 were normally distributed. Six individual vaccines were investigated in a single lot. The highest content of aluminium measured was 0.141 mg/vaccine. The lowest content of aluminium measured was 0.127 mg/vaccine. The mean content of six vaccines was 0.136 mg/vaccine (Table 2). The aluminium content of Prevnar13 was significantly higher ($P = 0.003$) (Fig. 1) than the amount stated by the manufacturer (0.125 mg/vaccine) on the patient information leaflet (Table 3).

3.12. Synflorix

The data for Synflorix were normally distributed. Three individual vaccines were investigated from a single lot. The highest content of aluminium measured was 0.399 mg/vaccine. The lowest content of aluminium measured was 0.396 mg/vaccine. The mean content of three vaccines was 0.398 mg/vaccine (Table 2). The aluminium content of Synflorix was significantly less ($P < 0.001$) (Fig. 1) than the amount stated by the manufacturer (0.500 mg/vaccine) on the patient information leaflet (Table 3).

3.13. Vaqta

The data for Vaqta were not normally distributed. Twenty individual vaccines were investigated across two lots. The aluminium content was not significantly different between lots ($P > 0.05$). The highest content of aluminium measured was 0.796 mg/vaccine. The lowest content of aluminium measured was 0.270 mg/vaccine. The median content of twenty vaccines was 0.340 mg/vaccine (Table 2). The aluminium content of Vaqta was significantly higher ($P < 0.001$) (Fig. 1) than the amount stated by the manufacturer (0.225 mg/vaccine) on the patient information leaflet (Table 3).

4. Discussion

We present the aluminium content of 13 paediatric vaccines (Supplementary Table 1). We have compared our raw data with values given by manufacturers on patient information leaflets. In addition, we have applied a generous ±10 % margin of manufacturing error to the latter published values when applying Bayesian methods (see Supplementary Table 2). Using a level of statistical significance of $P = 0.05$, 3 vaccines contained the amount of aluminium stated by the manufacturer on the patient information leaflet (Boostrix, Engerix B, Infanrix Hexa). Six vaccines contained significantly more aluminium (Pentacel, Havrix, Adacel, Pedvax, Prevnar 13, Vaqta) while 4 vaccines contained significantly less (Infanrix, Pediarix, Kinrix, Synflorix). Statistical significance, of course, does not tell the only story. For example, while the aluminium content of Boostrix is not significantly different to that stated by the manufacturer ($P > 0.05$) there remains a 92 % chance that the aluminium content of a Boostrix vaccine will exceed the official content (Table 3). In addition, the content of aluminium is extremely variable with many vaccines showing %RSD in excess of 10 % even within the same lot (Table 2). For example, an infant receiving Havrix could receive anything between 0.172 and 0.602 mg of aluminium per vaccine. The data presented herein will be an underestimate of the actual content of aluminium as it is inevitable that some aluminium adjuvant will remain within the syringe system following injection. This will also be true when vaccines are injected *in vivo*. Vaccines that include an aluminium adjuvant are cloudy suspensions and manufacturers recommend that they are shaken prior to injection. For a few vaccines we were unable to obtain ten or more individual products and so data are limited, especially Synflorix. However, we present the first robust data obtained using state-of-the-art methods on the aluminium content of vaccines currently being administered in infants. The data are not reassuring. They suggest that vaccine manufacturers have limited control over the aluminium content of their vaccines. The aluminium content of individual vaccines within vaccine lots vary significantly. The amount of aluminium an infant receives in a vaccine is, it would appear, akin to a lottery. The true significance of this lottery is unknown. Vaccine manufacturers do not provide experimental protocols or rationales to support the amount of aluminium used in vaccines. If the amount used relates to a vaccine's potency in eliciting antibody titres then this should be explained in complementary information including, for example, that provided with the vaccine. Equally, how this potency is affected by the quantity of aluminium should also be a matter of public record. For example, using the data previously noted for Havrix, does it matter for the vaccine's efficacy if the content of aluminium received by an infant is 0.172 or 0.602 mg/vaccine. The natural assumption is that it does matter, otherwise why state specific amounts of aluminium on patient information leaflets. If the vaccine manufacturers stated content of aluminium is significant then it is concerning that six of the thirteen vaccines measured had statistically higher contents of aluminium. In

practice, this would mean that many infants are receiving significantly more aluminium than recommended by the manufacturer. How is this additional aluminium affecting the efficacy and safety of the vaccine? Similarly, four out of the thirteen vaccines contained statistically significantly less aluminium than recommended by the manufacturer. Is the lower than prescribed amount of aluminium in these vaccines affecting their efficacy?

The data raise many open questions about the significance of the amount of aluminium included in vaccines. They demonstrate if nothing else that further clarity and transparency is required from vaccine manufacturers as well as regulatory organisations such as the EMA and FDA. The aluminium content of a vaccine is never trivial [37]. There is a long history of testing the efficacy of childhood vaccines against false placebos and warnings against this practice continue to go unheeded [38]. It should be a matter of concern that a recent freedom of information act request (FOIA Case Number 50882, and HHS Appeal No.; 19-0083-AA) revealed that the NIH were unable to provide a single study relied upon by them in relation to the safety of injection of aluminium adjuvants in infants. Human exposure to aluminium is an unequivocal consequence of everyday living [39]. Aluminium adjuvants in infant vaccines contribute towards the body burden of aluminium [37]. Aluminium in the body has the potential to be toxic and significantly neurotoxic [40]. Where aluminium is being used for apparent human benefit, as in vaccines, we cannot simply ignore the other side of the coin, its known toxicity. We cannot afford to be complacent about its injection into infants [22].

Aluminium adjuvants are critical to the efficacy of vaccines and are far from being benign components [22]. Information on their content, activity and safety is severely lacking and this void requires urgent attention. Until such information is forthcoming, aluminium adjuvants remain prime suspects in widely documented vaccination-related adverse events.

Author contributions

ES: Analytical support and contributed to writing of the manuscript. CL: Carried out all statistics and contributed to writing of the manuscript. SC and ES: Carried out the majority of measurements and contributed to writing of the manuscript. CE: Design of study, analytical support and contributed to writing the manuscript.

Declaration of Competing Interest

The authors report no declarations of interest.

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Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.jtemb.2021.126762.

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