International practice of corticosteroid replacement therapy in congenital adrenal hyperplasia - data from the I-CAH registry.

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Short title: Steroid replacement in CAH

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Abstract

Objective: Despite published guidelines no unified approach to hormone replacement in congenital adrenal hyperplasia (CAH) exists. We aimed to explore geographical and temporal variations in the treatment with glucocorticoids and mineralocorticoids in CAH.

Design: This retrospective multi-center study, including 31 centers (16 countries), analyzed data from the International-CAH Registry.

Methods: Data was collected from 461 patients aged 0-18 years with classic 21-hydroxylase deficiency (54.9% females) under follow-up between 1982 – 2018. Type, dose and timing of glucocorticoid and mineralocorticoid replacement was analyzed from 4174 patient visits.

Results: The most frequently used glucocorticoid was hydrocortisone (87.6%)., Overall, there were significant differences between age groups with regards to daily hydrocortisone-equivalent dose for body surface, with the lowest dose (median with interquartile range) of 12.0 (10.0 – 14.5) mg/ m^2/ day at age 1 - 8 years and the highest dose of 14.0 (11.6 - 17.4) mg/ m^2/ day at age 12-18 years. Glucocorticoid doses decreased after 2010 in patients 0-8 years (*p<0.001*) and remained unchanged in patients aged 8-18 years. Fludrocortisone was used in 92% of patients, with relative doses decreasing with age. A wide variation was observed among countries with regards to all aspects of steroid hormone replacement.

Conclusions: Data from the I-CAH Registry suggests international variations in hormone replacement therapy, with a tendency to treatment with high doses in children.

Introduction

Congenital adrenal hyperplasia (CAH) represents a group of autosomal recessive conditions leading to glucocorticoid (GC) deficiency. It is caused by defects in the steroidogenic enzymes involved in cortisol biosynthesis or the electron providing factor P450 oxidoreductase (1-4). The most common form, 21-hydroxylase deficiency (21OHD), associates significant morbidity and mortality (5-7). Classic CAH due to 21OHD is characterized by a complex imbalance of adrenal steroids resulting in androgen excess, GC deficiency and, in two thirds of affected individuals, mineralocorticoid deficiency (8). Currently, it is almost impossible to mimic the complex circadian physiology of adrenal steroid biosynthesis by oral glucocorticoid replacement regimes (9). A challenge of GC treatment in CAH remains meeting the adequate balance between normalization of adrenal androgens, often requiring supraphysiological doses, and avoiding GC over-exposure, to minimize negative long-term health problems (10-12).

International guidelines aiming to optimize the medical management of CAH exist (13-15). However, they remain relatively broad and are likely to result in variable clinical practice. Furthermore, given the variability in health care provision between different countries, it is reasonable to anticipate geographical heterogeneity in the medical management of CAH. Thus, it is likely that the approach to hormonal replacement therapy in CAH is not uniform across the globe, which may represent an additional challenge to optimizing management and improving health care delivery. In this study we used information available through the International-CAH Registry (16) and provided evidence for significant variation in current practice of glucocorticoids and mineralocorticoid replacement in patients with CAH.

Patients and Methods

Study design, setting and participants

We conducted a retrospective international cohort study using data recorded in the I-CAH Registry (www.i-cah.org). The I-CAH Registry is an international database of pseudonymized information on patients with CAH and is approved by the National Research Ethics Service in the United Kingdom as a research database of information collected as part of routine clinical care (16). Following informed consent from patients or guardians, data are deposited within the registry by the endocrinologist supervising their management. All patients diagnosed with 21OHD for whom clinical information was recorded in the I-CAH Registry until December 2018 were included in the study. Data collection was conducted using the I-DSD/CAH data fields included in the basic module (register ID, center, country, year of birth, age on presentation, disorder type, actual diagnosis, sex assigned at birth, current gender) and longitudinal module (date of visit, age, weight, height, body surface area (BSA), cushingoid features, virilization, daily adherence to treatment, glucocorticoid type, glucocorticoid dose, timing of glucocorticoid dose, fludrocortisone dose, fludrocortisone frequency). For children data were collected from each visit for the first two years of life and then, the first medical visit in every year until 18 years of age; for adults, data collection included the first medical visit in every year over the last five years.

Data analysis

Hormone replacement analysis consisted of exploring the type of drug used (for glucocorticoids), total daily dose and dose for BSA, timing of administrations and distribution of doses throughout the day. BSA was calculated using the Mosteller

formula (17). We established the following age subgroups: less than one year, one to eight years, eight to 12 years, 12-18 years, 18-30 years and over 30 year, corresponding to infancy, early childhood, middle childhood, adolescence, young adult stage and adulthood respectively. Temporary variations and changes in treatment trends were explored by separating data before and after 2010, chosen in relation to the guidance published by The Endocrine Society (14).

We expressed GC doses for BSA in hydrocortisone equivalent, using the conversion rate: 20 mg hydrocortisone = 4 mg prednisolone/prednisone = 250 ⌈g dexamethasone = 25 mg cortisone acetate, in relation to the suppressive effect on growth of different types of synthetic steroids (18,19). As recommended by The Endocrine Society guidelines (15), target ranges for GC replacement in 21OHD were defined 10-15 mg hydrocortisone/ m^2/ day. Visits reporting intravenous doses of GC during acute deteriorations (4 patient visits) were excluded from the analysis.

Statistical analysis

Data were analyzed using descriptive statistics and analysis of variance, with appropriate adjustments of statistical tests used in accordance to data normality, tested graphically and by using the Shapiro-Wilk test. Hormone replacement doses between groups were compared by Kruskal Wallis H, Mann Whitney U and independent T tests. A *p* value of <0.05 was considered statistically significant throughout the analysis. Statistical analysis and computation were conducted using SPSS Statistics Software version 26 and GraphPad Prism version 8. In exploring aspects of gluco- and mineralocorticoid replacement (daily doses, number and timing of doses), we interpreted data from every patient visit as an independent variable.

For the descriptive analysis of different treatment practices in different countries, we only included countries that had recorded at least 50 patient visits.

Results

The initial dataset included 4732 patient visits that took place between 1982-2018, of which only 2.6% (6% patients) being classified as non-classic CAH (NCCAH). Moreover, 89.3% visits related to patients younger than 18 years, with adult visits only available from three countries (seven centres). Consequently, we limited our analysis to paediatric visits involving patients with classic 21 hydroxylase deficiency (**Supplementary Figure 1**). We analyzed 4174 visits recording information on 461 patients (54.9% females) from 16 countries and 31 centers (**Supplementary Table 1**).

Glucocorticoid treatment

Type of glucocorticoids used

Hydrocortisone was used for GC replacement in 90-100% visits by all countries with one exception: in Brazil cortisone acetate was used in 51.8% visits, hydrocortisone in 27.2%, dexamethasone in 13.2% and prednisolone/prednisone in 6.7%. Prednisone was used in a small number of cases: eight visits corresponding to six patients from Brazil and the United Kingdom. Dexamethasone and prednisolone were used in only 2% of visits in children younger than 12 years, and more frequently in children aged 12-18 years (dexamethasone 27.6% and prednisolone 7.8%). Hydrocortisone was most frequently administered following a three daily doses regimen (85%) (**Figure 1A).** In 8% of patients, different types of GC were used over time (**Supplementary Table 2**).

Glucocorticoid administration regimes

The timing of the GC replacement doses varied widely (**Figure 1B and 1C**). The majority of children following a three daily GC regime administered the first dose between 6:00 and 8:00, the second dose between 14:00 and 16:00 and the third dose between 22:00 and 23:00. For children taking four daily doses, the observed trend was for the first dose to be given at 4:00, the second dose between 11:00 and 12:00, the third dose between 16:00 and 17:00 and last dose at 21:00. Of the patient visits reporting three and four daily GC administrations, a circadian dosing regime was identified in 33.1% and a reverse circadian regime in 12.1% of cases, the remaining patients receiving at least two equal doses throughout the day. There was significant difference in the administration strategy between age groups ($p<0.001$), our results showing that a circadian regime was more frequently used by children aged 8-12 and 12-18 years (47.4% and 50.5% of patient visits, respectively), in comparison to younger children. We also found an important increase in the use of circadian regimes after 2010 for children older than one year of age (**Supplementary Table 3**).

Glucocorticoid daily doses for different age groups before and after 2010

Daily relative GC doses for BSA (**Table 1, Figure 2A**) varied significantly among age groups as shown by the Kruskal Wallis H test ($p<0.001$). Specifically, patients younger than one year had higher doses than children aged 1- 8 years ($p<0.001$) and 8-12 years ($p=0.030$), patients aged 1-8 years had smaller doses for BSA compared to all other age groups ($p<0.001$), while patients aged 12-18 years had the highest doses among all age groups with significant difference compared to the 1-8 years ($p<0.001$) and 8-12 years ($p=0.004$) group. Analyzing the clinical practice in

relation to the international guidelines (14,15), we found that overall the recommended upper limit of 15 mg /m^2/ day was exceeded in 37% of patients younger than 1 year of age, 21% of 1-8 year olds, 28% within the 8-12 years group and 39% of patients in the 12-18 years group. No significant difference in relative GC doses between genders were found, with the exception of patients aged 12-18 years, where females received higher doses than males (*p=0.002*). The fluctuation of GC doses before and after 2010 was inconsistent among age and gender groups and subgroups (**Table 1, Figure 2B, Supplementary Tables 4 and 5**).

Taking the variable growth-suppressive effect of different artificial GCs into consideration, we explored the variations in the HC-equivalent dose for BSA across age groups for the different types of drugs used (**Table 2**). Thus, the HC-equivalent doses for BSA were comparable to those of hydrocortisone in children treated with cortisone acetate for all age groups. The use of dexamethasone and prednisolone was limited to a very small number of cases, but there was a tendency to exceed the recommended dose range.

Glucocorticoid replacement practice in different countries

This analysis only included the 11 countries that recorded 50 or more patient visits. A comparison of hormone replacement between different countries revealed large variations in types of medication, doses and regimens used. While over 90% of countries/ centers used hydrocortisone as the preferred GC to treat children, in Brazil the use of cortisone acetate was reported in 51.8% of patient visits, due to limited availability of hydrocortisone. Of the 33 patients treated with cortisone acetate, four patients received exclusively cortisone acetate (during visits recorded before and

after 2010), 12 patients were initially treated with cortisone acetate, which was then changed to hydrocortisone at different ages between 1 month and 10 years (for all patients treatment with hydrocortisone was started after 2014), 14 patients were treated with cortisone acetate until 12-16 years of age, then the treatment was changed to dexamethasone; two patients initially received cortisone acetate, followed by prednisolone between 4–14 years and 10–15 years, respectively, then dexamethasone. The use of circadian administration regimes varied among countries between 0% and 51.3% of patient visits and that of reverse circadian, between 5.0% and 27.9% (**Supplementary Table 6)**. Exploring replacement doses, there were wide variations between countries, especially for children younger than 8 years of age **(Figure 3, Table 3, Supplementary Table 7)**. Observing visits recorded in neonates, we noted different strategies of initiating GC replacement, with six of ten countries using hydrocortisone doses above 7.5 mg/day and some having a large variation in doses as wide as 2–15 mg/day (corresponding to 6.6-71.8 mg/ m^2/ day HC-equivalent) (**Supplementary Table 8**). While for the majority of neonates GC replacement consisted of hydrocortisone, six neonates were started on cortisone acetate (dose range 0.15-10 mg/ day, HC-equivalent range 0.5 – 32.4 mg/ m^2/ day), five neonates started on prednisolone (dose range 0.4–1 mg/ day, 5-21.1 mg/ m^2/ day HC-equivalent) and one started on dexamethasone 0.075 mg/day (17.8 mg/ m^2/ day HC-equivalent). One center actively reduced the GC doses over the first 1-3 months of life, while in the other countries initial neonatal doses were maintained, with a slowly decreasing relative dose over time.

Fludrocortisone replacement

Ninety-two percent of patients were treated with fludrocortisone. The majority of patient visits (60.2%) reported a single daily dose of fludrocortisone, 29.8% two and 4.7% three daily doses; for the remaining visits fludrocortisone frequency was not specified.

The total daily dose of fludrocortisone ranged between 50-200 μg/ day for the vast majority of patients of all ages, however, relative fludrocortisone doses varied widely across age subgroups (**Figure 4**). We identified an increase in the dose after 2010 for all ages for both total and relative daily doses (**Table 4**), while no significant differences were detected between gender groups (**Supplementary Table 9**). Comparing doses of fludrocortisone between countries revealed variations across age groups (**Supplementary Table 10**).

Discussion

Our study explored global trends of hormone replacement therapy in children with 21OHD, providing the first general overview of the medical management of CAH for a wide range of countries across the world. Previous data on this topic consisted mainly of literature reviews and national cohort studies (20), which made comparisons difficult.

Overall, the types of GC used across different age groups was in line with the recommendation of international guidelines. Hydrocortisone was commonly used in children most likely based on its reduced half-life and risk of adverse effects, especially in relation to growth suppression (19,21,22). Of note, cortisone acetate was used in a relatively high number of children and young people in Brazil due to lack of availability of oral hydrocortisone preparations. Nevertheless, in over 90% of patients cortisone acetate was replaced by another GC at a later stage. We noted that in some cases this was done in younger, prepubertal children after 2014, most likely following better access to hydrocortisone preparations. In contrast, for patients who were born and diagnosed before 2000, dexamethasone replaced cortisone acetate after the age of 12 years. While we do not have information regarding the clinical reasoning that led to these changes, we can speculate that the timing is related to the completion of linear growth. Using cortisone acetate in children with 21OHD is in line with international recommendations; however, previous publications described its variable bioactivity in relation to the dependency on 11ß-hydroxysteroid dehydrogenase type 1, suggesting higher variability of therapeutic efficiency (23,24). We observed a number of children visits reporting the use of dexamethasone and prednisolone. The published guidelines advise against long-acting GC due to the

significant growth-suppressive effect, especially in the case of dexamethasone (14,15). Since the use of dexamethasone was mainly limited to children aged 12-18 years, one might argue that patients in this age group may have completed linear growth and had more significant problems with complying to a three-daily glucocorticoid regime. However, it is important to highlight the impact of the type of GC used on other health outcomes, as hydrocortisone has been also shown to be associated with lower prevalence of metabolic comorbidities and better bone health in CAH in comparison to dexamethasone (20,25,26).

The wide variations in the timing of the GC doses may relate to the ongoing debate regarding the optimal GC regimen. The majority of children received three or four daily doses of short acting GC, which resonates with previous research focused on optimizing hydrocortisone replacement. Earlier studies demonstrated the high bioavailability and fast clearance of hydrocortisone, recommending frequent and modest daily administrations to better mimic the physiological cortisol profiles (27). Furthermore, clinical studies in patients with adrenal insufficiency recommended the use of three rather than two daily hydrocortisone doses, based on improved cortisol profiles (28-30). We identified a rise in the use of circadian administration regimes after 2010, which is likely related to the increasing focus on glucocorticoid regimes following a more physiological circadian pattern (31). Despite limited research evidence assessing the benefits of circadian hormone replacement in CAH, it represents the most commonly employed therapy in clinical practice, based on theoretical and practical reasons, aiming to mimic the physiology of the hypothalamic-pituitary-adrenal axis (32). Moreover, it is well established that evening hydrocortisone doses are associated with increased disturbances of glucose

regulation, including insulin resistance, in comparison to morning administrations (33). .

Daily glucocorticoid doses exceeded the recommended range of 10-15 mg/ m^2/ day in a third of children younger than one year and between 12-18 years. However, there was a marked reduction in the percentage after 2010 for children younger than 8 years. This may relate to the publication of the international guidelines in 2002 and 2010 (13,14). The reason for the absence of a similar effect in children aged 8 to 18 years remains unclear. However, adolescents are known to have reduced compliance, together with an increasing degree of independence and reduced parental supervision (34). In addition, cortisol pharmacokinetics are altered at puberty in children with CAH by the decreased cortisol re-activation, secondary to reduced 11ß-hydroxysteroid dehydrogenase type 1 activity caused by the physiological rise of growth hormone and IGF1 (35,36).

A broad variation between different countries in the approach to GC replacement therapy was observed. This finding is not entirely unexpected considering previously reported variations in GC regimens for different centers within the United Kingdom (37). In some countries the recommended glucocorticoid dose range for children was exceeded in as many as 75% of patient visits, while other countries recorded doses below 10 mg/m^2 per day in up to 57% of cases. Overall, infants received very high relative doses of GC reaching above 30 mg HC-equivalent/ m^2 per day. In neonates there were particularly marked differences in clinical practice, with half of the countries using hugely variable dose ranges between 4.2–75 mg/ m^2 per day HC-equivalent. Only one center actively reduced GC doses over the weeks following

initiation of treatment in a consistent manner. Importantly, studies analyzing growth in children with simple virilizing CAH and delayed treatment found accelerated growth only after the first 12-18 months of life (38,39), which appears to be a consequence of androgen insensitivity in infancy (40). In addition, the dose-dependent negative effect of GC on linear growth is known and the suppressive action is more marked during age intervals of high growth velocity, including infancy (41,42). Moreover, there is increased risk of developing metabolic and cardiovascular comorbidities in CAH (43), associated with chronic GC overexposure (44). Although such comorbidities are only becoming fully apparent in adulthood, there is increasing evidence of the onset during childhood (45). These points emphasize the importance of treatment with the lowest possible dose of glucocorticoids by actively reducing cumulative GC doses from a young age starting in infants and young children.

The large majority of fludrocortisone doses in children were within the recommended range of 50-200 µg/ day (15). However, high relative doses were used in younger individuals, with doses up to 800 µg/ m^2 per day and 600 µg/ m^2 per day in the under 1 year old and 1-8 years old group, respectively. This trend was consistent across different countries. We could not assess the practice in relation to the dosing strategies used and in the absence of information regarding clinical and biochemical standards of control it is not possible to estimate overtreatment. While there is evidence of renal resistance to mineralocorticoids in infants (46,47), it was also shown that fludrocortisone treatment is associated with hypertension in children with CAH (48) and there is a correlation between blood pressure and fludrocortisone dose in infants (49). Thus, our findings indicate that the use of absolute doses may

lead to overtreatment with fludrocortisone, suggesting a potential benefit to using relative doses of fludrocortisone at least in young children to fine-tune mineralocorticoid replacement.

The aim of this study was to provide an overview on the current practice of hormone replacement therapy in patients with CAH and we focused mainly on the types of medication used, doses and timing of administration. Thus, we acknowledge the absence of clinical and biochemical data as a weakness of the study and identify the need in the future for more in-depth analysis, including such information, in order to increase the clinical relevance of these findings. Another limitation related to data collection, consisting of the overall modest number of countries providing information on patients with CAH by using the I-CAH Registry, as a limited amount of data was available outside of Europe and South America. Moreover, the small number of adult patient visits recorded led to their exclusion from the analysis. However, the I-CAH registry allowed for analysis of a very large dataset in a rare condition, highlighting the value of real-world data and the benefit of using an international registry as a platform for exploring global clinical practice to address questions relevant to improving patient management.

Overall, our results suggest that hormone replacement therapy in children and young persons with classic 21OHD varies widely across different age groups and different countries. It appears that some of these discrepancies relate to physicians' preference rather than only to physiological difference between individuals. To understand these differences in clinical practice further evidence exploring the interdependency of different management strategies including CAH monitoring

methods is urgently warranted. In addition, future research should explore if different clinical practices are associated with differences in long-term outcome in CAH.

Abbreviations

21OHD: 21hydroxylase deficiency

BSA: body surface area

CAH: congenital adrenal hyperplasia

DSD: disorders of sexual development

GC: glucocorticoid

HC: hydrocortisone

I-DSD/CAH: International–DSD/CAH (Registry)

IGF1: insulin-like growth factor 1

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Author Contributions

Nils P Krone, Irina Bacila, Jillian Bryce, Salma Ali, and S Faisal Ahmed conceptualized and designed the study, designed the data collection instruments, coordinated and supervised data collection, performed the initial analyses, drafted the initial manuscript, and approved the final manuscript as submitted. Irina Bacila

and Nicole Freeman undertook the statistical analyses. All authors contributed to data acquisition, revision of the manuscript and have read and approved the final report. All authors take public responsibility and accountability for the results.

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Data Availability

The datasets generated or analysed during the current study are not available publicly but available to access through a data sharing agreement available at https://idsdorg.files.wordpress.com/2020/05/i-dsd-i-cah-data-sharing-agreement.docx

Disclosure Summary

RJMR is a Director of Diurnal Ltd.

Hedi L Claahsen-van der Grinten and Jeremy W Tomlinson are on the editorial board of EJE. They were not involved in the review or editorial process for this paper, on which they are listed as an author.

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Figure legends

Figure 1. Type of glucocorticoids and number of daily doses corresponding to different types of glucocorticoids used in children with CAH (**A**). Timing of glucocorticoid doses for children on three daily doses (**B**) and children on four daily doses regimes (**C**). Each bar represents the number of patient visits recording a dose given at that time; the different patterns correspond to the order of the doses throughout the day.

Figure 2. A. Glucocorticoid doses expressed as relative hydrocortisone equivalent (mg/ m^2/ day) in different age groups. The shaded area indicates the recommended dose range of 10 - 15mg/ m^2/ day. The black horizontal lines indicate the median with the interquartile range (error bars) for age group**. B.** Glucocorticoid doses expressed as hydrocortisone equivalent (mg/ m^2/ day) in different age groups before (clear circles) and after (black squares) 2010. For each subgroup the circles or squares correspond to the median and the error bars to the interquartile range. (*statistical significance in comparing doses before and after 2010). HC = hydrocortisone

Figure 3. Glucocorticoid doses used in children from different countries, expressed as hydrocortisone equivalent (mg/ m^2/ day). The shaded area indicates the recommended dose range of 10-15mg/ m^2/ day. The black horizontal lines indicate the median with the interquartile range (error bars) for each country. The countries included in the analysis were anonymized. We only included in the analysis countries that had recorded at least 50 patient visits. HC = hydrocortisone

Figure 4. Absolute (**A**) and relative (**B**) fludrocortisone (FC) doses for different age groups, expressed in µg /day and µg/ m^2/ day, respectively. The black horizontal lines indicate the median with the interquartile range (error bars) for age group.

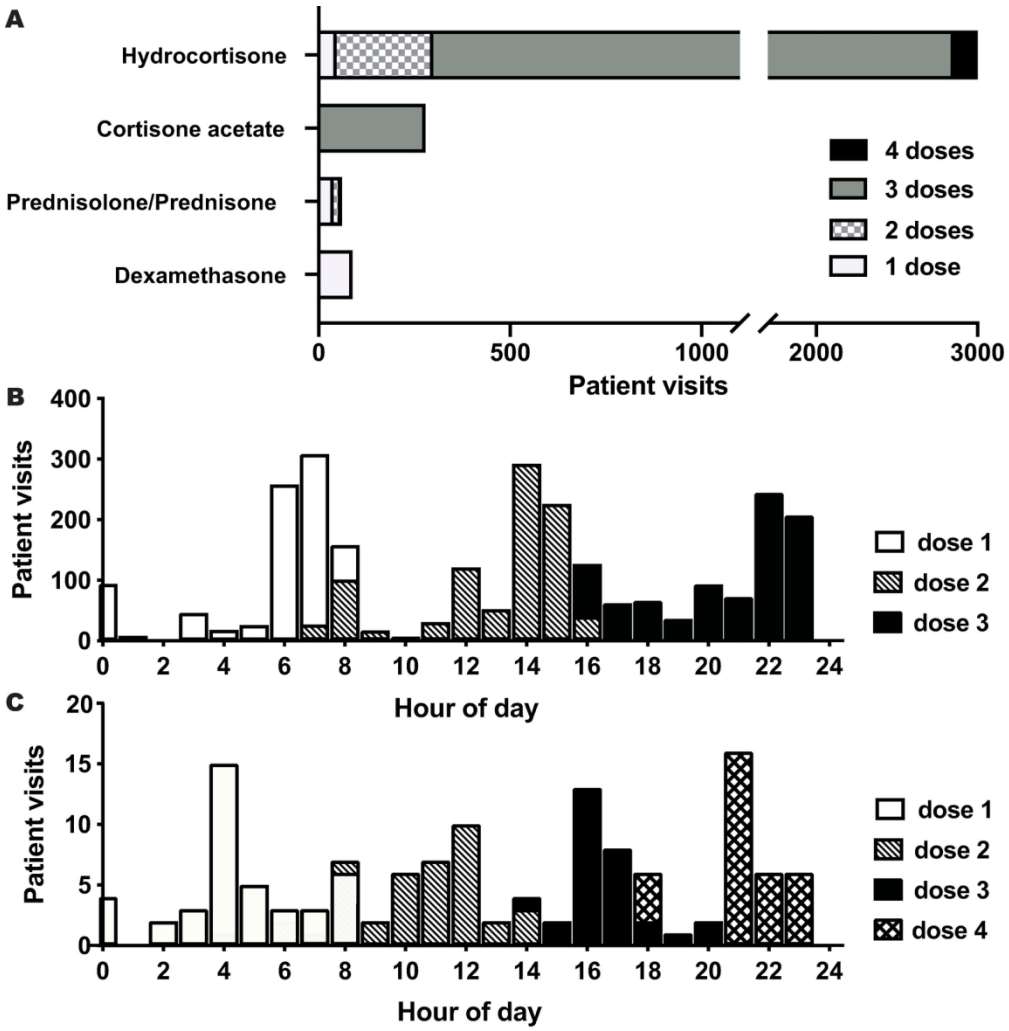

Figure 1. Type of glucocorticoids and number of daily doses corresponding to different types of glucocorticoids used in children with CAH (A). Timing of glucocorticoid doses for children on three daily doses (B) and children on four daily doses regimes (C). Each bar represents the number of patient visits recording a dose given at that time; the different patterns correspond to the order of the doses throughout the day.

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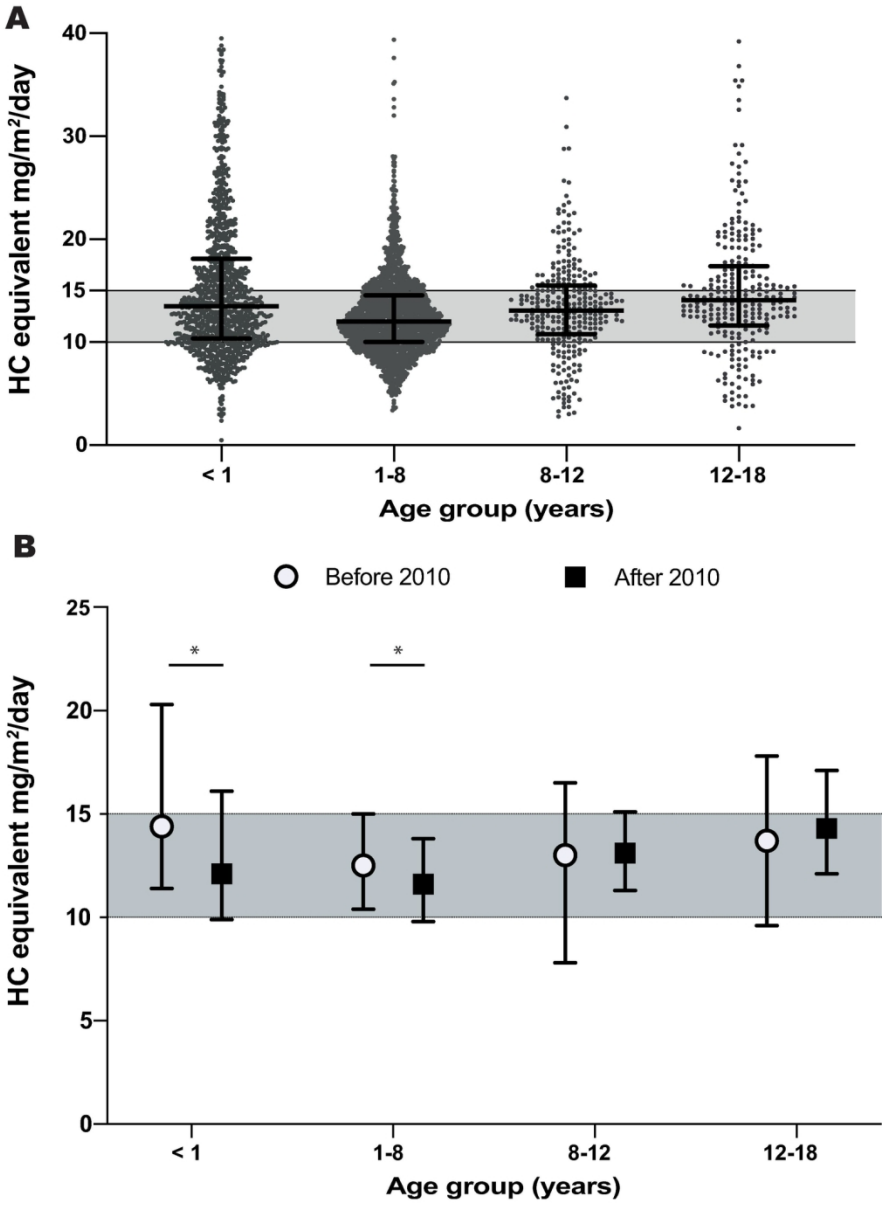

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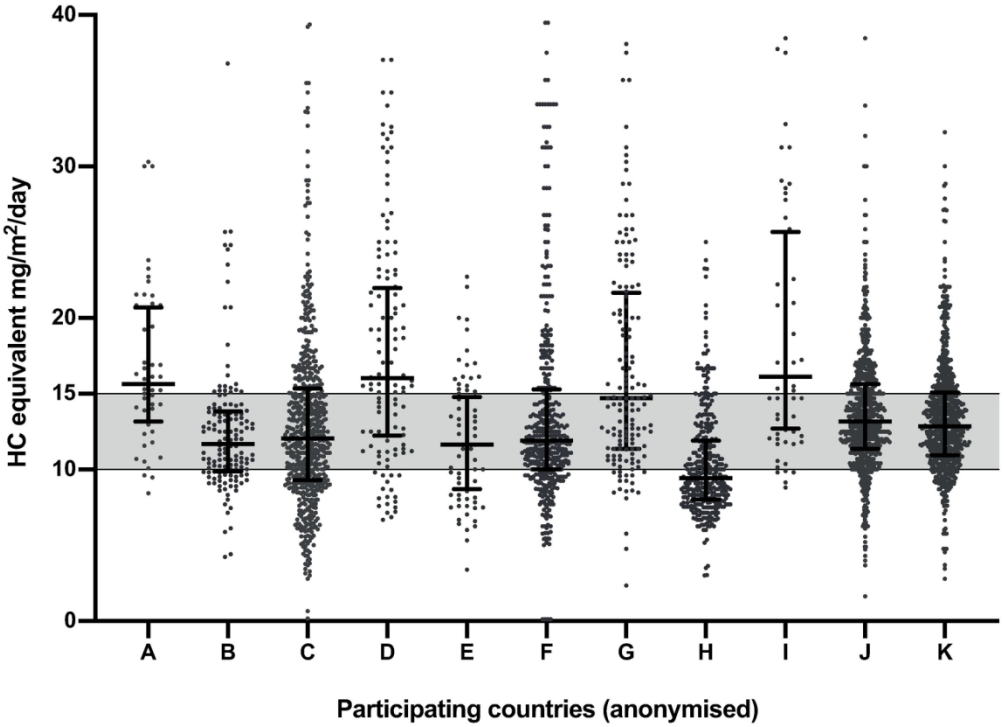

Figure 3. Glucocorticoid doses used in children from different countries, expressed as hydrocortisone equivalent (mg/ m2/ day). The shaded area indicates the recommended dose range of 10-15mg/ m2/ day. The black horizontal lines indicate the median with the interquartile range (error bars) for each country. The countries included in the analysis were anonymized. We only included in the analysis countries that had recorded at least 50 patient visits. HC = hydrocortisone

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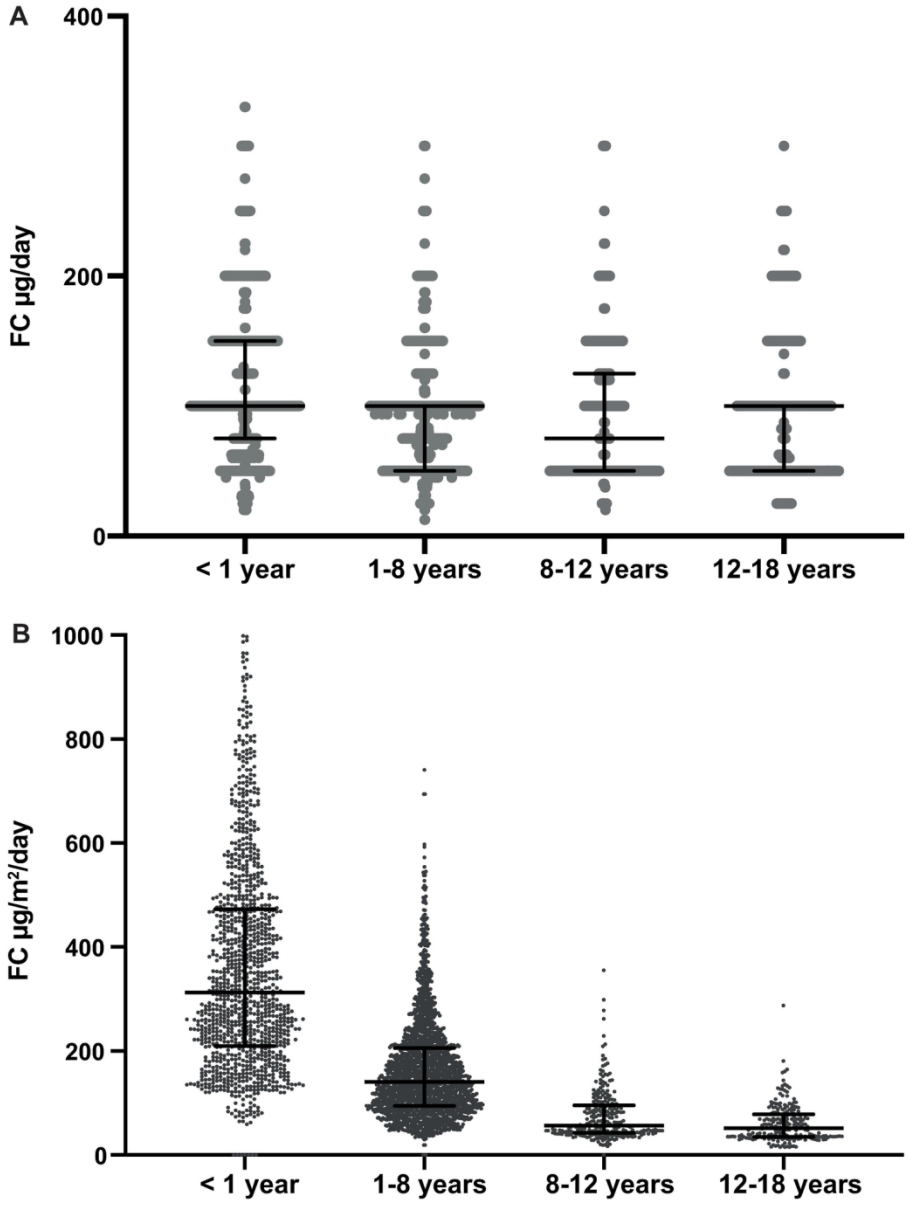

Figure 4. Absolute (A) and relative (B) fludrocortisone (FC) doses for different age groups, expressed in µg /day and µg/ m2/ day, respectively. The black horizontal lines indicate the median with the interquartile range (error bars) for age group.

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Table 1. Glucocorticoid doses for different age groups, before and after 2010

Age group	Overall	Before 2010	After 2010	Before vs after 2010
0 – 1 years	**13.4 (10.3 – 18.1)** [N=375, n=1037]	**14.3 (11.4 – 20.3)** [N=197, n=527]	**12.2 (9.9 – 16.1)** [N=178, n=546]	*p<0.001*
1 – 8 years	**12.0 (10.0 – 14.5)** [N=420, n=1893]	**12.5 (10.3 – 15.0)** [N=187, n=823]	**11.6 (9.8 – 13.9)** [N=233, n=1061]	*p<0.001*
8 – 12 years	**13.0 (10.7 – 15.5)** [N=89, n=288]	**13.0 (7.8 – 16.5)** [N=24, n=104]	**13.0 (11.3 – 15.1)** [N=65, n=184]	*p=0.553*
12 – 18 years	**14.0 (11.6 – 17.4)** [N=78, n=259]	**13.7 (9.6 – 17.8)** [N=22, n=114]	**14.3 (12.1 – 17.1)** [N=56, n=145]	*p=0.235*

The doses are expressed as hydrocortisone (HC)-equivalent (mg/ m^2/ day – median with interquartile range). The last column presents the statistical difference in doses between the two time intervals. [N = number of patients and n = number of patient visits available for the analysis]

Table 2. Relative doses for different types of glucocorticoids used in children

Age group	Hydrocortisone	Cortisone acetate	Dexamethasone	Prednisolone
0 – 1 years	**13.5 (10.4 – 18.3)** [N=338, n=997]	**11.4 (9.6 – 15.4)** [N=22, n=55]	**17.6 – 20.5** [N=3, n=3]	**8.7 (6.4 – 16.0)** [N=4, n=18]
1 – 8 years	**12.0 (10.0 – 14.4)** [N=344, n=1740]	**12.6 (10.1 – 16.5)** [N=23, n=139]	**16.7** [N=1, n=1]	**6.1 (5.4 – 22.5)** [N=5, n=13]
8 – 12 years	**12.9 (11.3 – 15.2)** [N=68, n=209]	**13.0 (7.6 – 16.0)** [N=17, n=73]	**20.5 – 24.2** [N=3, n=3]	**20.8 – 19.8** [N=2, n=3]
12 – 18 years	**13.6 (11.0 – 15.8)** [N=46, n=114]	**14.0 (8.8 – 14.8)** [N=12, n=44]	**13.8 (12.0 – 20.8)** [N=20, n=81]	**21.3 (13.9 – 25.4)** [N=11, n=20]

The doses are expressed as HC-equivalent (mg/ m^2/ day – median with interquartile range). [N = number of patients and n = number of patient visits on which the analysis was based]

Table 3. Glucocorticoid doses used in children from different countries

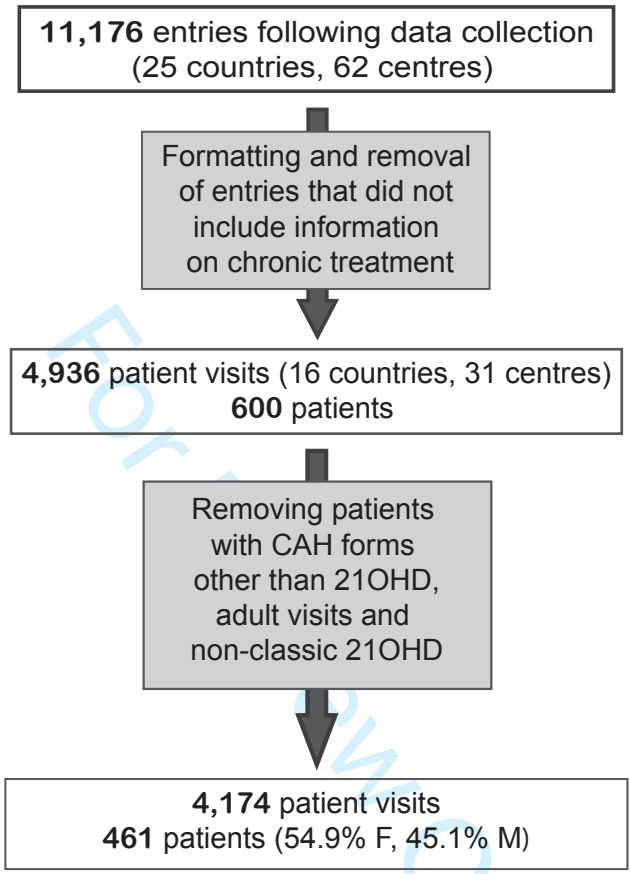
Country (anonymized)	Daily GC doses (HC-equivalent mg/ m^2/ day)		
	< 10	**10 - 15**	**> 15**
A [N=13, n=56]	5.4%	35.7%	58.9%
B [N=24, n=141]	27.0%	56.0%	17.0%
C [N=57, n=599]	32.2%	41.0%	26.7%
D [N=17, n=145]	12.3%	28.1%	58.9%
E [N=22, n=88]	37.5%	38.6%	23.9%
F [N=57, n=441]	24.0%	50.1%	25.9%
G [N=27, n=153]	12.4%	41.8%	45.8%
H [N=56, n=350]	57.3%	32.5%	10.0%
I [N=9, n=50]		24.5%	75.5%
J [N=81, n=760]	11.3%	59.0%	29.6%
K [N=78, n=693]	13.5%	59.5%	25.2%

The results are expressed in percentages in relation to number of patient visits. Only the countries with a number of patient visits ≥ 50 were included in the analysis. [N = number of patients and n = number of patient visits on which the analysis was based]

Table 4. Fludrocortisone doses for different age groups, before and after 2010

Age group	Dose unit	Overall	Before 2010	After 2010	Before vs after 2010
					Mann-Whitney U
0 – 1 y	**µg/ day**	**100 (75 – 150)** [N=379,n=1218]	**100 (62.5 – 150)** [N=201,n=621]	**100 (75 – 150)** [N=178,n=597]	*p<0.001*
	µg/ m²/ day	**312 (209 – 473)** [N=372,n=1143]	**279 (188 – 416)** [N=195,n=567]	**365 (228 – 536)** [N=177,n=576]	*p<0.001*
1 – 8 y	**µg/ day**	**100 (50 – 100)** [N=417,n=2142]	**94 (50 – 100)** [N=185,n=921]	**100 (50 – 100)** [N=232,n=1221]	*p=0.016*
	µg/ m²/ day	**139 (94 – 205)** [N=414,n=2093]	**149 (99 – 212)** [N=181,n=877]	**134 (90 – 195)** [N=233,n=1216]	*p=0.001*
8 – 12 y	**µg/ day**	**75 (50 – 125)** [N=85,n=278]	**50 (50 – 75)** [N=21,n=81]	**100 (50 – 150)** [N=64,n=197]	*p<0.001*
	µg/ m²/ day	**56 (42 – 95)** [N=82,n=262]	**45 (38 – 55)** [N=19,n=76]	**74 (44 – 107)** [N=63,n=186]	*p<0.001*
12 – 18 y	**µg/ day**	**100 (50 – 100)** [N=72,n=238]	**50 (50 – 50)** [N=19,n=75]	**100 (60 – 150)** [N=53,n=163]	*p<0.001*
	µg/ m²/ day	**51 (34 – 77)** [N=71,n=221]	**35 (29 – 44)** [N=18,n=74]	**63 (41 – 91)** [N=53,n=147]	*p<0.001*

Absolute doses are expressed in µg/ day and relative doses in µg/ m²/ day. The doses are expressed as median with interquartile range. The last column presents the statistical difference in doses between the two time intervals. [N = number of patients and n = number of patient visits where data was available for the analysis]



Supplementary Figure 1. Data collection pathway. Data collection resulted in a dataset of 11,176 recorded events from 62 centers in 25 countries. Formatting the raw data, removing "null" entries and duplicates and patients suffering from CAH due to causes other than 21OHD, resulted in a set of 4,174 patient visits recorded for 461 patients from 16 countries and 31 centers. (M: males, F: females, CAH: congenital adrenal hyperplasia, 21OHD: 21-hydroxylase deficiency).

Supplementary Table 1. Geographical distribution of participants (patients and patient visits)

Country (Centre)	Patients	Patient visits
	Number (Percentage)	Number (Percentage)
Argentina (Buenos Aires)	14 (3.0%)	100 (2.4%)
Belgium (Ghent)	25 (5.4%)	228 (5.5%)
Brazil (Sap Paulo, Porto Alegre)	57 (12.4%)	628 (15.0%)
Bulgaria (Varna)	1 (0.2%)	1 (0.02%)
Denmark (Aarhus)	17 (3.7%)	151 (3.6%)
Egypt (Cairo Ain Shams University)	22 (4.8%)	198 (4.7%)
Germany (Berlin, Magdeburg, Munich Technical University)	57 (12.4%)	474 (11.2%)
Hungary (Budapest)	1 (0.2%)	2 (0.02%)
Israel (Petah-Tikvah)	4 (0.9%)	24 (0.6%)
Italy (Bologna, Torino-Regina Margherita Children's Hospital)	27 (5.9%)	232 (5.6%)
Netherlands (Nijmegen, Amsterdam, Utrecht, Rotterdam)	57 (12.4%)	450 (10.8%)
Romania (Craiova)	1 (0.2%)	10 (0.2%)
Serbia (Belgrade)	9 (2.0%)	56 (1.3%)
Sri Lanka (Colombo)	7 (1.2%)	12 (0.3%)
Turkey (Istanbul Marmara, Istanbul University, Istanbul Zeynep Kamil)	81 (17.6%)	771 (18.5%)
United Kingdom (Birmingham, Cambridge, Glasgow, London, Manchester, Sheffield Children's Hospital, Sheffield Hallamshire Hospital)	81 (17.6%)	837 (20.1%)
Total	461	4174

The results are expressed in numbers and percentages.

Supplementary Table 2. Patients treated with multiple types of glucocorticoids over time

Types of glucocorticoids (used chronologically)	Number of patients
Cortisone acetate, Dexamethasone	11
Cortisone acetate, Hydrocortisone	11
Hydrocortisone, Prednisolone	10
Cortisone acetate, Dexamethasone, Prednisolone	7
Dexamethasone, Hydrocortisone	3
Cortisone acetate, Hydrocortisone, Prednisolone	2
Cortisone acetate, Prednisolone	1

Supplementary Table 3. The type of GC administration regimen (circadian vs reverse circadian) in different age groups, before and after 2010.

Age group	GC regimen	Overall	Before 2010	After 2010	Chi-Square Test (before vs after 2010)
0 – 1 years [N=260, n=806]	Circadian	25.1%	21.7%	28.8%	***p = 0.066***
	Reverse circadian	8.2%	8.7%	7.6%	
	Other:	66.7%	69.6%	63.6%	
	E	54%	57.8%	49.7%	
	MA	2.9%	4.7%	0.8%	
	ME	9.6%	6.8%	12.6%	
	AE	0.4%	0.2%	0.5%	
1 – 8 years [N=283, n=1467]	Circadian	34.4%	23.7%	42.5%	***p < 0.001***
	Reverse circadian	14.2%	16.1%	12.7%	
	Other:	51.5%	60.3%	44.8%	
	E	25.6%	31.9%	20.8%	
	MA	5.5%	7.1%	4.2%	
	ME	19.4%	19.9%	19.1%	
	AE	1.0%	1.4%	0.7%	
8 – 12 years [N=63, n=196]	Circadian	47.4%	13.8%	53.3%	***p = 0.002***
	Reverse circadian	13.3%	17.2%	12.6%	
	Other:	39.3%	69.0%	34.1%	
	E	19.4%	34.5%	16.8%	
	MA	7.1%	10.3%	6.6%	
	ME	9.7%	17.2%	8.4%	
	AE	3.1%	6.9%	2.4%	
12 –18 years [N=42, n=101]	Circadian	50.5%	0%	53.7%	***p = 0.005*** (NB: Only 6 visits before 2010)
	Reverse circadian	11.9%	0%	12.6%	
	Other:	37.6%	100%	33.7%	
	E	8.9%	16.7%	8.4%	
	MA	11.9%	16.7%	11.6%	
	ME	12.9%	66.7%	9.5%	
	AE	4.0%		4.2%	

Results are expressed in percentages in relation to the number of patient visits. Other administration regimes: E = all daily doses were even; MA = the morning and afternoon doses were even and higher than the evening dose; ME = morning and evening doses were even

and higher than the afternoon dose; AE = the afternoon and evening doses were even and higher than the morning dose. [N = number of patients and n = number of patient visits on which the analysis was based]

Supplementary Table 4. Glucocorticoid doses in relation to the recommended dose range

Age group [Before 2010]	Before 2010			After 2010			Age group [After 2010]
	<10 mg/m²	**10-15 mg/m²**	**>15 mg/m²**	**<10 mg/m²**	**10-15 mg/m²**	**>15 mg/m²**	
0 – 1 y [N=197, n=526]	16.6%	36.7%	46.1%	26.2%	43.6%	29.5%	**0 – 1 y** [N=178, n=546]
1 – 8 y [N=188, n=832]	21.9%	51.8%	25.8%	26.8%	55.6%	17.4%	**1 – 8 y** [N=233, n=1061]
8 – 12 y [N=24, n=104]	32.7%	32.7%	34.6%	10.8%	62.9%	25.3%	**8 – 12 y** [N=65, n=184]
12 –18 y [N=22, n=114]	24.3%	40.9%	33.9%	11.6%	44.5%	43.2%	**12 –18 y** [N=56, n=145]

Results are expressed in percentages in relation to the number of patient visits. [N = number of patients and n = number of patient visits on which the analysis is based]

Supplementary Table 5. Glucocorticoid doses for age and gender groups, before and after 2010

Age group	Before 2010		After 2010		Before vs after 2010 Females	Before vs after 2010 Males
	Females	Males	Females	Males		
0 – 1 years	**15.2 (11.4–21.2)** [N=108,n=282]	**14.2 (11.1–19.5)** [N=89,n=245]	**12.4 (9.9–16.3)** [N=98,n=305]	**11.8 (9.9–15.6)** [N=80,n=241]	***p<0.001***	***p<0.001***
	p =0.121		***p =0.660***			
1 – 8 years	**12.8 (10.6–15.6)** [N=100,n=454]	**12.1 (10.0–14.6)** [N=87,n=378]	**11.5 (9.7–13.7)** [N=126,n=589]	**11.7 (9.9–14.3)** [N=107,n=472]	***p<0.001***	***p=0.524***
	p <0.001		***p =0.124***			
8 – 12 years	**14.0 (7.8–16.5)** [N=15,n=69]	**12.3 (7.1–16.4)** [N=9,n=35]	**12.9 (11.0–15.0)** [N=31,n=93]	**13.1 (11.4–15.2)** [N=34,n=91]	***p=0.728***	***p=0.394***
	p =0.744		***p =0.821***			
12 – 18 years	**14.3 (12.4–19.3)** [N=14,n=76]	**12.2 (8.0–14.4)** [N=8,n=38]	**14.3 (11.5–20.7)** [N=31,n=81]	**14.2 (12.3–15.8)** [N=25,n=64]	***p=0.983***	***p=0.009***
	p =0.001		***p =0.201***			

The doses are expressed as hydrocortisone (HC)-equivalent (mg/ m^2/ day – median with interquartile range). The *p*-values correspond to the statistical significance of the differences in doses between genders and time intervals. [N = number of patients and n = number of patient visits on which the analysis was based]

Supplementary Table 6. The type of GC administration regimen (circadian vs reverse circadian) in different countries.

Country (anonymized)	GC regimen					
	Circadian	Reverse Circadian	Other			
			E	MA	ME	AE
A [N=9, n=37]	2.7%	16.2%	35.1%	2.7%	43.2%	
B [N=22, n=140]	28.6%	5.0%	27.9%	0.7%	36.4%	1.4%
C [N=18, n=61]	0%	27.9%	41.0%		27.9%	3.3%
D [N=17, n=145]	41.4%	18.6%	40.0%	11.0%	27.6%	
F [N=57, n=454]	43.4%	7.3%	26.0%	4.8%	18.3%	0.2%
G [N=25, n=130]	10.0%	10.0%	59.2%	5.4%	15.4%	
H [N=55, n=338]	14.2%	24.9%	49.1%	1.2%	10.4%	0.3%
J [N=49, n=492]	21.1%	17.7%	38.2%	8.9%	10.2%	3.9%
K [N=70, n=706]	51.3%	5.1%	25.8%	6.8%	10.6%	0.4%

Results are expressed in percentages in relation to the number of patient visits. Other administration regimes: E = all daily doses were even; MA = the morning and afternoon doses were even and higher than the evening dose; ME = morning and evening doses were even and higher than the afternoon dose; AE = the afternoon and evening doses were even and higher than the morning dose. [N = number of patients and n = number of patient visits where data was available for the analysis]

Supplementary Table 7. Glucocorticoid doses used by different countries for children

Country (anonymized)	<1 year	1-8 years	8-12 years	12-18 years
A	**16.6 (14.6–21.8)** [N=12, n=22]	**14.5 (11.5–18.3)** [N=9, n=28]	**20.7(14.1–21.2)** [N=2, n=5]	
B	**10.5 (9.1–12.6)** [N=13, n=29]	**11.5 (9.8–13.5)** [N=17, n=89]	**14.2(10.2–22.3)** [N=7, n=11]	**14.5(13.0–25.3)** [N=4, n=12]
C	**10.7 (9.3–13.7)** [N=43, n=154]	**11.8 (9.4–15.3)** [N=39, n=218]	**12.2 (6.9–16.1)** [N=21, n=90]	**13.7 (10.6–19.1)** [N=21, n=137]
D	**21.6 (17.2– 28.4)** [N=16, n=56]	**14.2 (11.1–16.9)** [N=17, n=86]		
E	**10.7 (8.7–13.8)** [N=18, n=29]	**12.2 (8.1–15.2)** [N=19, n=59]		
F	**13.7 (11.0–22.2)** [N=51, n=192]	**11.1 (9.4–12.8)** [N=50, n=242]	**11.9 (11.0-12.9)** [N=3, n=5]	
G	**21.8 (16.7–26.2)** [N=22, n=64]	**12.2 (10.4–14.6)** [N=23, n=88]		
H	**10.0 (8.2–13.4)** [N=53, n=165]	**9.0 (7.7–10.9)** [N=48, n=185]		
I	**26.7 (15.8–35.4)** [N=8, n=20]	**19.0 (14.6–26.5)** [N=7, n=19]	**15.2(11.9–25.6)** [N=3, n=7]	**15.3 – 22.9** [N=1, n=3]
J	**14.1 (12.0–16.8)** [N=73, n=181]	**12.8 (11.0–15.0)** [N=75, n=483]	**14.3 (12.2-17.2)** [N=14, n=74]	**15.3 (12.2–16.3)** [N=7, n=23]
K	**14.0 (11.6–18.6)** [N=44, n=144]	**12.4 (10.8–14.5)** [N=55, n=374]	**12.5(10.7–13.9)** [N=34, n=94]	**13.2 (11.4–16.4)** [N=33, n=80]

The doses are expressed as HC-equivalent in mg/ m^2/ day (median with interquartile range). Only the countries with an overall number of patient visits ≥ 50 were included in the analysis. [N = number of patients, n = number of patient visits]

Supplementary Table 8. Hydrocortisone doses used in neonates by different countries

Country (anonymized)	Total daily dose range (mg/ day)	Dose for BSA median (interquartile range) (mg/ m^2/ day)
A [N=3, n=3]	**5.0**	**23.7**
B [N=11, n=13]	**2- 15**	**11.9 (7.6 – 13.5)**
C [N=15, n=31]	**0.12 - 8**	**10.8 (6.6 – 16.3)**
D [N=10, n=11]	**3 - 9**	**24.6 (20.7 – 31.3)**
E [N=3, n=4]	**2.5 - 5**	**12.8 (10.7 – 20.5)**
F [N=40, n=43]	**3 - 10**	**32.7 (26.2 – 34.2)**
G [N=12, n=16]	**2.4 - 8.3**	**27.8 (23.0 – 34.0)**
H [N=46, n=51]	**1 - 6.5**	**13.5 (11.5 – 15.5)**
I [N=4, n=6]	**7.5 - 15**	**44.9 (35.4 – 71.8)**
K [N=24, n=32]	**1 - 7.5**	**18.7 (13.9 – 20.7)**

The doses are expressed as total daily dose range and dose for BSA (median with interquartile range). [N = number of patients and n = number of patient visits where data was available for the analysis]

Supplementary Table 9. Fludrocortisone doses for age and gender subgroups

Age group	Dose unit	Females	Males	Females vs males (Independent T test)
0 – 1 years	**µg/ day**	**100 (62.5 – 150)** [N=210, n=672]	**100 (75 – 150)** [N=169, n=546]	*p=0.390*
	µg/ m²/ day	**315 (209 – 482)** [N=204, n=628]	**309 (215 – 465)** [N=168, n=515]	*p=0.569*
1 – 8 years	**µg/ day**	**94 (50 – 100)** [N=223, n=1173]	**100 (50 – 100)** [N=194, n=969]	*p=0.972*
	µg/ m²/ day	**139 (94 – 207)** [N=220, n=1142]	**142 (93 – 204)** [N=194, n=951]	*p=0.754*
8 – 12 years	**µg/ day**	**50 (50 – 120)** [N=44, n=151]	**100 (50 – 150)** [N=41, n=127]	*p=0.055*
	µg/ m²/ day	**52 (43 – 92)** [N=41, n=144]	**62 (38 – 102)** [N=41, n=118]	*p=0.838*
12 – 18 years	**µg/ day**	**87.5 (50 – 100)** [N=43, n=151]	**100 (50 – 150)** [N=29, n=87]	*p=0.024*
	µg/ m²/ day	**48 (34 – 70)** [N=42,n=137]	**54 (32 – 93)** [N=29,n=84]	*p=0.217*

Absolute doses are expressed in µg/ day and relative doses in µg/ m^2/ day. The doses are expressed as median with interquartile range. The last column presents the statistical difference in doses between the two time intervals. [N = number of patients and n = number of patient visits where data was available for the analysis]

Supplementary Table 10. Fludrocortisone doses in different age groups of children from different countries

Country	Dose unit	<1 year	1-8 years	8-12 years	12-18 years
A	µg/ day	**100 (50-100)** [N=13, n=39]	**50 (50-100)** [N=12, n=50]	**50 (50-50)** [N=3, n=9]	
	µg/ m²/ day	**280 (150-478)** [N=13, n=34]	**109 (66-182)** [N=12, n=48]	**37 (29-52)** [N=3, n=9]	
B	µg/ day	**100 (90-160)** [N=13, n=43]	**80 (60-100)** [N=15,n=129]	**120 (62.5-120)** [N=6, n=17]	**100 (60-100)** [N=4, n=15]
	µg/ m²/ day	**249 (209-670)** [N=13, n=35]	**97 (82-175)** [N=15, n=122]	**88 (60-107)** [N=6, n=15]	**56 (41-67)** [N=4, n=15]
C	µg/ day	**200 (150-200)** [N=43, n=161]	**100 (50-150)** [N=34, n=213]	**50 (50-50)** [N=17, n=68]	**50 (50-50)** [N=16, n=92]
	µg/ m²/ day	**579 (444-730)** [N=43, n=159]	**156 (78-272)** [N=35, n=213]	**43 (36-49)** [N=17, n=68]	**35 (30-41)** [N=16, n=92]
D	µg/ day	**100 (75-150)** [N=16, n=56]	**100 (75-150)** [N=16, n=86]		
	µg/ m²/ day	**345 (218-443)** [N=16, n=56]	**216 (153-269)** [N=16, n=84]		
E	µg/ day	**100 (75-100)** [N=18, n=43]	**100 (50-100)** [N=19, n=136]		
	µg/ m²/ day	**274 (228-390)** [N=18, n=42]	**126 (85-190)** [N=19, n=134]		
F	µg/ day	**100 (75-150)** [N=50, n=197]	**75 (50-100)** [N=48, n=250]	**50 (50-50)** [N=1, n=7]	
	µg/ m²/ day	**325 (220-486)** [N=49, n=186]	**127 (96-169)** [N=48, n=248]	**42 (40-46)** [N=3, n=7]	
G	µg/ day	**100 (50-100)** [N=23, n=82]	**75 (50-100)** [N=26, n=131]		
	µg/ m²/ day	**241 (166-351)** [N=22, n=78]	**149 (94-192)** [N=26, n=130]		
H	µg/ day	**62.5(62.5-100)** [N=54,n=216]	**62.5(62.5-00)** [N=50, n=216]		
	µg/ m²/ day	**216 (153-312)** [N=52, n=194]	**141 (106-188)** [N=50, n=214]		
I	µg/ day	**50 (50-50)** [N=6, n=19]	**50 (50-50)** [N=6, n=22]	**100 (50-100)** [N=2, n=5]	
	µg/ m²/ day	**192 (144-250)** [N=6, n=19]	**94 (78-105)** [N=6, n=21]	**86 (34-94)** [N=2, n=5]	

J	**µg/day**	**100 (100-150)** [N=71, n=176]	**100 (50-100)** [N=71, n=473]	**87.5 (50-125)** [N=12, n=61]	**82 (50-100)** [N=6, n=21]
	µg/m^2/ day	**323 (243-446)** [N=71, n=176]	**138 (87-209)** [N=71, n=473]	**63 (44-89)** [N=12, n=61]	**47 (32-61)** [N=6, n=21]
K	**µg/day**	**100 (100-150)** [N=44, n=166]	**100 (75-150)** [N=56, n=413]	**125 (75-150)** [N=36, n=108]	**100(100-200)** [N=34,n=103]
	µg/m^2/ day	**311 (217-416)** [N=46, n=145]	**151 (108-212)** [N=56, n=382]	**90 (53-129)** [N=34, n=94]	**82 (62-99)** [N=33, n=86]

Absolute doses are expressed in µg/ day and relative doses in µg/ m^2/ day. The doses are expressed as median with interquartile range. [N = number of patients and n = number of patient visits where data was available for the analysis]