Appendix

The systematic use of metabolomic epidemiology, biobanks, and electronic medical records for precision medicine initiatives in asthma: new findings suggest new guidelines to optimize treatment

Authors:
Priyadarshini Kachroo Ph.D.[1*], Isobel D. Stewart Ph.D.[2*], Rachel S. Kelly Ph.D.[1], Meryl Stav M.S.[1], Kevin Mendez M.S.[1], Amber Dahlin Ph.D.[1], Djøra I. Soeteman Ph.D.[1,3], Su H. Chu Ph.D.[1], Mengna Huang Ph.D.[1], Margaret Cote[1], Hannah M. Knilhtilä M.D.[1], Kathleen Lee-Sarwar M.D.[1], Michael McGeachie Ph.D. [1], Alberta Wang M.D.[1],Ann Chen Wu M.D.[4], Yamini Virkud M.D.[5], Pei Zhang, Ph.D. [6,8], Nicholas J. Wareham, M.D.[2], Elizabeth W. Karlson M.D.[7], Craig E. Wheelock Ph.D.[6,8], Clary Clish Ph.D.[9], Scott T. Weiss M.D.[1], Claudia Langenberg, Ph.D.[2], Jessica A. Lasky-Su, Sc.D.[1]

[1] Channing Division of Network Medicine, Department of Medicine, Brigham and Women's Hospital and Harvard Medical School, Boston, MA, USA
[2] University of Cambridge, Cambridge, England
[3] Center for Health Decision Science, Harvard T.H. Chan School of Public Health, Boston, MA, USA
[4] Harvard Pilgrim Health Care Institute and Department of Population Medicine, Harvard Medical School, Boston, MA USA
[5] Massachusetts General Hospital and Harvard Medical School, Boston, MA USA
[6] Gunma University, Gunma, Japan
[7] Division of Rheumatology, Inflammation and Immunity, Brigham and Women's Hospital and Harvard Medical School, Boston, MA, USA
[8] Karolinska Institute, Stockholm, Sweden
[9] Broad Institute, Cambridge, MA USA

Table of Contents

Methods 3
COHORTS/STUDY POPULATIONS 3
Discovery Study Population: EPIC-Norfolk 3
Replication Study Population: Mass General Brigham Biobank-Asthma (MGBB-Asthma) 4
Metabolomic Profiling for EPIC-Norfolk and MGBB-Asthma 5
Randomized controlled trial (RCT) of ICS: Childhood Asthma Management Program (CAMP) 6
Clinical Quantification of Cortisol: MGBB-Cortisol 7
Statistical Analysis 8
Relative Quantification of Steroid Metabolites between Asthma (with and without ICS intake) and Controls in MGBB-Asthma 8

Results 8
Metabolite-ICS associations in MGBB-Asthma and CAMP 8

Supplementary Figures 11
Figure S1 11
Figure S2 13

Supplementary Tables 14

Table S1 14
Table S2 16
Table S3 17
Table S4 20
Table S5 22
Table S6 23
Table S7 24
Table S8 24

References 27

METHODS

COHORTS/STUDY POPULATIONS

Discovery Study Population: EPIC-Norfolk

The European Prospective Investigation of Cancer (EPIC)[1] is a large multi-center, longitudinal cohort study investigating the impact of nutrition and lifestyle factors on cancer risk in 519,978 men and women (aged 35-70 years) from 23 locations across 10 European countries. EPIC-Norfolk[1] includes 25,000 men and women, of predominantly European descent. Plasma samples were obtained at the baseline visit between 1993 and 1998 and were stored in the gas phase of liquid nitrogen at -175 degrees Celsius from this collection onward. At baseline, participants provided blood samples and completed a health and lifestyle questionnaire. Asthma status was ascertained based on a combination of self-report of previous diagnosis (where the question was: "Has the doctor ever told you that you have any of the following: Asthma"?), and any additional cases ascertained at a baseline health check performed by a registered nurse.

Plasma metabolomic data generated on a subset of 10,754 participants from the EPIC-Norfolk cohort were used as the discovery population in this analysis. Of these 10,754 participants, 661 (6%) individuals had a positive asthma status (as previously defined), while the remainder were considered non-asthmatic controls. Individuals with a self-reported previous doctor's diagnosis of bronchitis, and/or emphysema at baseline were excluded. Participants provided consent to participate in the study. Participants were not requested to fast prior to blood sampling and were largely unfasted. The study was approved by the Norwich Local Ethics Committee (REC Ref. 98CN01). **Table 1A** summarizes the characteristics of the EPIC-Norfolk participants in this study.

Replication Study Population: Mass General Brigham Biobank-Asthma (MGBB-Asthma)

The Mass General Brigham Biobank (MGBB) (https://biobank.partners.org) is a collection of DNA, serum, and plasma samples from 81,502 fully consented subjects recruited from the Partners Healthcare System, that is linked to the Research Patient Data Registry (RPDR), a data warehouse that gathers data from multiple electronic medical record (EMR) systems throughout Partners Healthcare and stores it in a SQL Server database. Researchers may query the RPDR using an online query tool; RPDR currently contains data on 4.6M patients, with 227M encounters, and approximately 900M distinct, coded clinical facts stored in the database dating back to 1986 including demographic data, diagnoses (e.g., ICD-9/ICD-10 codes), procedures (e.g., Current Procedural Terminology (CPT) codes), pharmacy data (*e.g.* RxNorm), inpatient and outpatient encounter information, provider information, laboratory data, imaging, and pathology data. We applied a validated phenotyping algorithm[2] for asthma diagnosis using RPDR. For our study, we identified 287 individuals with asthma (positive predictive value > 85%) and 323 controls (negative predictive value > 99%) generating the MGBB-Asthma population (total N = 610 subjects**, Table 1A**). Plasma samples for the MGBB asthma cohort were collected between October 2010 and March 2017 and were stored immediately in an -80 degree freezer. Non-fasted plasma samples were available on all of these individuals, which were used for metabolomic profiling. This study was approved by the IRB of Mass General Brigham and all study participants provided written consent at enrollment.

Using the MGBB, we also obtained information on asthma medication use (**Table S1).** Quantitative ICS intake was ascertained using presence and total prescription count of the following medications: Beclomethasone, Dipropionate, Budenoside, Ciclesonide,

Dexamethasone, Flunisolide, Fluticasone, Fluticasone/Salmeterol, Mometasone and Triamcinolone. To avoid misclassification, we further created stringent binary classifications for presence/absence of ICS medications. Based on the frequency/distribution of subjects with ICS intake, subjects with at least four ICS prescriptions were categorized as subjects with ICS intake. Subjects with less than four prescription counts of their ICS medication were categorized as without ICS intake including those without any ICS intake. Any healthy controls with intake of ICS and individuals with COPD were excluded from the analysis.

Metabolomic Profiling for EPIC-Norfolk and MGBB-Asthma

In both studies, metabolomic profiling was conducted by Metabolon Inc. (Durham, NC, USA). The methods have been described in detail previously[3]. In short, four non-targeted Liquid Chromatography Couple Mass Spectroscopy (LCMS) platforms were performed enabling the broadest coverage of the metabolome: 1) Amines and polar metabolites that ionize in the positive ion mode; 2) Central metabolites and polar metabolites that ionize in the negative ion mode; 3) Polar and non-polar lipids; 4) Free fatty acids, bile acids, and metabolites of intermediate polarity. All reagents and columns for this project were purchased in bulk from a single lot and all instruments were calibrated for mass resolution and mass accuracy daily. Coefficients of variation were measured in blinded QC samples randomly distributed among study samples, and batch variation is controlled for. Metabolites were identified by their mass-to-charge ratio (m/z), retention time (rt), and through a comparison to a library of purified known standards. Peaks were quantified using the area-under-the-(ROC) curve. Metabolite measures were median normalized across run days (with medians set to 1).

In EPIC-Norfolk, measurements were made in citrate plasma samples taken at baseline, for two sets of samples, each consisting of approximately 6,000 quasi-randomly selected individuals. Individuals with high levels of metabolite missingness were excluded. Metabolites present in at least 30 asthma cases in both measurement sets were included in the analyses. Metabolite measures were natural log transformed, windsorized at 5 SD and standardized ($\mu = 0$, SD = 1). Analyses were performed within each of the two metabolite measurements sets individually, and results were meta-analysed to pool the associations from the two measurement sets, using a fixed effects inverse variance weighted meta-analysis. In the MGBB-Asthma, missing metabolite values were imputed by replacement with half the minimum value for each metabolite in all samples. Metabolites with an interquartile range of zero were excluded from further analysis (n=129) with 904 metabolites remaining for the analysis (**Table S2**).

Randomized controlled trial (RCT) of ICS: Childhood Asthma Management Program (CAMP)

CAMP is a double blind RCT that randomized 1,041 children with mild-to-moderate asthma aged 5 to 12 years to inhaled steroid budesonide or nedocromil (each compared with placebo), treatment twice daily during 4.3 years of the trial[4,5]. The participants were managed further by their physician for a 4.8-year post-trial period. Approval was obtained from the institutional review boards at each of the participating institutions of CAMP. Samples for the CAMP clinical trial were obtained from the clinical sites at the randomization visit between December 1993 and September 1995 and again at the end of the clinical trial between December 1997 and December 1999. Samples were stored in an -80 degree freezer. Cortisone and cortisol were both among the metabolites measured via global untargeted metabolomic profiling at the Broad Institute using

560 serum samples **(Table 1B)** collected from CAMP individuals at baseline and four years later at the end of the trial. The quality control information is detailed elsewhere[6,7]. The RCT design of CAMP was used to further validate the cause-effect relationship between long-term ICS use and changes in cortisone and cortisol levels.

Clinical Quantification of Cortisol: MGBB-Cortisol

We queried the RPDR in MGBB to identify individuals who obtained cortisol testing (most often as a part of adrenal insufficiency testing, specifically the ACTH stim test) during the course of their recorded EMRs. Given the diurnal variation in cortisol levels, we obtained detailed information on specifications of the cortisol measurement; including the time and date of the blood draw. For individuals with multiple cortisol measurements on different dates, we recorded the minimum, maximum, mean, and median cortisol values, only considering the initial measurement from any single visit. To avoid confounding by other medications and to avoid misclassification due to controls that may have taken ICS medications for reasons other than asthma, we used a stringent measure of ICS intake: subjects having a count of greater than or equal to at least 10 prescriptions of their most common ICS medication were categorized as "ICS use", while subjects with less than 10 prescription counts of their most common ICS medication were categorized as "no ICS use". We then stratified these groups based on asthma affection, resulting in the following four categories: 1) no asthma/no ICS use; 2) no asthma/ICS use; 3) asthma/no ICS use; 4) asthma/ICS use. Subjects with COPD were excluded from this analysis. We identified a total of 2,235 individuals that were included in these analyses to generate the MGBB-Cortisol cohort (**Table 1B**).

Statistical Analysis

An overview of analytic approach and methods describing discovery and replication for associations between metabolites (independent predictor variable) and asthma affection status (dependent variable) is described in the main paper.

Relative Quantification of Steroid Metabolites between Asthma (with and without ICS intake) and Controls in MGBB-Asthma

To quantify the relative reduction in steroid metabolite levels based on ICS use, asthmatics were stratified by the Asthma Status/ICS variable used in the clinical quantification of cortisol. Multivariable logistic regression models were utilized to assess steroid metabolite levels between asthma cases and controls; asthma cases with ICS intake and controls; asthma cases with no ICS and controls; and, asthma cases with and without ICS intake. All models were adjusted for age, gender, race, and BMI.

RESULTS

Metabolite-ICS associations in MGBB-Asthma and CAMP

Seventeen of the 35 significant metabolites in EPIC-Norfolk cohort (**Figure S1A-B**, details in main paper) were replicated in MGBB-Asthma cohort at an FDR threshold of 5% that were annotated to major steroid hormone biosynthesis pathways (**Figure S2**). In MGBB-Asthma, we focused on the 34 steroid metabolites that were measured by Metabolon Inc. (Durham, NC, USA): 20 androgenic steroids, five pregnenolone steroids, two corticosteroids, and seven progestin steroids. All steroid metabolites except five progestin steroids were significantly lowered in asthmatics with ICS intake and with asthmatics overall compared with controls at an

FDR threshold of 5% (**Table S4, Figure 2B**). In asthmatics with ICS intake compared to asthmatics with no ICS, all steroid metabolites except two androgenic steroids and seven progestin steroids were significantly lowered (**Figure 2B**, **Table S4**). For asthmatics with no ICS compared to controls, none of the steroid metabolites were significant at an FDR threshold of 5%, however at a nominal P-value cut-off of 5%, nine androgenic steroids and two pregnenolone steroids were significantly lowered (**Figure 2B, Table S4**). Across all measured steroid metabolites, the median OR was 0.95 between asthmatics with no ICS and controls, which is 0.09 (9%) higher than between asthmatics with ICS intake and controls. Stratified by steroid pathway into androgenic, pregnenolone, corticosteroid, and progestin steroids, the median ORs between asthmatics with no ICS and controls were 0.95, 0.94, 0.96, 0.96, and between asthmatics with ICS intake and controls were 0.85, 0.84, 0.86, 0.90, respectively (**Figure 2B, Table S5-S8**). Across all four analyses, no steroid metabolite levels were significantly increased (OR > 1) even at a P-value cut-off of 5%.

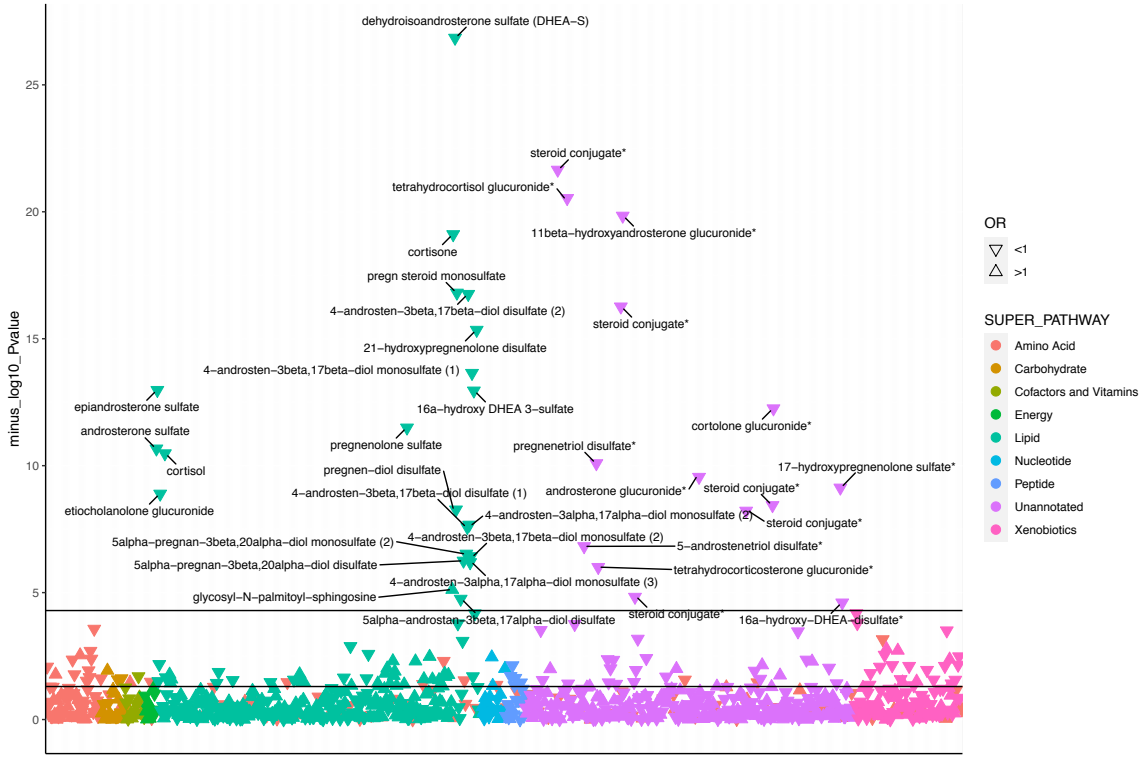
The overall reduction in steroid levels metabolites (median OR = 0.86) in asthmatics with ICS intake is consistent with adrenal suppression. This reduction was highly significant (FDR threshold of 5%) in all androgenic steroids, pregnenolone steroids, and corticosteroids, but only in 2/7 (28.6%) of progestin steroids. As classification of asthmatics with no ICS is based on less than four prescription counts of their ICS medication, there may be mild adrenal suppression associated with low ICS usage. Therefore, while no steroids were highly significant (FDR threshold of 5%), there was a suggestive decrease in 9/20 (45%) androgenic steroids and 2/5 (40%) pregnanolone steroids, but 0/2 (0%) of corticosteroids. This result suggests that a panel including androgenic steroids and pregnanolone steroid metabolites would be preferred over

cortisol measures to identify decreases in steroid levels prior to clinical-grade adrenal suppression.

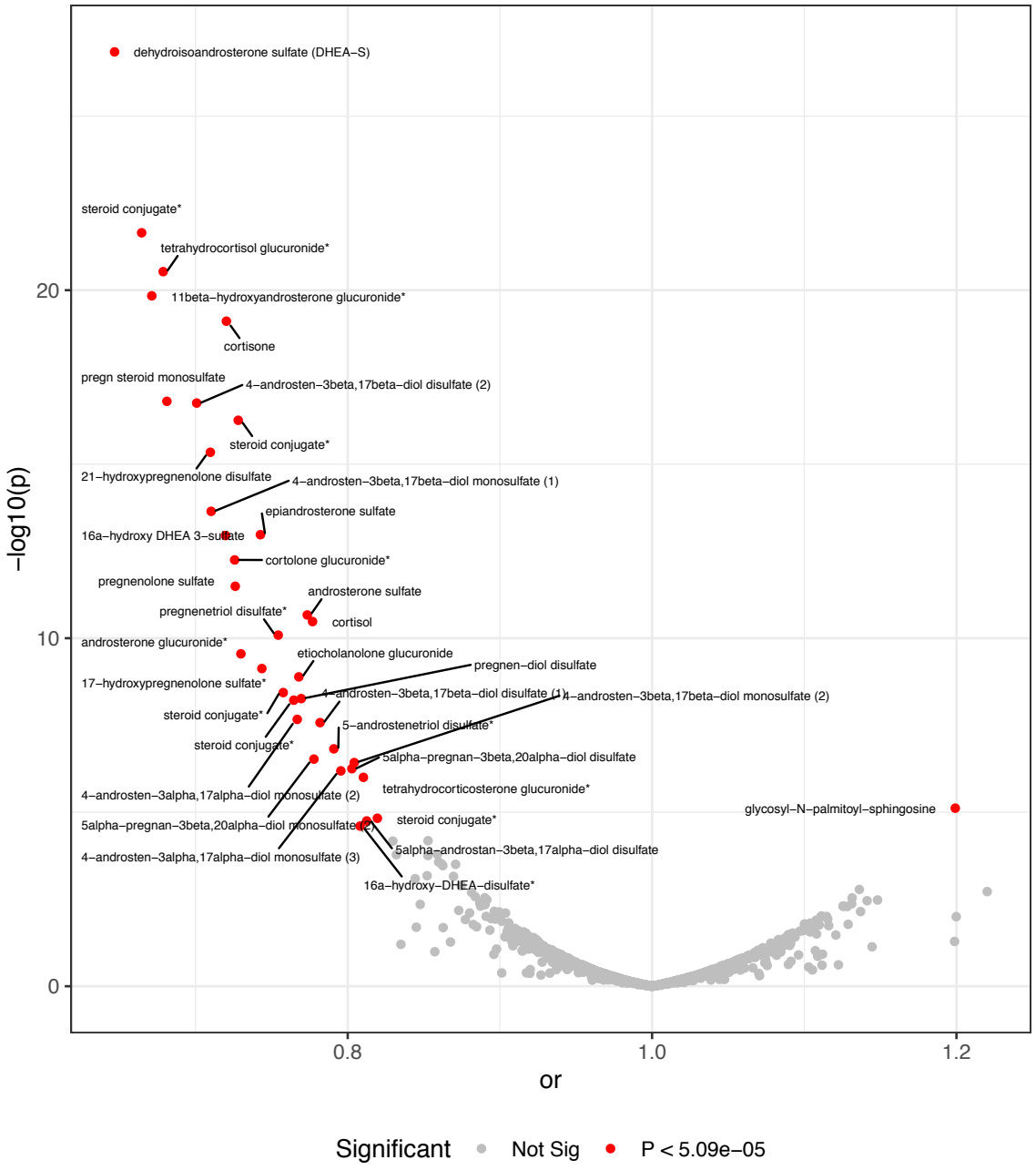
Supplementary Figures

Figure S1. Plasma metabolites in EPIC-Norfolk cohort. A.) Manhattan plot of metabolites sorted by their pathways on x-axis and negative log10 of P-value on the y-axis. The cut off horizontal lines on the y-axis highlight the metabolites significantly associated with asthma outcome at a P-value <0.05 and at the bonferroni threshold (n=35 metabolites, P-value<$5.14x10^{-5}$). The legend key shape and color shows the direction of effect for the metabolites and the main pathway they belong to, respectively. B.) Volcano Plot showing the effect size of the metabolites with OR on the x-axis and negative log10 of P-value on the y-axis. The metabolites colored in red are significant at a bonferroni threshold of P-value<$5.14x10^{-5}$.

A.

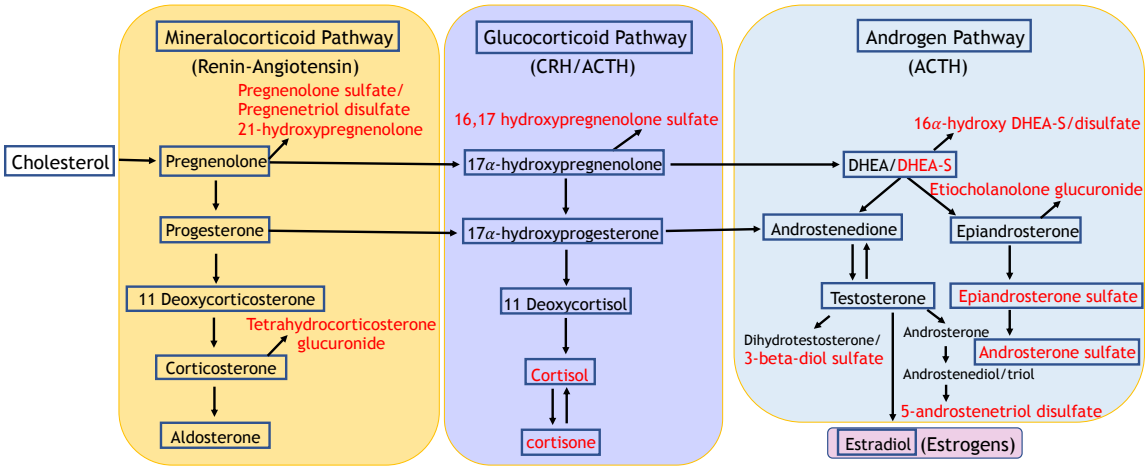


B.


dehydroisoandrosterone sulfate (DHEA−S)
steroid conjugate*
tetrahydrocortisol glucuronide*
11beta−hydroxyandrosterone glucuronide*
cortisone
pregn steroid monosulfate
4−androsten−3beta,17beta−diol disulfate (2)
steroid conjugate*
21−hydroxypregnenolone disulfate
4−androsten−3beta,17beta−diol monosulfate (1)
epiandrosterone sulfate
16a−hydroxy DHEA 3−sulfate
cortolone glucuronide*
pregnenolone sulfate
androsterone sulfate
pregnenetriol disulfate*
cortisol
androsterone glucuronide*
etiocholanolone glucuronide
pregnen−diol disulfate
17−hydroxypregnenolone sulfate*
4−androsten−3beta,17beta−diol disulfate (1)
4−androsten−3beta,17beta−diol monosulfate (2)
steroid conjugate*
5−androstenetriol disulfate*
steroid conjugate*
5alpha−pregnan−3beta,20alpha−diol disulfate
tetrahydrocorticosterone glucuronide*
4−androsten−3alpha,17alpha−diol monosulfate (2)
glycosyl−N−palmitoyl−sphingosine
5alpha−pregnan−3beta,20alpha−diol monosulfate (2)
steroid conjugate*
5alpha−androstan−3beta,17alpha−diol disulfate
4−androsten−3alpha,17alpha−diol monosulfate (3)
16a−hydroxy−DHEA−disulfate*
20
10
0
−log10(p)
0.8
1.0
1.2
or
Significant
Not Sig
P < 5.09e−05

237
238
239

Figure S2. Principal steroid hormone biosynthesis pathways with mineralocorticoid, glucocorticoid and androgen metabolites highlighting the replicated metabolites between EPIC-Norfolk cohort and Mass General Brigham Biobank. Our annotated metabolites colored in red have been mapped to these pathways with their precursors or intermediates.
Abbreviations: CRH: Corticotropin releasing hormone; ACTH: Adrenocorticotropic hormone



Supplementary Tables

Table S1. Inclusion and exclusion definitions and data dictionary for asthma and asthma medications from Mass General Brigham Biobank electronic medical health record (MGBB EMR) data. Since the data in biobank is dynamic, the data download was restricted till March 24, 2020 (the date at the time of last pull for final analysis)

Diagnosis/Medications	**Folder one level up in MGBB**	**International Classification of Diseases (ICD)-9 and ICD-10 codes where applicable/available**
Asthma - current or past history (custom PPV) [>= 0.80PPV]	Asthma (AST)	NA
Asthma - current or past history (PPV 0.90)	Asthma (AST)	NA
Asthma - no history (NPV 0.99)	Asthma (AST)	Absence of J45
Plasma	Biobank Sample Types	NA
Asthma	Chronic lower respiratory diseases (J40-J47)	J45
Chronic obstructive pulmonary disease with (acute) exacerbation	Other chronic obstructive pulmonary disease	J44.1
Prednisone	Glucocorticoids	RXNORM:8640
Methylprednisolone	Glucocorticoids	RXNORM:6902
Dexamethasone	Glucocorticoids	RXNORM:3264
Dexamethasone	Anti-inflammatories,inhalation	RXNORM:3264
Ipratropium	Bronchodilators,anticholinergic	RXNORM:7213
Tiotropium	Bronchodilators,anticholinergic	RXNORM:69120
Omalizumab	Immune Suppressants	RXNORM:302379
Beclomethasone dipropionate	Anti-inflammatories,inhalation	RXNORM:1348
Triamcinolone	Glucocorticoids	RXNORM:10759
Triamcinolone	Anti-inflammatories,inhalation	RXNORM:10759
Fluticasone	Anti-inflammatories,inhalation	RXNORM:41126
Budesonide	Glucocorticoids	RXNORM:19831

Budesonide	Anti-inflammatories,inhalation	RXNORM:19831
Mometasone	Anti-inflammatories,inhalation	RXNORM:108118
Ciclesonide	Anti-inflammatories,inhalation	RXNORM:274964
Flunisolide	Anti-inflammatories,inhalation	RXNORM:25120
Fluticasone/salmeterol	Antiasthma,other	RXNORM:284635
Budesonide/formoterol	Antiasthma,other	RXNORM:389132
Formoterol/mometasone	Antiasthma,other	RXNORM:1002293
Fluticasone/vilanterol	Antiasthma,other	RXNORM:1424888
Montelukast	Antiasthma,other	RXNORM:88249
Zafirlukast	Antiasthma,other	RXNORM:114970
Zileuton	Antiasthma,other	RXNORM:40575
Albuterol	Bronchodilators,sympathomimetic,inhalation	RXNORM:435
Albuterol	Bronchodilators,sympathomimetic,oral	RXNORM:435
Levalbuterol	Bronchodilators,sympathomimetic,inhalation	RXNORM:237159
Ipratropium/albuterol 3ml	Antiasthma,antileukotrienes	B00913606
Albuterol/ipratropium	Antiasthma,other	RXNORM:214199

Table S2. Distribution of metabolites in MGBB by platform and by super-pathway (n=904)

Platform	n	%
LC/MS Neg	508	56.2
LC/MS Polar	83	11.9
LC/MS Pos Early	242	2.7
LC/MS Pos Late	71	19.7

Super-pathway	n	%
Lipid	184	20.4
Xenobiotics	108	11.9
Carbohydrate	24	2.7
Amino Acid	178	19.7
Cofactors and Vitamins	29	3.2
Nucleotide	31	3.4
Energy	9	1
Peptide	29	3.2
Partially Characterized molecules	3	0.3
Unannotated metabolites	309	34.2

Table S3. Plasma metabolites significantly associated with prevalent asthma outcome (n=35, FDR < 5.14×10^{-5}) in discovery EPIC-Norfolk cohort

Metabolite	Names from metabolon*	HMDB	N	Ncase	se	p	or	or_95l	or_95h
dehydroisoandrosterone sulfate (DHEA-S)		HMDB0001032	10,751	661	0.04	1.42E-27	0.65	0.60	0.70
X - 21364	possibly a steroid conjugate		10,665	642	0.04	2.24E-22	0.66	0.61	0.72
X - 11444	tetrahydrocortisol glucuronide		10,717	648	0.04	2.93E-21	0.68	0.63	0.74
X - 12846	retired for 11beta-hydroxyandrosterone glucuronide		10,342	607	0.04	1.45E-20	0.67	0.62	0.73
cortisone		HMDB0002802	10,712	649	0.04	7.77E-20	0.72	0.67	0.77
Pregn steroid monosulfate			10,751	660	0.05	1.56E-17	0.68	0.62	0.74
4-androsten-3beta,17beta-diol disulfate (2)		HMDB0003818	10,739	658	0.04	1.75E-17	0.70	0.65	0.76
X - 12844	possibly a steroid conjugate		10,744	656	0.04	5.47E-17	0.73	0.68	0.78
21-hydroxypregnenolone disulfate		HMDB0004026	10,655	642	0.04	4.54E-16	0.71	0.65	0.77
4-androsten-3beta,17beta-diol monosulfate (1)		HMDB0003818	10,711	653	0.04	2.26E-14	0.71	0.65	0.78
epiandrosterone sulfate		HMDB0000365	10,720	656	0.04	1.06E-13	0.74	0.69	0.80
16a-hydroxy DHEA 3-sulfate			10,696	647	0.04	1.12E-13	0.72	0.66	0.79
X - 17359	retired for cortolone glucuronide		10,382	615	0.04	5.63E-13	0.73	0.67	0.79
pregnenolone sulfate		HMDB0000774	10,187	593	0.05	3.21E-12	0.73	0.66	0.79
androsterone sulfate		HMDB0002759	10,741	661	0.04	2.15E-11	0.77	0.72	0.83

cortisol		HMDB0000063	10,738	657	0.04	3.32E-11	0.78	0.72	0.84
X - 11440	retired for pregnenetriol disulfate		10,738	661	0.04	8.15E-11	0.75	0.69	0.82
X - 22379	retired for androsterone glucuronide		10,524	617	0.05	2.81E-10	0.73	0.66	0.80
X - 24544	17-hydroxypregnenolone sulfate	HMDB0000416	9,580	547	0.05	7.44E-10	0.74	0.68	0.82
etiocholanolone glucuronide		HMDB0004484	10,468	623	0.04	1.29E-09	0.77	0.71	0.84
X - 17357	possibly a steroid conjugate		8,923	502	0.05	3.65E-09	0.76	0.69	0.83
pregnen-diol disulfate			10,754	661	0.04	5.43E-09	0.77	0.70	0.84
X - 17340	possibly a steroid conjugate		9,992	593	0.05	6.04E-09	0.76	0.70	0.84
4-androsten-3alpha,17alpha-diol monosulfate (2)			9,775	563	0.05	2.15E-08	0.77	0.70	0.84
4-androsten-3beta,17beta-diol disulfate (1)		HMDB0003818	10,750	660	0.04	2.66E-08	0.78	0.72	0.85
X - 21470	5-androstenetriol disulfate		10,099	581	0.04	1.51E-07	0.79	0.72	0.86
5alpha-pregnan-3beta,20alpha-diol monosulfate (2)			9,551	547	0.05	2.98E-07	0.78	0.71	0.86
4-androsten-3beta,17beta-diol monosulfate (2)			10,462	628	0.04	3.75E-07	0.80	0.74	0.87
5alpha-pregnan-3beta,20alpha-diol disulfate			10,257	611	0.04	5.61E-07	0.80	0.74	0.87
4-androsten-3alpha,17alpha-diol monosulfate (3)			10,675	647	0.05	6.48E-07	0.80	0.73	0.87

X - 11470	tetrahydrocorticosterone glucuronide		10,069	589	0.04	9.95E-07	0.81	0.74	0.88
glycosyl-N-palmitoyl-sphingosine			10,742	659	0.04	7.64E-06	1.20	1.11	1.30
X - 15492	possibly a steroid conjugate		10,612	635	0.05	1.49E-05	0.82	0.75	0.90
5alpha-androstan-3beta,17alpha-diol disulfate			8,839	505	0.05	1.78E-05	0.81	0.74	0.89
X - 24546	16a-hydroxy-DHEA-disulfate		8,880	500	0.05	2.50E-05	0.81	0.73	0.89

263
264

265 **Table S4.** Steroid metabolite associations based on logistic regression models (adjusted for confounders) between asthma cases
266 and controls; asthma cases with ICS intake and controls; asthma cases with no ICS and controls; and asthma cases with ICS intake
267 and with no ICS in MGBB-Asthma
268

Metabolite	Metabolite Subclass	Asthma vs. Control			Asthma ICS vs. Control			Asthma NO ICS vs. Control			Asthma ICS vs. Asthma NO ICS		
		OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR
androstenediol (3beta,17beta) monosulfate (1)	Androgenic Steroids	0.84 (0.79, 0.88)	$5.5x10^{-11}$	$1.9x10^{-9}$	0.81 (0.77, 0.86)	$1.8x10^{-14}$	$6.0x10^{-13}$	0.92 (0.86, 0.98)	0.006	0.07	0.90 (0.83, 0.97)	0.005	0.01
androstenediol (3beta,17beta) disulfate (2)	Androgenic Steroids	0.84 (0.79, 0.89)	$4.6x10^{-9}$	$3.2x10^{-8}$	0.81 (0.77, 0.86)	$3.2x10^{-12}$	$2.0x10^{-11}$	0.92 (0.85, 0.99)	0.03	0.13	0.90 (0.83, 0.97)	0.007	0.02
androstenediol (3beta,17beta) disulfate (1)	Androgenic Steroids	0.84 (0.80, 0.89)	$8.0x10^{-9}$	$4.5x10^{-8}$	0.81 (0.77, 0.86)	$6.4x10^{-13}$	$7.3x10^{-12}$	0.93 (0.87, 1.00)	0.06	0.15	0.89 (0.82, 0.96)	0.005	0.01
androstenediol (3alpha, 17alpha) monosulfate (3)	Androgenic Steroids	0.84 (0.80, 0.90)	$1.8x10^{-8}$	$7.6x10^{-8}$	0.82 (0.78, 0.87)	$3.0x10^{-11}$	$1.3x10^{-10}$	0.93 (0.86, 0.99)	0.03	0.13	0.90 (0.83, 0.98)	0.02	0.03
androstenediol (3beta,17beta) monosulfate (2)	Androgenic Steroids	0.85 (0.80, 0.89)	$4.5x10^{-10}$	$7.6x10^{-9}$	0.84 (0.80, 0.88)	$2.0x10^{-11}$	$9.7x10^{-11}$	0.91 (0.86, 0.97)	0.003	0.07	0.91 (0.85, 0.98)	0.02	0.03
5alpha-androstan-3beta,17beta-diol disulfate	Androgenic Steroids	0.85 (0.80, 0.90)	$1.3x10^{-8}$	$6.3x10^{-8}$	0.83 (0.79, 0.88)	$1.0x10^{-10}$	$3.5x10^{-10}$	0.92 (0.86, 0.98)	0.02	0.13	0.91 (0.85, 0.99)	0.03	0.04
epiandrosterone sulfate	Androgenic Steroids	0.85 (0.81, 0.90)	$4.7x10^{-9}$	$3.2x10^{-8}$	0.83 (0.79, 0.87)	$3.0x10^{-12}$	$2.0x10^{-11}$	0.93 (0.87, 1.00)	0.04	0.13	0.91 (0.85, 0.98)	0.01	0.03
dehydroisoandrosterone sulfate (DHEA-S)	Androgenic Steroids	0.86 (0.81, 0.90)	$3.6x10^{-8}$	$1.2x10^{-7}$	0.83 (0.79, 0.87)	$3.6x10^{-12}$	$2.0x10^{-11}$	0.95 (0.89, 1.02)	0.16	0.24	0.89 (0.82, 0.96)	0.003	0.01
androsterone sulfate	Androgenic Steroids	0.86 (0.81, 0.91)	$1.0x10^{-7}$	$3.2x10^{-7}$	0.84 (0.80, 0.89)	$3.9x10^{-10}$	$1.1x10^{-9}$	0.93 (0.87, 1.00)	0.04	0.14	0.93 (0.86, 1.00)	0.06	0.07
androstenediol (3alpha, 17alpha) monosulfate (2)	Androgenic Steroids	0.87 (0.83, 0.92)	$4.2x10^{-7}$	$1.1x10^{-6}$	0.84 (0.80, 0.89)	$1.3x10^{-10}$	$3.9x10^{-10}$	0.95 (0.89, 1.02)	0.13	0.24	0.90 (0.84, 0.96)	0.004	0.01
5alpha-androstan-3beta,17beta-diol monosulfate (2)	Androgenic Steroids	0.88 (0.84, 0.93)	$6.0x10^{-7}$	$1.5x10^{-6}$	0.87 (0.83, 0.91)	$3.8x10^{-8}$	$7.1x10^{-8}$	0.94 (0.89, 0.99)	0.03	0.13	0.92 (0.85, 0.99)	0.03	0.05
androsterone glucuronide	Androgenic Steroids	0.88 (0.84, 0.93)	$4.0x10^{-6}$	$7.2x10^{-6}$	0.86 (0.82, 0.91)	$2.3x10^{-8}$	$4.9x10^{-8}$	0.95 (0.90, 1.02)	0.15	0.24	0.90 (0.84, 0.97)	0.005	0.01
etiocholanolone glucuronide	Androgenic Steroids	0.88 (0.84, 0.93)	$3.5x10^{-7}$	$9.8x10^{-7}$	0.86 (0.82, 0.90)	$4.3x10^{-10}$	$1.1x10^{-9}$	0.95 (0.90, 1.01)	0.10	0.22	0.90 (0.84, 0.96)	0.002	0.01
5alpha-androstan-3alpha,17beta-diol monosulfate (1)	Androgenic Steroids	0.89 (0.85, 0.93)	$9.0x10^{-7}$	$2.1x10^{-6}$	0.89 (0.85, 0.93)	$4.5x10^{-7}$	$7.7x10^{-7}$	0.93 (0.88, 0.98)	0.006	0.07	0.97 (0.90, 1.04)	0.35	0.41
5alpha-androstan-3beta,17alpha-diol disulfate	Androgenic Steroids	0.90 (0.86, 0.94)	$1.8x10^{-6}$	$3.4x10^{-6}$	0.89 (0.85, 0.92)	$3.5x10^{-8}$	$7.0x10^{-8}$	0.96 (0.92, 1.01)	0.12	0.23	0.91 (0.85, 0.98)	0.009	0.02
5alpha-androstan-3alpha,17alpha-diol monosulfate	Androgenic Steroids	0.90 (0.86, 0.95)	$1.3x10^{-4}$	$1.9x10^{-4}$	0.88 (0.84, 0.93)	$1.8x10^{-6}$	$2.7x10^{-6}$	0.98 (0.92, 1.03)	0.39	0.46	0.90 (0.82, 0.97)	0.01	0.02
5alpha-androstan-3alpha,17beta-diol disulfate	Androgenic Steroids	0.91 (0.87, 0.95)	$1.2x10^{-4}$	$1.9x10^{-4}$	0.89 (0.85, 0.93)	$1.2x10^{-6}$	$1.8x10^{-6}$	0.97 (0.92,1.02)	0.30	0.39	0.90 (0.84, 0.97)	0.008	0.02
16a-hydroxy DHEA 3-sulfate	Androgenic Steroids	0.92 (0.87, 0.97)	0.003	0.005	0.89 (0.84, 0.94)	$8.8x10^{-6}$	$1.3x10^{-5}$	1.01 (0.95, 1.07)	0.76	0.78	0.89 (0.82, 0.96)	0.002	0.01
5alpha-androstan-3alpha,17beta-diol monosulfate (2)	Androgenic Steroids	0.93 (0.88, 1.00)	0.04	0.05	0.91 (0.86, 0.97)	0.007	0.008	0.98 (0.92, 1.05)	0.64	0.68	0.87 (0.77, 0.98)	0.03	0.04

andro steroid monosulfate C19H28O6S (1)	Androgenic Steroids	0.94 (0.89, 0.98)	0.006	0.008	0.91 (0.87, 0.95)	$7.1x10^{-5}$	$9.2x10^{-5}$	1.00 (0.95, 1.06)	0.88	0.88	0.89 (0.82, 0.95)	$8.0x10^{-4}$	0.01
pregnenediol sulfate (C21H34O5S)	Pregnenolone Steroids	0.84 (0.79, 0.89)	$2.2x10^{-8}$	$8.2x10^{-8}$	0.82 (0.77, 0.87)	$3.9x10^{-11}$	$1.5x10^{-10}$	0.91 (0.85, 0.99)	0.02	0.13	0.89 (0.82, 0.97)	0.007	0.02
pregnenolone sulfate	Pregnenolone Steroids	0.85 (0.81, 0.90)	$2.6x10^{-9}$	$3.0x10^{-8}$	0.82 (0.78, 0.86)	$1.4x10^{-13}$	$2.4x10^{-12}$	0.93 (0.87, 0.99)	0.02	0.13	0.90 (0.83, 0.97)	0.004	0.01
pregnenediol disulfate (C21H34O8S2)	Pregnenolone Steroids	0.86 (0.81, 0.91)	$1.0x10^{-6}$	$2.1x10^{-6}$	0.84 (0.79, 0.89)	$3.4x10^{-9}$	$8.3x10^{-9}$	0.95 (0.88, 1.02)	0.13	0.24	0.88 (0.81, 0.96)	0.003	0.01
21-hydroxypregnenolone disulfate	Pregnenolone Steroids	0.87 (0.83, 0.92)	$1.1x10^{-6}$	$2.2x10^{-6}$	0.85 (0.81, 0.90)	$4.0x10^{-9}$	$9.0x10^{-9}$	0.94 (0.88, 1.01)	0.09	0.21	0.90 (0.84, 0.97)	0.007	0.02
17alpha-hydroxypregnenolone 3-sulfate	Pregnenolone Steroids	0.92 (0.87, 0.97)	$1.4x10^{-3}$	$2.0x10^{-3}$	0.89 (0.84, 0.94)	$2.2x10^{-5}$	$3.0x10^{-5}$	0.98 (0.93, 1.04)	0.48	0.54	0.86 (0.78, 0.94)	0.001	0.01
cortisone	Corticosteroids	0.89 (0.83, 0.94)	$5.8x10^{-5}$	$9.8x10^{-5}$	0.86 (0.81, 0.91)	$8.3x10^{-8}$	$1.5x10^{-7}$	0.95 (0.87, 1.04)	0.28	0.39	0.90 (0.83, 0.97)	0.005	0.01
cortisol	Corticosteroids	0.89 (0.84, 0.95)	$4.0x10^{-4}$	$6.0x10^{-4}$	0.86 (0.81, 0.91)	$5.9x10^{-7}$	$9.5x10^{-7}$	0.97 (0.90, 1.06)	0.53	0.59	0.89 (0.82, 0.96)	0.005	0.01
pregnanediol-3-glucuronide	Progestin Steroids	0.95 (0.90, 0.99)	0.02	0.03	0.94 (0.90, 0.99)	0.02	0.02	0.97 (0.91, 1.03)	0.27	0.38	0.94 (0.88, 1.00)	0.07	0.08
5alpha-pregnan-3beta,20beta-diol monosulfate (1)	Progestin Steroids	0.95 (0.90, 0.99)	0.02	0.03	0.95 (0.91, 1.00)	0.04	0.05	0.96 (0.90, 1.01)	0.14	0.24	0.97 (0.90, 1.03)	0.29	0.36
5alpha-pregnan-diol disulfate	Progestin Steroids	0.96 (0.92, 1.00)	0.06	0.07	0.98 (0.94, 1.02)	0.22	0.25	0.96 (0.92, 1.00)	0.07	0.17	0.99 (0.94, 1.05)	0.79	0.82
5alpha-pregnan-3beta,20alpha-diol disulfate	Progestin Steroids	0.96 (0.92, 1.01)	0.14	0.15	0.97 (0.93, 1.02)	0.21	0.23	0.97 (0.92, 1.03)	0.33	0.40	0.98 (0.92, 1.05)	0.54	0.57
pregnanolone/allopregnanolone sulfate	Progestin Steroids	0.97 (0.92, 1.01)	0.13	0.15	0.98 (0.94, 1.03)	0.44	0.44	0.95 (0.90, 1.00)	0.06	0.15	1.01 (0.94, 1.07)	0.86	0.86
5alpha-pregnan-3beta,20alpha-diol monosulfate (2)	Progestin Steroids	0.97 (0.93, 1.02)	0.19	0.19	0.97 (0.93, 1.02)	0.25	0.27	0.97 (0.92, 1.03)	0.32	0.40	0.98 (0.92, 1.04)	0.50	0.55
5alpha-pregnan-3beta-ol,20-one sulfate	Progestin Steroids	1.00 (0.95, 1.06)	0.93	0.93	1.03 (0.98, 1.08)	0.33	0.34	0.96 (0.91, 1.02)	0.21	0.32	1.03 (0.96, 1.11)	0.44	0.50

Abbreviations: ICS, Inhaled Corticosteroid Use

Table S5: Steroid metabolite associations (stratified by sub-pathways - Corticosteroids) based on logistic regression models (adjusted for confounders) between asthma cases and controls; asthma cases with ICS intake and controls; asthma cases with no ICS and controls; and asthma cases with ICS intake and with no ICS in MGBB-Asthma

Metabolite	Metabolite Subclass	Asthma vs. Control			Asthma ICS vs. Control			Asthma NO ICS vs. Control			Asthma ICS vs. Asthma NO ICS		
		OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR
cortisone	Corticosteroids	0.89 (0.83, 0.94)	5.8×10^{-5}	9.8×10^{-5}	0.86 (0.81, 0.91)	8.3×10^{-8}	1.5×10^{-7}	0.95 (0.87, 1.04)	0.28	0.39	0.90 (0.83, 0.97)	0.005	0.01
cortisol	Corticosteroids	0.89 (0.84, 0.95)	4.0×10^{-4}	6.0×10^{-4}	0.86 (0.81, 0.91)	5.9×10^{-7}	9.5×10^{-7}	0.97 (0.90, 1.06)	0.53	0.59	0.89 (0.82, 0.96)	0.005	0.01

Table S6: Steroid metabolite associations (stratified by sub-pathways - Pregnenolone Steroids) based on logistic regression models (adjusted for confounders) between asthma cases and controls; asthma cases with ICS intake and controls; asthma cases with no ICS and controls; and asthma cases with ICS intake and with no ICS in MGBB-Asthma

Metabolite	Metabolite Subclass	Asthma vs. Control			Asthma ICS vs. Control			Asthma NO ICS vs. Control			Asthma ICS vs. Asthma NO ICS		
		OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR
pregnenediol sulfate (C21H34O5S)	Pregnenolone Steroids	0.84 (0.79, 0.89)	$2.2x10^{-8}$	$8.2x10^{-8}$	0.82 (0.77, 0.87)	$3.9x10^{-11}$	$1.5x10^{-10}$	0.91 (0.85, 0.99)	0.02	0.13	0.89 (0.82, 0.97)	0.007	0.02
pregnenolone sulfate	Pregnenolone Steroids	0.85 (0.81, 0.90)	$2.6x10^{-9}$	$3.0x10^{-8}$	0.82 (0.78, 0.86)	$1.4x10^{-13}$	$2.4x10^{-12}$	0.93 (0.87, 0.99)	0.02	0.13	0.90 (0.83, 0.97)	0.004	0.01
pregnenediol disulfate (C21H34O8S2)	Pregnenolone Steroids	0.86 (0.81, 0.91)	$1.0x10^{-6}$	$2.1x10^{-6}$	0.84 (0.79, 0.89)	$3.4x10^{-9}$	$8.3x10^{-9}$	0.95 (0.88, 1.02)	0.13	0.24	0.88 (0.81, 0.96)	0.003	0.01
21-hydroxypregnenolone disulfate	Pregnenolone Steroids	0.87 (0.83, 0.92)	$1.1x10^{-6}$	$2.2x10^{-6}$	0.85 (0.81, 0.90)	$4.0x10^{-9}$	$9.0x10^{-9}$	0.94 (0.88, 1.01)	0.09	0.21	0.90 (0.84, 0.97)	0.007	0.02
17alpha-hydroxypregnenolone 3-sulfate	Pregnenolone Steroids	0.92 (0.87, 0.97)	$1.4x10^{-3}$	$2.0x10^{-3}$	0.89 (0.84, 0.94)	$2.2x10^{-5}$	$3.0x10^{-5}$	0.98 (0.93, 1.04)	0.48	0.54	0.86 (0.78, 0.94)	0.001	0.01

Table S7: Steroid metabolite associations (stratified by sub-pathways - Progestin Steroids) based on logistic regression models (adjusted for confounders) between asthma cases and controls; asthma cases with ICS intake and controls; asthma cases with no ICS and controls; and asthma cases with ICS intake and with no ICS in MGBB-Asthma

Metabolite	Metabolite Subclass	Asthma vs. Control			Asthma ICS vs. Control			Asthma NO ICS vs. Control			Asthma ICS vs. Asthma NO ICS		
		OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR
pregnanediol-3-glucuronide	Progestin Steroids	0.95 (0.90, 0.99)	0.02	0.03	0.94 (0.90, 0.99)	0.02	0.02	0.97 (0.91, 1.03)	0.27	0.38	0.94 (0.88, 1.00)	0.07	0.08
5alpha-pregnan-3beta,20beta-diol monosulfate (1)	Progestin Steroids	0.95 (0.90, 0.99)	0.02	0.03	0.95 (0.91, 1.00)	0.04	0.05	0.96 (0.90, 1.01)	0.14	0.24	0.97 (0.90, 1.03)	0.29	0.36
5alpha-pregnan-diol disulfate	Progestin Steroids	0.96 (0.92, 1.00)	0.06	0.07	0.98 (0.94, 1.02)	0.22	0.25	0.96 (0.92, 1.00)	0.07	0.17	0.99 (0.94, 1.05)	0.79	0.82
5alpha-pregnan-3beta,20alpha-diol disulfate	Progestin Steroids	0.96 (0.92, 1.01)	0.14	0.15	0.97 (0.93, 1.02)	0.21	0.23	0.97 (0.92, 1.03)	0.33	0.40	0.98 (0.92, 1.05)	0.54	0.57
pregnanolone/allopregnanolone sulfate	Progestin Steroids	0.97 (0.92, 1.01)	0.13	0.15	0.98 (0.94, 1.03)	0.44	0.44	0.95 (0.90, 1.00)	0.06	0.15	1.01 (0.94, 1.07)	0.86	0.86
5alpha-pregnan-3beta,20alpha-diol monosulfate (2)	Progestin Steroids	0.97 (0.93, 1.02)	0.19	0.19	0.97 (0.93, 1.02)	0.25	0.27	0.97 (0.92, 1.03)	0.32	0.40	0.98 (0.92, 1.04)	0.50	0.55
5alpha-pregnan-3beta-ol,20-one sulfate	Progestin Steroids	1.00 (0.95, 1.06)	0.93	0.93	1.03 (0.98, 1.08)	0.33	0.34	0.96 (0.91, 1.02)	0.21	0.32	1.03 (0.96, 1.11)	0.44	0.50

Table S8: Steroid metabolite associations (stratified by sub-pathways - Androgenic Steroids) based on logistic regression models (adjusted for confounders) between asthma cases and controls; asthma cases with ICS intake and controls; asthma cases with no ICS and controls; and asthma cases with ICS intake and with no ICS in MGBB-Asthma

Metabolite	Metabolite Subclass	Asthma vs. Control			Asthma ICS vs. Control			Asthma NO ICS vs. Control			Asthma ICS vs. Asthma NO ICS		
		OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR	OR (95% CI)	P-Value	FDR
androstenediol (3beta,17beta) monosulfate (1)	Androgenic Steroids	0.84 (0.79, 0.88)	$5.5x10^{-11}$	$1.9x10^{-9}$	0.81 (0.77, 0.86)	$1.8x10^{-14}$	$6.0x10^{-13}$	0.92 (0.86, 0.98)	0.006	0.07	0.90 (0.83, 0.97)	0.005	0.01
androstenediol (3beta,17beta) disulfate (2)	Androgenic Steroids	0.84 (0.79, 0.89)	$4.6x10^{-9}$	$3.2x10^{-8}$	0.81 (0.77, 0.86)	$3.2x10^{-12}$	$2.0x10^{-11}$	0.92 (0.85, 0.99)	0.03	0.13	0.90 (0.83, 0.97)	0.007	0.02
androstenediol (3beta,17beta) disulfate (1)	Androgenic Steroids	0.84 (0.80, 0.89)	$8.0x10^{-9}$	$4.5x10^{-8}$	0.81 (0.77, 0.86)	$6.4x10^{-13}$	$7.3x10^{-12}$	0.93 (0.87, 1.00)	0.06	0.15	0.89 (0.82, 0.96)	0.005	0.01
androstenediol (3alpha, 17alpha) monosulfate (3)	Androgenic Steroids	0.84 (0.80, 0.90)	$1.8x10^{-8}$	$7.6x10^{-8}$	0.82 (0.78, 0.87)	$3.0x10^{-11}$	$1.3x10^{-10}$	0.93 (0.86, 0.99)	0.03	0.13	0.90 (0.83, 0.98)	0.02	0.03
androstenediol (3beta,17beta) monosulfate (2)	Androgenic Steroids	0.85 (0.80, 0.89)	$4.5x10^{-10}$	$7.6x10^{-9}$	0.84 (0.80, 0.88)	$2.0x10^{-11}$	$9.7x10^{-11}$	0.91 (0.86, 0.97)	0.003	0.07	0.91 (0.85, 0.98)	0.02	0.03
5alpha-androstan-3beta,17beta-diol disulfate	Androgenic Steroids	0.85 (0.80, 0.90)	$1.3x10^{-8}$	$6.3x10^{-8}$	0.83 (0.79, 0.88)	$1.0x10^{-10}$	$3.5x10^{-10}$	0.92 (0.86, 0.98)	0.02	0.13	0.91 (0.85, 0.99)	0.03	0.04
epiandrosterone sulfate	Androgenic Steroids	0.85 (0.81, 0.90)	$4.7x10^{-9}$	$3.2x10^{-8}$	0.83 (0.79, 0.87)	$3.0x10^{-12}$	$2.0x10^{-11}$	0.93 (0.87, 1.00)	0.04	0.13	0.91 (0.85, 0.98)	0.01	0.03
dehydroisoandrosterone sulfate (DHEA-S)	Androgenic Steroids	0.86 (0.81, 0.90)	$3.6x10^{-8}$	$1.2x10^{-7}$	0.83 (0.79, 0.87)	$3.6x10^{-12}$	$2.0x10^{-11}$	0.95 (0.89, 1.02)	0.16	0.24	0.89 (0.82, 0.96)	0.003	0.01
androsterone sulfate	Androgenic Steroids	0.86 (0.81, 0.91)	$1.0x10^{-7}$	$3.2x10^{-7}$	0.84 (0.80, 0.89)	$3.9x10^{-10}$	$1.1x10^{-9}$	0.93 (0.87, 1.00)	0.04	0.14	0.93 (0.86, 1.00)	0.06	0.07
androstenediol (3alpha, 17alpha) monosulfate (2)	Androgenic Steroids	0.87 (0.83, 0.92)	$4.2x10^{-7}$	$1.1x10^{-6}$	0.84 (0.80, 0.89)	$1.3x10^{-10}$	$3.9x10^{-10}$	0.95 (0.89, 1.02)	0.13	0.24	0.90 (0.84, 0.96)	0.004	0.01
5alpha-androstan-3beta,17beta-diol monosulfate (2)	Androgenic Steroids	0.88 (0.84, 0.93)	$6.0x10^{-7}$	$1.5x10^{-6}$	0.87 (0.83, 0.91)	$3.8x10^{-8}$	$7.1x10^{-8}$	0.94 (0.89, 0.99)	0.03	0.13	0.92 (0.85, 0.99)	0.03	0.05
androsterone glucuronide	Androgenic Steroids	0.88 (0.84, 0.93)	$4.0x10^{-6}$	$7.2x10^{-6}$	0.86 (0.82, 0.91)	$2.3x10^{-8}$	$4.9x10^{-8}$	0.95 (0.90, 1.02)	0.15	0.24	0.90 (0.84, 0.97)	0.005	0.01
etiocholanolone glucuronide	Androgenic Steroids	0.88 (0.84, 0.93)	$3.5x10^{-7}$	$9.8x10^{-7}$	0.86 (0.82, 0.90)	$4.3x10^{-10}$	$1.1x10^{-9}$	0.95 (0.90, 1.01)	0.10	0.22	0.90 (0.84, 0.96)	0.002	0.01
5alpha-androstan-3alpha,17beta-diol monosulfate (1)	Androgenic Steroids	0.89 (0.85, 0.93)	$9.0x10^{-7}$	$2.1x10^{-6}$	0.89 (0.85, 0.93)	$4.5x10^{-7}$	$7.7x10^{-7}$	0.93 (0.88, 0.98)	0.006	0.07	0.97 (0.90, 1.04)	0.35	0.41
5alpha-androstan-3beta,17alpha-diol disulfate	Androgenic Steroids	0.90 (0.86, 0.94)	$1.8x10^{-6}$	$3.4x10^{-6}$	0.89 (0.85, 0.92)	$3.5x10^{-8}$	$7.0x10^{-8}$	0.96 (0.92, 1.01)	0.12	0.23	0.91 (0.85, 0.98)	0.009	0.02
5alpha-androstan-3alpha,17alpha-diol monosulfate	Androgenic Steroids	0.90 (0.86, 0.95)	$1.3x10^{-4}$	$1.9x10^{-4}$	0.88 (0.84, 0.93)	$1.8x10^{-6}$	$2.7x10^{-6}$	0.98 (0.92, 1.03)	0.39	0.46	0.90 (0.82, 0.97)	0.01	0.02
5alpha-androstan-3alpha,17beta-diol disulfate	Androgenic Steroids	0.91 (0.87, 0.95)	$1.2x10^{-4}$	$1.9x10^{-4}$	0.89 (0.85, 0.93)	$1.2x10^{-6}$	$1.8x10^{-6}$	0.97 (0.92,1.02)	0.30	0.39	0.90 (0.84, 0.97)	0.008	0.02
16a-hydroxy DHEA 3-sulfate	Androgenic Steroids	0.92 (0.87, 0.97)	0.003	0.005	0.89 (0.84, 0.94)	$8.8x10^{-6}$	$1.3x10^{-5}$	1.01 (0.95, 1.07)	0.76	0.78	0.89 (0.82, 0.96)	0.002	0.01
5alpha-androstan-3alpha,17beta-diol monosulfate (2)	Androgenic Steroids	0.93 (0.88, 1.00)	0.04	0.05	0.91 (0.86, 0.97)	0.007	0.008	0.98 (0.92, 1.05)	0.64	0.68	0.87 (0.77, 0.98)	0.03	0.04

andro steroid monosulfate C19H28O6S (1)	Androgenic Steroids	0.94 (0.89, 0.98)	0.006	0.008	0.91 (0.87, 0.95)	7.1×10^{-5}	9.2×10^{-5}	1.00 (0.95, 1.06)	0.88	0.88	0.89 (0.82, 0.95)	8.0×10^{-4}	0.01

354

355

References:

1 Day N, Oakes S, Luben R, *et al.* EPIC-Norfolk: study design and characteristics of the cohort. European Prospective Investigation of Cancer. *Br J Cancer* 1999; **80 Suppl 1**: 95–103.

2 Yu S, Liao KP, Shaw SY, *et al.* Toward high-throughput phenotyping: unbiased automated feature extraction and selection from knowledge sources. *J Am Med Inform Assoc* 2015; **22**: 993–1000.

3 Kelly RS, Virkud Y, Giorgio R, Celedon JC, Weiss ST, Lasky-Su J. Metabolomic profiling of lung function in Costa-Rican children with asthma. *Biochim Biophys acta Mol basis Dis* 2017; **1863**: 1590–5.

4 The Childhood Asthma Management Program (CAMP): design, rationale, and methods. Childhood Asthma Management Program Research Group. *Control Clin Trials* 1999; **20**: 91–120.

5 Strunk RC, Sternberg AL, Szefler SJ, Zeiger RS, Bender B, Tonascia J. Long-term budesonide or nedocromil treatment, once discontinued, does not alter the course of mild to moderate asthma in children and adolescents. *J Pediatr* 2009; **154**: 682–7.

6 Kelly RS, Chawes BL, Blighe K, *et al.* An Integrative Transcriptomic and Metabolomic Study of Lung Function in Children With Asthma. *Chest* 2018; **154**: 335–48.

7 Kelly RS, Sordillo JE, Lutz SM, *et al.* Pharmacometabolomics of Bronchodilator Response in Asthma and the Role of Age-Metabolite Interactions. *Metabolites* 2019; **9**. DOI:10.3390/metabo9090179.