# Sex Differential Expression in HLA Complex Genes and Sex-specific Disease Associations of HLA Region Polymorphisms

## *" HLA Kompleks Genlerinin Ekspresyon Düzeylerinde Cinsiyet Farklılığı "*


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*School of Life Sciences, Pharmacy & Chemistry*

*Kingston University London*


**Istanbul, 29 March 2024**


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# Outline

HLA & Gender Effect in Immunology

HLA & Gender Effect in Genetics

Trait Associations

SNP Effects

eQTL / pQTL Effects

DNA Methylation

Gene / Protein Expression

Conclusion

# Why Is This Important?

HLA region harbours the highest number disease-associated SNPs

Cancer, infections and autoimmune diseases show gender effect

Some disease associations may be sex-specific and missed

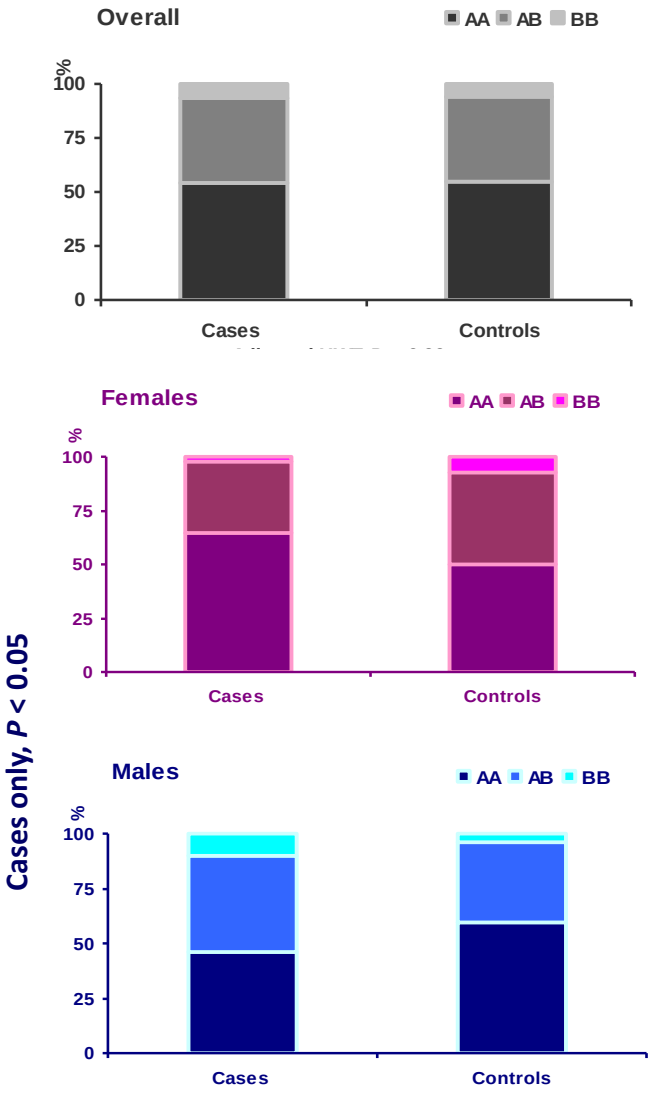
If a marker shows sexually antagonistic associations, the overall analysis will show no association

# Why Is This Important?

***Sexually antagonistic associations result in no overall effect!***

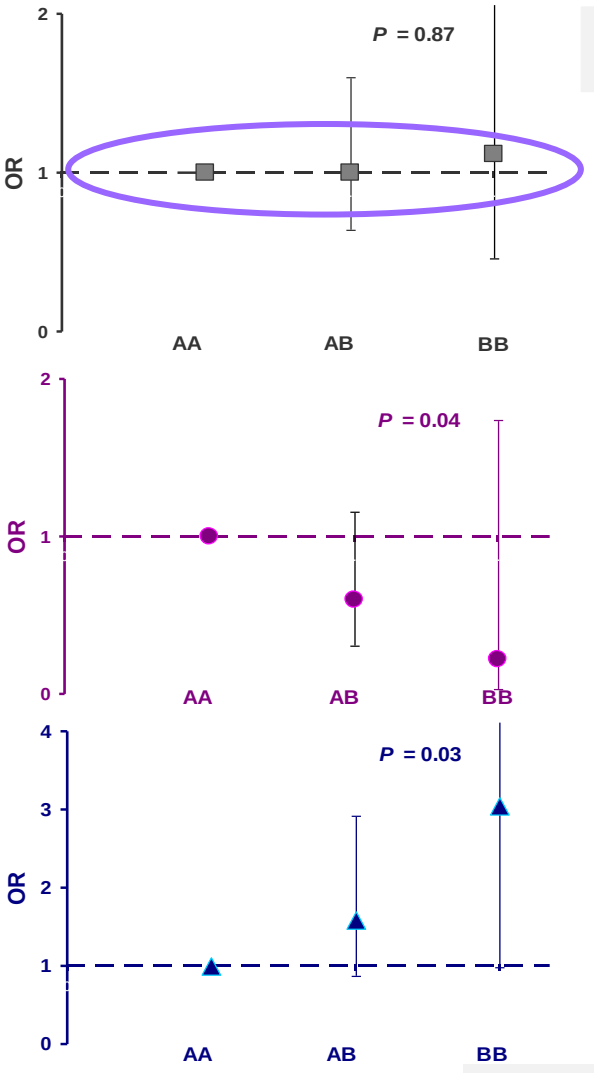
# Effect Modification by Sex

***NRAMP2* rs422982**




Cases only, $P < 0.05$

Controls only, $P < 0.05$

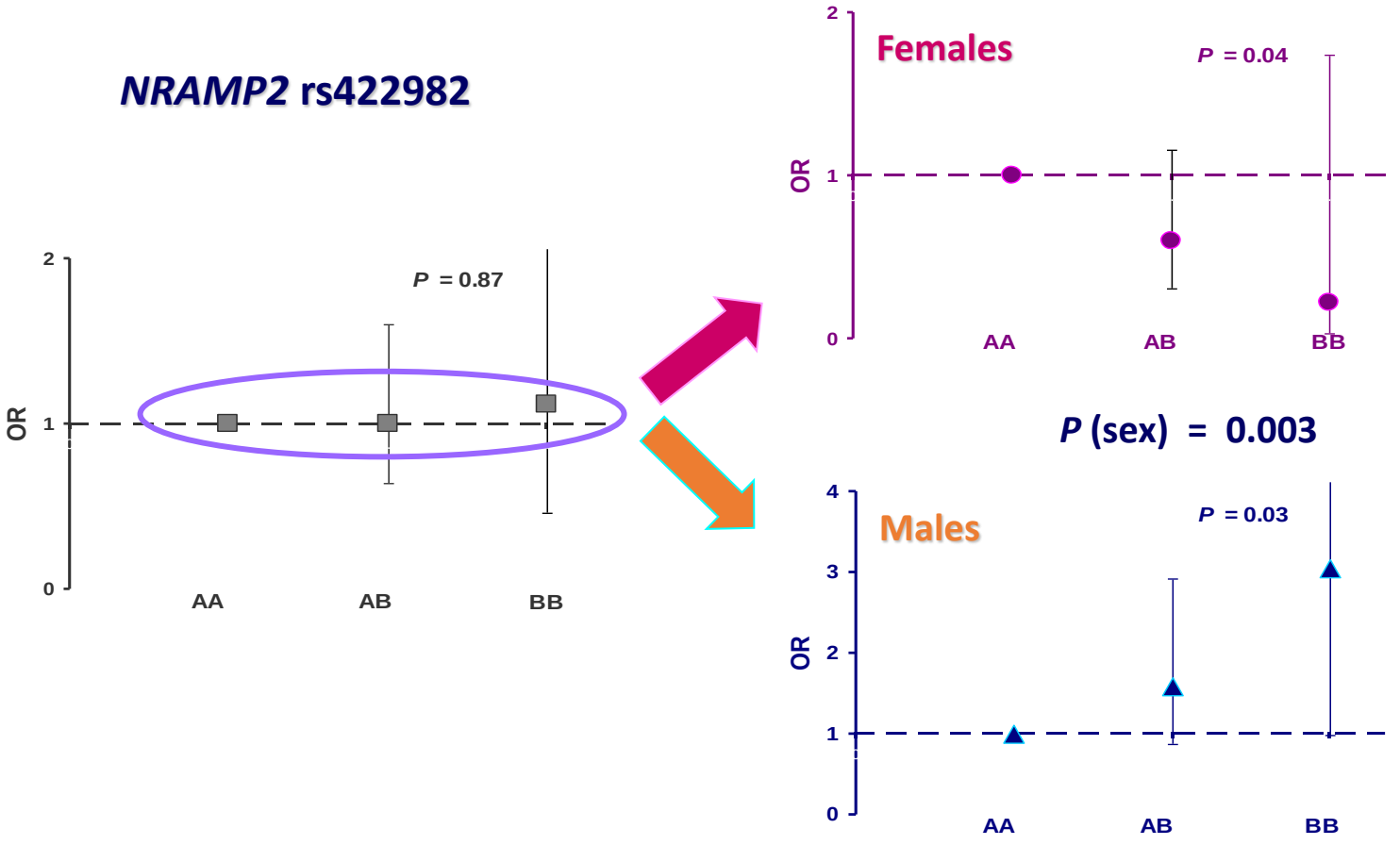



***non-HLA SNPs***

$P$ (sex) = 0.003

*Dorak et al, unpublished*

# Effect Modification by Sex




*non-HLA SNPs*

*Dorak et al, unpublished*

# Effect Modification by Sex

**Sex-differential in trait associations**

ASSOCIATION STUDIES ARTICLE

## Sex-specific autosomal genetic effects across 26 human complex traits

Wan-Yu Lin[1,2,*,†], Chang-Chuan Chan[2,3], Yu-Li Liu[4], Albert C. Yang[5,6], Shih-Jen Tsai[6,7,8,‡] and Po-Hsiu Kuo[1,2,*,¶]

*Human Molecular Genetics*, 2020, Vol. 29, No. 7 1218–1228

doi: 10.1093/hmg/ddaa040
Advance Access Publication Date: 11 March 2020
Association Studies Article

***non-HLA SNPs***

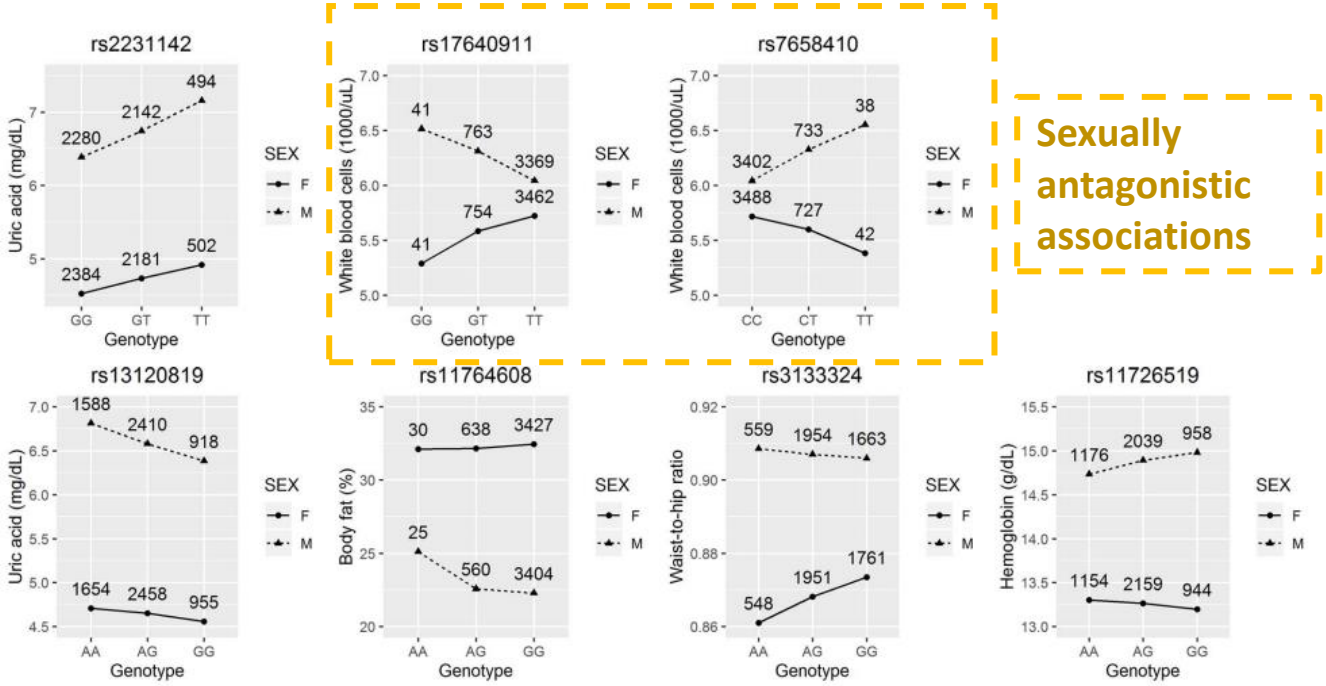



**Figure 1.** SNP-by-sex interactions detected at the genome-wide significance level ($5 \times 10^{-8}$, top row) or by the two-step approach (bottom row).

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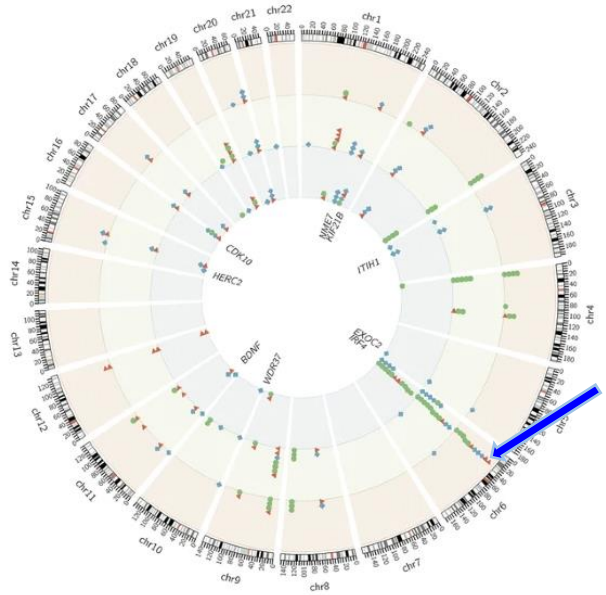
# Effect Modification by Sex

## "Absence of evidence is NOT evidence of absence"

*One of the reasons for getting no positive results in a genetic association study may be effect modification by sex*

*If there is any reason for a sex-specific effect (a priori hypothesis), sex stratification must be applied without worrying about multiple comparisons (and a statistical interaction test)*

# HLA & Disease Associations




Overview of PheWAS associations in the genome after functional annotation. **a** This matrix shows the number of functional SNPs for their respective phenotype. **b** The *Circos plot* showing the PheWAS associations in different types of functional data. *Red triangles* represent the associations in the GWAS Catalog only, *green circles* represent GWAS Catalog associations replicated by PheWAS ($P < 0.05$), and *blue diamonds* represent new phenotype associations identified by PheWAS ($P < 4.6 \times 10^{-6}$ or FDR < 0.1)

**including:**

- **Schizophrenia**
- **Alzheimer disease**
- **Parkinson disease**
- **Lung cancer**
- **Hodgkin lymphoma**

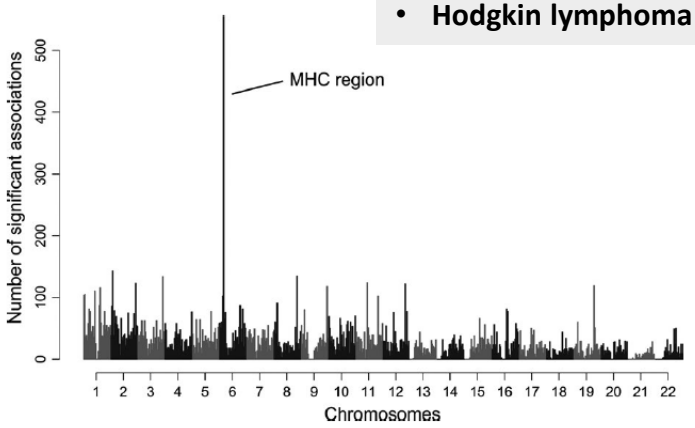



**FIGURE 1** Number of significant GWAS associations along the genome. The chromosomal location of significant trait associations from GWAS (*N* = 18,682) is shown for all autosomes. Data from NHGRI GWAS catalog. Reproduced from "Lenz TL, Spirin V, Jordan DM, Sunyaev SR. Excess of Deleterious Mutations around HLA Genes Reveals Evolutionary Cost of Balancing Selection. Mol Biol Evol 2016;33(10):2555-64. https://doi.org/10.1093/molbev/msw127" by permission of Oxford University Press on behalf of the Society for Molecular Biology and Evolution

**…. despite already showing the highest number of disease associations, the true extent of the involvement of the MHC region in disease genetics may not have been uncovered.**

Received: 9 March 2017 | Revised: 16 June 2017 | Accepted: 20 July 2017

DOI: 10.1111/iji.12332

REVIEW

WILEY INTERNATIONAL JOURNAL OF IMMUNOGENETICS

## What has GWAS done for HLA and disease associations?

A. E. Kennedy[1] | U. Ozbek[2,3] | M. T. Dorak[4]

# HLA & Gender Effect

**Sex-differential in immune system**

PNAS

## Sex bias in MHC I-associated shaping of the adaptive immune system

**Tilman Schneider-Hohendorf[a], Dennis Görlich[b], Paula Savola[c], Tiina Kelkka[c], Satu Mustjoki[c], Catharina C. Gross[a], Geoffrey C. Owens[d], Luisa Klotz[a], Klaus Dornmair[e], Heinz Wiendl[a], and Nicholas Schwab[a,1]**

[a]Department of Neurology, University of Muenster, 48149 Muenster, Germany; [b]Institute of Biostatistics and Clinical Research, University of Muenster, 48149 Muenster, Germany; [c]Hematology Research Unit Helsinki, Department of Clinical Chemistry and Hematology, University of Helsinki and Helsinki University Hospital Comprehensive Cancer Center, 00029 Helsinki, Finland; [d]Department of Neurosurgery, David Geffen School of Medicine at the University of California Los Angeles, Los Angeles, CA 90095; and [e]Institute of Clinical Neuroimmunology, University Hospital and Biomedical Center, Ludwig-Maximilians University Munich, 80539 Munich, Germany

PNAS | **February 27, 2018** | vol. 115 | no. 9

....

**We found that HLA-associated shaping of TCRBV usage differed between the sexes.**

....

**These findings are consistent with studies attributing autoimmunity to processes of epitope spreading and expansion of low-avidity T cell clones ....**

# HLA & Gender Effect

**Sex-differential in trait associations**

European Journal of Human Genetics (2002) 10, 259–265

www.nature.com/ejhg

ARTICLE

## Sex stratification of an inflammatory bowel disease genome search shows male-specific linkage to the HLA region of chromosome 6

Sheila A Fisher*,[1], Jochen Hampe[2], Andrew JS Macpherson[3], Alastair Forbes[4], John E Lennard-Jones[4], Stefan Schreiber[2], Mark E Curran[5], Christopher G Mathew[1] and Cathryn M Lewis[1]

*[1]Division of Medical and Molecular Genetics, Guy's, King's and St Thomas' School of Medicine, King's College London, UK; [2]Department of General Internal Medicine, University Hospital Kiel, Christian-Albrechts-University, Kiel, Germany; [3]Division of Medicine, Guy's, King's and St Thomas' School of Medicine, King's College London, UK; [4]St Mark's Hospital, Harrow, UK; [5]DNA Sciences, Fremont, California, USA*

***But, following GWASs did not examine this sex effect !***

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# HLA & Gender Effect

**Sex-differential in trait associations**

blood®

Volume 94, Issue 2, 15 July 1999, Pages 694-700

Articles

## Unravelling an HLA-DR Association in Childhood Acute Lymphoblastic Leukemia

M. Tevfik Dorak, Tom Lawson, Helmut K.G. Machulla, Chris Darke, Ken I. Mills, Alan K. Burnett

blood®

Volume 94, Issue 11, 1 December 1999, Page 3957

CORRESPONDENCE

## The C282Y Mutation of *HFE* Is Another Male-Specific Risk Factor for Childhood Acute Lymphoblastic Leukemia

M. Tevfik Dorak, Alan K. Burnett, Mark Worwood, Sproul Anne M., Gibson Brenda E.S.

# HLA & Gender Effect

## Sex-differential in trait associations



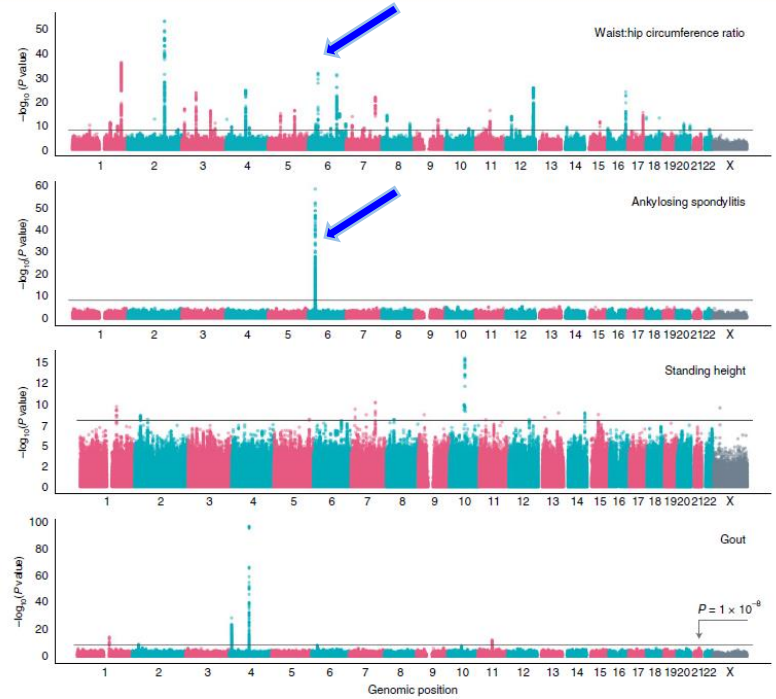


**Fig. 3 | Manhattan plots for traits with most lead sdSNPs.** The *x* axis corresponds to the genomic position in the genome and the *y* axis to the $-\log_{10}(P$ value) of the two-sided Student's *t*-test (Methods), for which the null hypothesis is that there is no difference between the sexes. Each point corresponds to a genetic variant. Points that go above the statistical significance line at $-\log_{10}(P) = 1 \times 10^{-8}$ are considered to be sdSNPs. Traits represented include: waist:hip circumference ratio, ankylosing spondylitis, standing height and gout.

## Sex differences in genetic architecture in the UK Biobank

Elena Bernabeu[1], Oriol Canela-Xandri[2], Konrad Rawlik[1], Andrea Talenti[1], James Prendergast[1] and Albert Tenesa[1,2] ✉

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# HLA & Gender Effect

## Sex-differential in trait associations

ASSOCIATION STUDIES ARTICLE

### Sex-specific autosomal genetic effects across 26 human complex traits

Wan-Yu Lin[1,2,*,†], Chang-Chuan Chan[2,3], Yu-Li Liu[4], Albert C. Yang[5,6], Shih-Jen Tsai[6,7,8,‡] and Po-Hsiu Kuo[1,2,*,¶]

*Human Molecular Genetics*, 2020, Vol. 29, No. 7 1218–1228

doi: 10.1093/hmg/ddaa040
Advance Access Publication Date: 11 March 2020
Association Studies Article

***non-HLA SNPs***

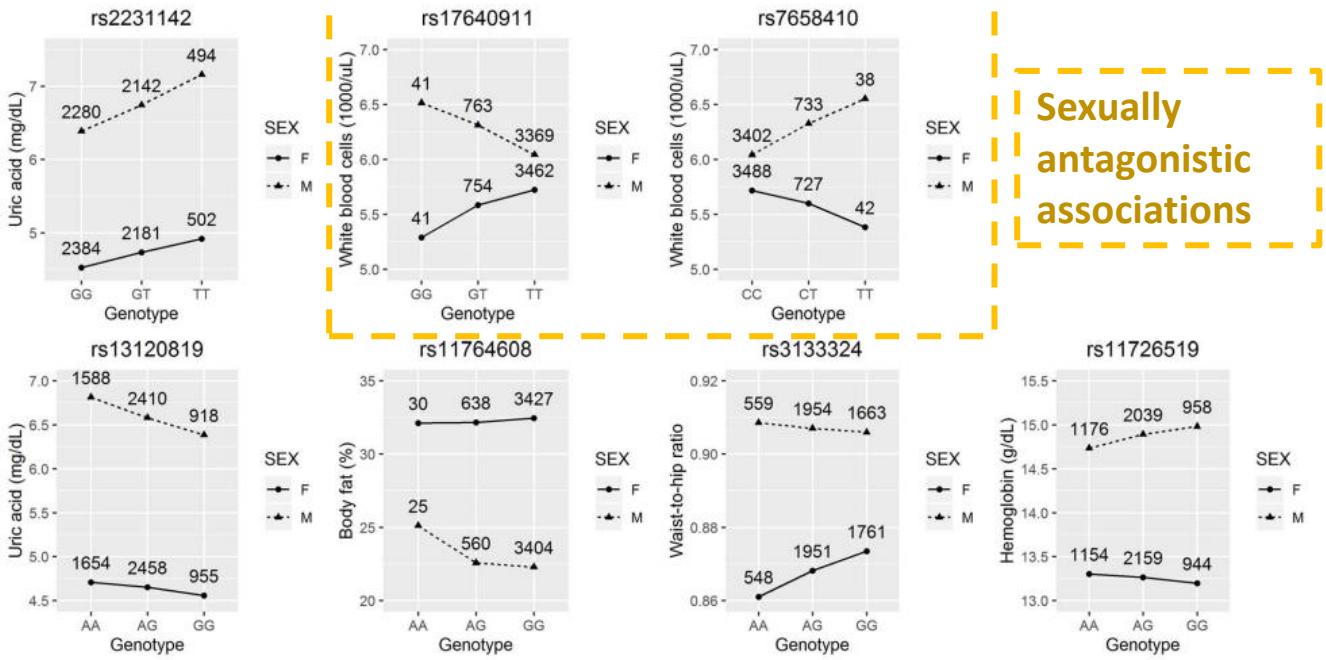


**Figure 1.** SNP-by-sex interactions detected at the genome-wide significance level ($5 \times 10^{-8}$, top row) or by the two-step approach (bottom row).

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# HLA & Gender Effect

## Sex-differential in trait associations

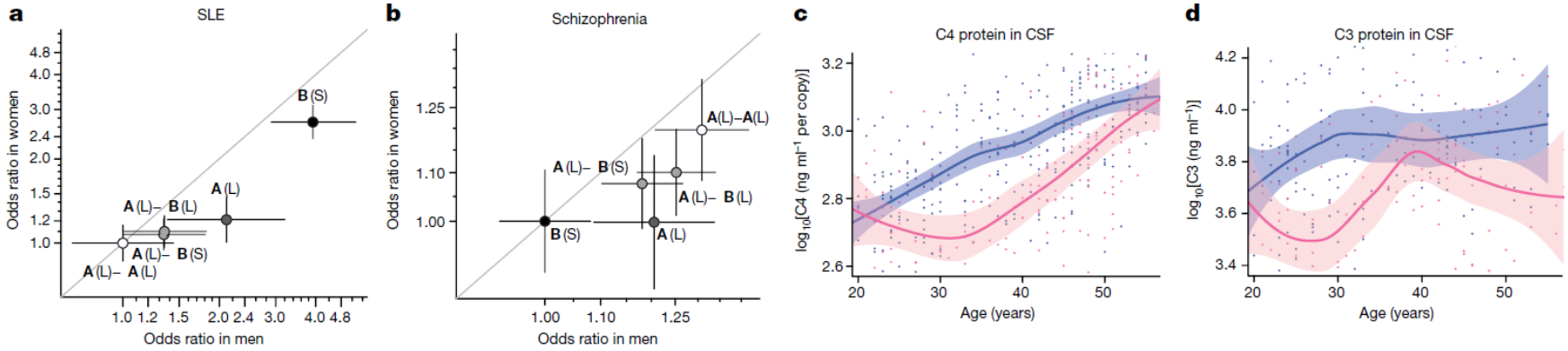


**Fig. 3 | Sex differences in the magnitude of C4 genetic effects and complement protein concentrations. a**, SLE risk (odds ratios) associated with the four most common C4 alleles in men (*x* axis) and women (*y* axis) among 6,748 affected and 11,516 unaffected individuals of European ancestry. For each sex, the lowest-risk allele (C4-A(L)-A(L)) is used as a reference (odds ratio of 1.0). Shading of each point reflects the relative level of SLE risk (darker indicates greater risk) conferred by *C4A* and *C4B* copy numbers as in Fig. 2b. Error bars represent 95% confidence intervals around the effect size estimate for each sex. **b**, Schizophrenia risk (odds ratios) associated with the four most common C4 alleles in men (*x* axis) and women (*y* axis) among 28,799 affected and 35,986 unaffected individuals of European ancestry, aggregated by the Psychiatric Genomics Consortium[43]. For each sex, the lowest-risk allele (C4-B(S)) is used as a reference (odds ratio of 1.0). For visual comparison with **a**, shading of each allele reflects the relative level of SLE risk. Error bars represent 95% confidence intervals around the effect size estimate for each sex. **c**, Concentrations of C4 protein in CSF sampled from 340 adult men (blue) and 167 adult women (pink) as a function of age with locally estimated scatterplot smoothing (LOESS). Concentrations are normalized to the number of C4 gene copies in an individual's genome (a strong independent source of variance, Extended Data Fig. 7a) and shown on a $\log_{10}$ scale as a LOESS curve. Shaded regions represent 95% confidence intervals derived during LOESS. **d**, Levels of C3 protein in CSF from 179 adult men and 125 adult women as a function of age. Concentrations are shown on a $\log_{10}$ scale as a LOESS curve. Shaded regions represent 95% confidence intervals derived during LOESS.

## The *C4* gene is within the HLA class III region

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Nature | Vol 582 | 25 June 2020

## Complement genes contribute sex-biased vulnerability in diverse disorders

https://doi.org/10.1038/s41586-020-2277-x
Received: 14 October 2019
Accepted: 28 February 2020
Published online: 11 May 2020
Check for updates

Nolan Kamitaki[1,2]✉, Aswin Sekar[1,2], Robert E. Handsaker[1,2], Heather de Rivera[1,2], Katherine Tooley[1,2], David L. Morris[3], Kimberly E. Taylor[4], Christopher W. Whelan[1,2], Philip Tombleson[3], Loes M. Olde Loohuis[5,6], Schizophrenia Working Group of the Psychiatric Genomics Consortium*, Michael Boehnke[7], Robert P. Kimberly[8], Kenneth M. Kaufman[9], John B. Harley[9], Carl D. Langefeld[10], Christine E. Seidman[11,12], Michele T. Pato[13], Carlos N. Pato[13], Roel A. Ophoff[5,6], Robert R. Graham[14], Lindsey A. Criswell[4], Timothy J. Vyse[3]✉ & Steven A. McCarroll[1,2]✉

# HLA & Gender Effect

**Sex-differential in SNP effects**

- **We obtained cell-specific functionality scores (range=0 to 1) of 435,924 SNPs within xMHC from FUN-LDA, which provides separate composite scores based on genomic effects in ENCODE and Epigenomics Roadmap project cells.**
- **We extracted the SNPs with scores substantially (>0.95) different between hematopoietic stem cells (HSC) from males and females {E050/E051}. 321 SNPs had substantial score differential in HSC between male and female cells. They were spread across xMHC.**
- **These SNPs showed 80 cis- and 201 trans-eQTL effects. Among the cis targets were xMHC genes previously identified as sex-associated in their expression such as HLA-DRB1, -DPB1, -F, IER3, MDC1 and various histone genes.**
- **The UK BioBank GWAS data revealed associations with anthropometric traits and autoimmune disorders with sexual differential in their prevalence.**

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FUN-LDA: A Latent Dirichlet Allocation Model for Predicting Tissue-Specific Functional Effects of Noncoding Variation: Methods and Applications

Daniel Backenroth,[1] Zihuai He,[1] Krzysztof Kiryluk,[2] Valentina Boeva,[3,4] Lynn Petukhova,[5,6] Ekta Khurana,[7] Angela Christiano,[6,8] Joseph D. Buxbaum,[9,10] and Iuliana Ionita-Laza[1,*]

***Dorak et al, unpublished***

# HLA & Gender Effect

**Sex-differential eQTL effects**

*Human Molecular Genetics, 2014, Vol. 23, No. 7* ***1947–1956***
*doi:10.1093/hmg/ddt582*
*Advance Access published on November 15, 2013*

## Sex- and age-interacting eQTLs in human complex diseases

**Chen Yao[1,2,3], Roby Joehanes[1,2,3], Andrew D. Johnson[1,2], Tianxiao Huan[1,2,3], Tõnu Esko[8,9,10], Saixia Ying[4], Jane E. Freedman[5], Joanne Murabito[1,6], Kathryn L. Lunetta[7], Andres Metspalu[8,11], Peter J. Munson[4] and Daniel Levy[1,2,3,*]**

## Sex-heterogeneous SNPs disproportionately influence gene expression and health

Michela Traglia, Margaux Bout, Lauren A. Weiss

Kingston University London

# HLA & Gender Effect

## Sex-differential eQTL effects

*Human Molecular Genetics, 2014, Vol. 23, No. 7* **1947–1956**
*doi:10.1093/hmg/ddt582*
*Advance Access published on November 15, 2013*

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**Table 1.** Sex-interaction eQTLs in human complex traits

| eQTL | Chromosome: Position | Trait/disease | Expressed gene | *P*-value (interaction) | *P*-value (main effect) |
|---|---|---|---|---|---|
| rs9302752 | 16 : 50 719 103 | Leprosy | NOD2 | 8.15E-10 | 2.41E-35 |
| rs9270986 (rs3129889 rs3135388 rs9271366) | 6 : 32 574 060 | Multiple sclerosis | HLA-DRB5 | 4.33E-09 | 1.29E-218 |
| rs3129860 | 6 : 32 401 079 | Diabetes mellitus, Type 1 | HLA-DRB5 | 2.57E-05 | 1.99E-121 |
| rs1335515 | 14 : 58 385 365 | Attention deficit disorder with hyperactivity | KIAA0586 | 4.00E-06 | NS |
| rs12145451 | 1 : 212 425 291 | Heart failure | PPP2R5A | 3.35E-06 | 1.27E-15 |
| rs8047080 (rs3868142 rs3868143 rs11860295) | 16 : 67 402 588 | Lipoproteins, HDL | TSNAXIP1 | 6.08E-06 | NS |
| rs599634 | 6 : 50 013 663 | Body mass index | MUT | 8.35E-06 | NS |
| rs846793 | 6 : 101 305 983 | Memory | GRIK2 | 8.6E-06 | NS |
| rs11634397 | 15 : 80 432 222 | Diabetes mellitus, Type 2 | C15orf37 | 9.60E-06 | NS |
| rs17291650 | 12 : 51 213 433 | Death, sudden, cardiac | LIMA1 | 1.66E-05 | NS |
| rs6867983 | 5 : 55 854 153 | Triglycerides | IL6ST | 2.67E-05 | NS |
| rs3132610 | 6 : 30 544 401 | Lupus erythematosus, systemic | HCG8 | 3.71E-05 | NS |
| rs644148 | 19 : 44 970 935 | Personality | BLOC1S3 | 3.93E-05 | NS |
| rs10503734 | 8 : 23 528 230 | Blood pressure | NKX3-1 | 5.07E-05 | NS |
| X-Chromosome eQTL | | | | | |
| rs5991441 | X : 42 930 414 | Lipids | CXorf23 (X: 19 930 978–19 988 416) | 2.70E-10 | NS |
| rs5991573 | X : 42 930 110 | Blood pressure | | 2.73E-10 | NS |
| rs5991545 | X : 42 889 004 | Diabetes mellitus | | 2.17E-09 | NS |

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# HLA & Gender Effect

## Sex-differential pQTL effects

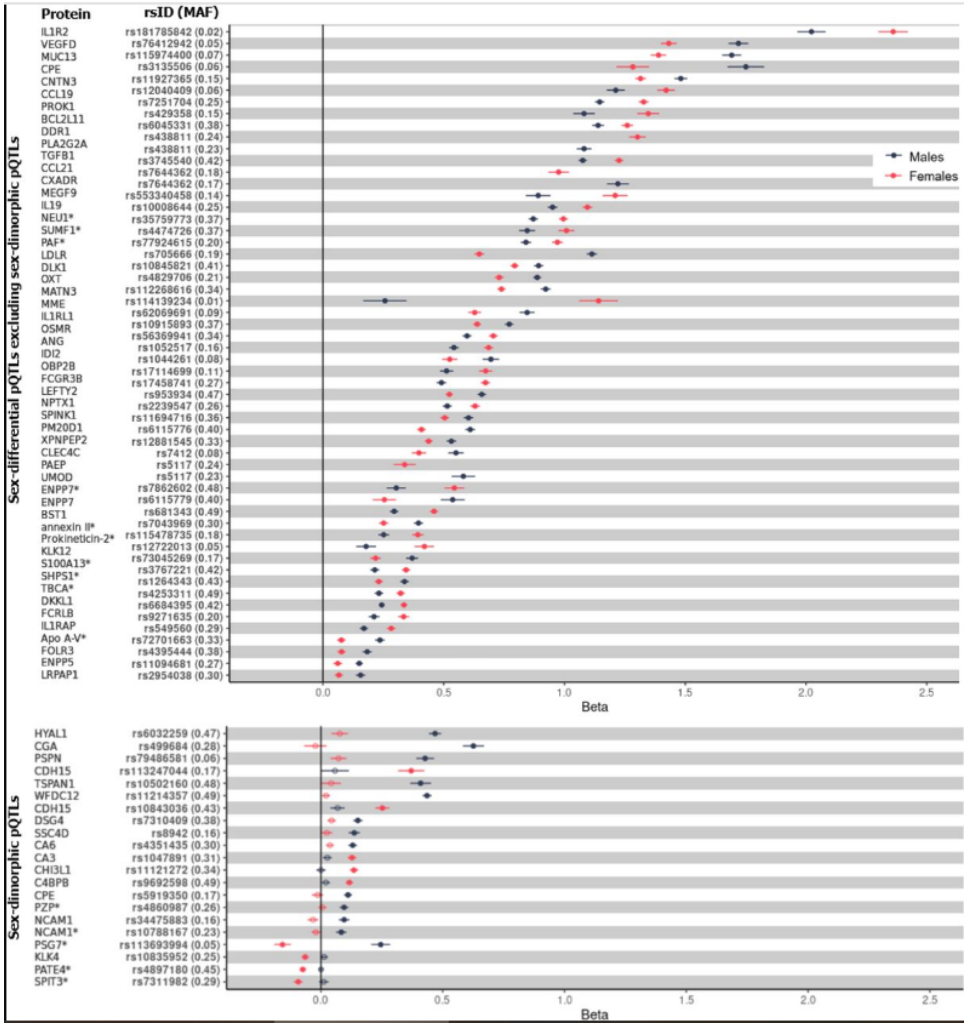


**Figure 2: Forest plot of all identified sex-differential protein quantitative trait loci (sd-pQTLs) from both aptamer- ($p_{het}1.01\times10^{-11}$) and antibody-based ($p_{het}3.42\times10^{-11}$) technologies.**

**Includes:**
***DDR1*** rs6045331
***NEU1*** rs35759773

**Similar and different: systematic investigation of proteogenomic variation between sexes and its relevance for human diseases**

Mine Koprulu, Eleanor Wheeler, Nicola D. Kerrison, Spiros Denaxas, Julia Carrasco-Zanini, Chloe M. Orkin, Harry Hemingway, Nicolas J. Wareham, Maik Pietzner, Claudia Langenberg

**doi:** https://doi.org/10.1101/2024.02.16.24302936

# HLA & Gender Effect

## Sex-differential in microRNA gene regulation

Sharma and Eghbali *Biology of Sex Differences* 2014, **5**:3
http://www.bsd-journal.com/content/5/1/3

BIOLOGY OF SEX DIFFERENCES

REVIEW Open Access

### Influence of sex differences on microRNA gene regulation in disease

Salil Sharma and Mansoureh Eghbali*

**Table 1 MicroRNAs showing sex-biased expression in different pathological conditions**

| Disease/development | MicroRNAs | Expression changes between males and females | Hormone regulated | Chromosome linked |
|---|---|---|---|---|
| Autoimmune diseases | | | | |
| Systemic lupus erythematosus | miR-182 cluster, miR-31, and miR-148a [84] | M < F | Estrogens | - |
| Neurodegenerative diseases | | | | |
| Schizophrenia | miR-30b [85] | M > F | Estrogens | |
| | let-7f-2, miR-18b, miR-505, miR-502, miR-188, miR-325, miR-660 and miR-509-3 [41] | No expression changes reported (only mutational changes between disease and control) | | X chromosome |
| Cerebral ischemia | miR-23a [37] | M < F | - | - |
| Neurodevelopment | miR-322, miR-574, and miR-873 [65] | M < F | Estrogens | - |
| Metabolic diseases | miR-221, let-7 g [86] | M < F | - | - |
| Breast cancer | miR17, let-7a [87]; | M < F | - | - |
| | miR-137 [35] | M < F promoter methylation of miR-137 | - | - |
| Liver fibrosis | miR-29a, miR-29b [88] | M < F | Estrogens | - |

(*None of these microRNAs are encoded in chromosome 6*)

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# HLA & Gender Effect

## Sex-differential in DNA methylation

Grant *et al. Clinical Epigenetics* (2022) 14:62
https://doi.org/10.1186/s13148-022-01279-7

Clinical Epigenetics

RESEARCH Open Access

### Characterising sex differences of autosomal DNA methylation in whole blood using the Illumina EPIC array

Olivia A. Grant[1,2,3], Yucheng Wang[1,4], Meena Kumari[2], Nicolae Radu Zabet[1,3*] and Leonard Schalkwyk[1*]

*Published online 17 August 2021* *Nucleic Acids Research, 2021, Vol. 49, No. 16* **9097–9116**
*https://doi.org/10.1093/nar/gkab682*

### Autosomal sex-associated co-methylated regions predict biological sex from DNA methylation

**Evan Gatev[1,2,3,4,5,6], Amy M. Inkster[4,5], Gian Luca Negri[7], Chaini Konwar[4,5,6], Alexandre A. Lussier[8,9,10], Anne Skakkebaek[11,12], Marla B. Sokolowski[13,14], Claus H. Gravholt[12,15], Erin C. Dunn[8,9,10], Michael S. Kobor[4,5,6,14] and Maria J. Aristizabal[4,5,6,13,14,16,*]**

Inoshita *et al. Biology of Sex Differences* (2015) 6:11
DOI 10.1186/s13293-015-0029-7

BIOLOGY OF SEX DIFFERENCES

RESEARCH Open Access

### Sex differences of leukocytes DNA methylation adjusted for estimated cellular proportions

Masatoshi Inoshita[1], Shusuke Numata[1*], Atsushi Tajima[2,3], Makoto Kinoshita[1], Hidehiro Umehara[1], Hidenaga Yamamori[4,5], Ryota Hashimoto[5,6], Issei Imoto[2] and Tetsuro Ohmori[1]

### Genetic and environmental influences interact with age and sex in shaping the human methylome

Jenny van Dongen[1,*], Michel G. Nivard[1,*], Gonneke Willemsen[1], Jouke-Jan Hottenga[1], Quinta Helmer[1], Conor V. Dolan[1], Erik A. Ehli[2], Gareth E. Davies[2], Maarten van Iterson[3], Charles E. Breeze[4], Stephan Beck[4], BIOS Consortium[†], H. Eka Suchiman[3], Rick Jansen[5], Joyce B. van Meurs[6], Bastiaan T. Heijmans[3,**], P. Eline Slagboom[3,**] & Dorret I. Boomsma[1,**]

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# HLA & Gender Effect

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**We identified 266 significant sex-associated DMRs on the autosomes between males and females located at 231 unique sets of genes (Additional file 2)**

ADDITIONAL FILE 2 (xMHC):

| chr6_start | end | overlapping.promoters |
|---|---|---|
| 26195488 | 26197733 | HIST1H2AD, HIST1H3D, HIST1H1PS1 |
| 27205325 | 27206911 | NA |
| 27568835 | 27570548 | NA |
| 28457624 | 28460106 | NA |
| 28552582 | 28558113 | SCAND3, RP5-1186N24.3 |
| 28601271 | 28603779 | RP11-373N24.2 |
| 28677112 | 28678885 | NA |
| 30684284 | 30692147 | TUBB, MDC1XXbac-BPG252P9.9 |
| 30848178 | 30854551 | **DDR1** |
| 31777883 | 31783545 | HSPA1A, HSPA1L |
| 32728786 | 32730859 | HLA-DQB2 |
| 33313260 | 33313437 | NA |

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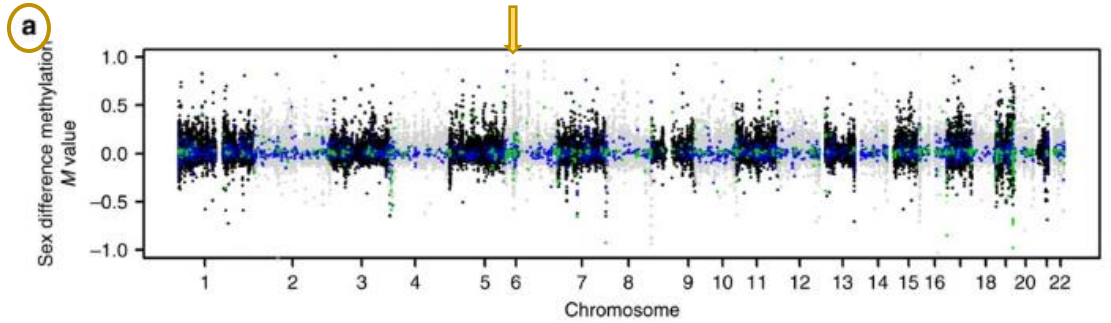
# HLA & Gender Effect

## Sex-differential in DNA methylation

### Genetic and environmental influences interact with age and sex in shaping the human methylome

Jenny van Dongen[1,*], Michel G. Nivard[1,*], Gonneke Willemsen[1], Jouke-Jan Hottenga[1], Quinta Helmer[1], Conor V. Dolan[1], Erik A. Ehli[2], Gareth E. Davies[2], Maarten van Iterson[3], Charles E. Breeze[4], Stephan Beck[4], BIOS Consortium[†], H. Eka Suchiman[3], Rick Jansen[5], Joyce B. van Meurs[6], Bastiaan T. Heijmans[3,**], P. Eline Slagboom[3,**] & Dorret I. Boomsma[1,**]

**Figure 4: Main effects of age and sex and their interaction with genetic and environmental effects.**




(**a**) Main and interaction effects of sex. The *x* axis denotes chromosomal position. The *y* axis denotes the $\beta$-value from the regression of methylation *M* value on sex (while correcting for age, white blood cell counts and technical covariates). Sites where the genetic variance showed significant sex interaction are indicated in blue. Sites where the environmental variance showed significant sex interaction are shown in green. The *y* axis is truncated at −1 and 1 to improve visibility, excluding a small number of sites with regression $\beta$-values up to −2.4 and 2.6. (**b**) Mean and interaction effects of age. The *x* axis denotes chromosomal position. The *y* axis denotes the $\beta$-value from the regression of methylation *M* value on age (while correcting for sex, white blood cell counts and technical covariates). Sites where the genetic variance showed significant age interaction are indicated in blue. Sites where the environmental variance showed significant age interaction are shown in green. Results are based on data from 2,603 individuals.

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# HLA & Gender Effect

## Sex-differential in DNA methylation

Inoshita *et al. Biology of Sex Differences* (2015) 6:11
DOI 10.1186/s13293-015-0029-7

BIOLOGY OF SEX DIFFERENCES

RESEARCH Open Access

### Sex differences of leukocytes DNA methylation adjusted for estimated cellular proportions

Masatoshi Inoshita[1], Shusuke Numata[1*], Atsushi Tajima[2,3], Makoto Kinoshita[1], Hidehiro Umehara[1], Hidenaga Yamamori[4,5], Ryota Hashimoto[5,6], Issei Imoto[2] and Tetsuro Ohmori[1]

**Additional File 2:**
**xMHC-CpG islands that show differences in methylation levels between males and females:**

| Rank | CpG | Gene | Chr | Pos_hg19 | Mean β (M) | Mean β (F) | Diff β | Gender p-value |
|---|---|---|---|---|---|---|---|---|
| 90 | cg17871437 | *BRD2* | 6 | 32940801 | 0.019 | 0.049 | -0.030 | 6.28E-13 |
| 131 | cg21144120 | | 6 | 28661199 | 0.352 | 0.439 | -0.087 | 3.08E-11 |
| 134 | cg00934883 | *IER3* | 6 | 30711966 | 0.091 | 0.101 | -0.010 | 3.84E-11 |
| 149 | cg23250574 | | 6 | 28661310 | 0.222 | 0.312 | -0.089 | 8.72E-11 |
| 201 | cg05973262 | *NOTCH4* | 6 | 32191895 | 0.494 | 0.536 | -0.042 | 1.26E-09 |
| 204 | cg24642916 | *GPSM3* | 6 | 32163304 | 0.024 | 0.030 | -0.006 | 1.37E-09 |
| 240 | cg09314434 | *CSNK2B* | 6 | 31632207 | 0.406 | 0.462 | -0.057 | 3.56E-09 |
| 253 | cg27011480 | *HLA-L* | 6 | 30228083 | 0.151 | 0.220 | -0.068 | 5.42E-09 |
| 362 | cg02030913 | *NFKBIL1* | 6 | 31515391 | 0.123 | 0.085 | 0.037 | 5.99E-08 |
| 382 | cg01530154 | | 6 | 28661712 | 0.175 | 0.210 | -0.035 | 9.35E-08 |
| 386 | cg08864245 | *SYNGAP1* | 6 | 33395936 | 0.019 | 0.008 | 0.010 | 1.02E-07 |
| 404 | cg09523275 | *NKAPL* | 6 | 28227093 | 0.274 | 0.321 | -0.047 | 1.22E-07 |

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# HLA & Gender Effect

## Sex-differential in gene expression

Gershoni and Pietrokovski *BMC Biology* (2017) 15:7
DOI 10.1186/s12915-017-0352-z

BMC Biology

RESEARCH ARTICLE Open Access

### The landscape of sex-differential transcriptome and its consequent selection in human adults

Moran Gershoni* and Shmuel Pietrokovski

OXFORD

*Briefings in Bioinformatics*, 19(2), 2018, 188–198

doi: 10.1093/bib/bbw125
Advance Access Publication Date: 21 December 2016
Paper

### Identification and analysis of the human sex-biased genes

Sisi Guo, Yuan Zhou, Pan Zeng, Guoheng Xu, Guoqing Wang and Qinghua Cui

Jansen *et al. BMC Genomics* 2014, **15**:33
http://www.biomedcentral.com/1471-2164/15/33

BMC Genomics

RESEARCH ARTICLE Open Access

### Sex differences in the human peripheral blood transcriptome

Rick Jansen[1*], Sandra Batista[5], Andrew I Brooks[4], Jay A Tischfield[4], Gonneke Willemsen[2], Gerard van Grootheest[1], Jouke-Jan Hottenga[2], Yuri Milaneschi[1], Hamdi Mbarek[2], Vered Madar[3], Wouter Peyrot[1], Jacqueline M Vink[2], Cor L Verweij[6], Eco JC de Geus[2], Johannes H Smit[1], Fred A Wright[3], Patrick F Sullivan[5], Dorret I Boomsma[2†] and Brenda WJH Penninx[1†]

### Sex Differences in Gene Expression and Regulatory Networks across 29 Human Tissues

Camila M. Lopes-Ramos,[1] Cho-Yi Chen,[2] Marieke L. Kuijjer,[3] Joseph N. Paulson,[4] Abhijeet R. Sonawane,[5] Maud Fagny,[6] John Platig,[5] Kimberly Glass,[1,5] John Quackenbush,[1,5,7,*] and Dawn L. DeMeo[5,8,9,*]

### The human transcriptome across tissues and individuals

Marta Melé,[1,2*] Pedro G. Ferreira,[1,3,4,5*] Ferran Reverter,[1,6,7*] David S. DeLuca,[8] Jean Monlong,[1,7,9] Michael Sammeth,[1,7,10] Taylor R. Young,[8] Jakob M Goldmann,[1,7,11] Dmitri D. Pervouchine,[1,7,12] Timothy J. Sullivan,[8] Rory Johnson,[1,7] Ayellet V. Segrè,[8] Sarah Djebali,[1,7] Anastasia Niarchou,[3,4,5] The GTEx Consortium, Fred A. Wright,[13] Tuuli Lappalainen,[3,4,5,14,15] Miquel Calvo,[6] Gad Getz,[8,16] Emmanouil T. Dermitzakis,[3,4,5] Kristin G. Ardlie,[8†] Roderic Guigó[1,7,17,18†]

### The impact of sex on gene expression across human tissues

Meritxell Oliva*†, Manuel Muñoz-Aguirre†, Sarah Kim-Hellmuth†, Valentin Wucher, Ariel D. H. Gewirtz, Daniel J. Cotter, Princy Parsana, Silva Kasela, Brunilda Balliu, Ana Viñuela, Stephane E. Castel, Pejman Mohammadi, François Aguet, Yuxin Zou, Ekaterina A. Khramtsova, Andrew D. Skol, Diego Garrido-Martín, Ferran Reverter, Andrew Brown, Patrick Evans, Eric R. Gamazon, Anthony Payne, Rodrigo Bonazzola, Alvaro N. Barbeira, Andrew R. Hamel, Angel Martinez-Perez, José Manuel Soria, GTEx Consortium, Brandon L. Pierce, Matthew Stephens, Eleazar Eskin, Emmanouil T. Dermitzakis, Ayellet V. Segrè, Hae Kyung Im, Barbara E. Engelhardt, Kristin G. Ardlie, Stephen B. Montgomery, Alexis J. Battle, Tuuli Lappalainen, Roderic Guigó, Barbara E. Stranger*

### Large Scale Gene Expression Meta-Analysis Reveals Tissue-Specific, Sex-Biased Gene Expression in Humans

*Benjamin T. Mayne[1,2], Tina Bianco-Miotto[1,3], Sam Buckberry[4,5], James Breen[1,6], Vicki Clifton[7], Cheryl Shoubridge[1,2] and Claire T. Roberts[1,2*]*

### Tissue-specific sex differences in human gene expression

Irfahan Kassam[1], Yang Wu[1], Jian Yang[1,2], Peter M. Visscher[1,2] and Allan F. McRae[1,2,*]

SAGD
Sex-Associated Gene Database

SAGD: a comprehensive sex-associated gene database from transcriptomes

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# HLA & Gender Effect

**Sex-differential in gene expression**

## The human transcriptome across tissues and individuals

Marta Melé,[1,2*] Pedro G. Ferreira,[1,3,4,5*] Ferran Reverter,[1,6,7*] David S. DeLuca,[8] Jean Monlong,[1,7,9] Michael Sammeth,[1,7,10] Taylor R. Young,[8] Jakob M Goldmann,[1,7,11] Dmitri D. Pervouchine,[1,7,12] Timothy J. Sullivan,[8] Rory Johnson,[1,7] Ayellet V. Segrè,[8] Sarah Djebali,[1,7] Anastasia Niarchou,[3,4,5] The GTEx Consortium, Fred A. Wright,[13] Tuuli Lappalainen,[3,4,5,14,15] Miquel Calvo,[6] Gad Getz,[8,16] Emmanouil T. Dermitzakis,[3,4,5] Kristin G. Ardlie,[8†] Roderic Guigó[1,7,17,18†]

| 1 | Table S10 | Genes differentially expressed between males and females | | | | |
|---|---|---|---|---|---|---|
| 2 | | | | | | |
| 3 | gene_id | gene_name | GeneGroup | chr | coeff | BH |
| 28 | ENSG00000080007.6 | DDX43 | protein_coding | chr6 | -0.689309281 | 6.95E-09 |
| 43 | ENSG00000203907.5 | OOEP | protein_coding | chr6 | -0.315642044 | 5.87E-05 |
| 72 | ENSG00000124557.7 | BTN1A1 | protein_coding | chr6 | 0.113386817 | 0.001167335 |
| 95 | ENSG00000214922.3 | HLA-F-AS1 | lncRNA | chr6 | 0.230836095 | 0.00633945 |

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# HLA & Gender Effect

## Sex-differential in gene expression

### Tissue-specific sex differences in human gene expression

Irfahan Kassam[1], Yang Wu[1], Jian Yang[1,2], Peter M. Visscher[1,2] and Allan F. McRae[1,2,*]

**Description:** Summary statistics for 131 TSSD transcripts. A total of **65 autosomal** and 66 X-linked transcripts showed **significant heterogeneity in sex-difference effect sizes across tissue-types** at a Bonferroni corrected significance threshold of P_TSSD <=1.58 x 10^{-6}. **Concordant direction of male/female sex-differences was observed for 45 (12 autosomal and 33 X-linked) TSSD transcripts**. The remaining **86 (53 autosomal and 33 X-linked) TSSD transcripts showed discordant direction of sex-differences in at least one of the expressed tissues.** Shown are the number of tissues the transcripts are expressed in. The tissue effect, sex effect, and TSSD effect correspond to test statistics from Model 1 (see Methods and

| | | | | | Sex Effect | | Tissue Effect | | TSSD Effect | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Chr | Start | Stop | Gene | F-statist | P.sex | F-statistic | P.tissu | F-statist | P.tssd | Concordanc |
| 53 | 6 | 74104857 | 74127292 | DDX43 | 97.36 | 1.69E-21 | 0.79 | 8.22E-01 | 4.98 | 3.81E-22 | concordant |
| 94 | 6 | 144261437 | 144385735 | PLAGL1 | 0.38 | 5.37E-01 | 0.41 | 1.00E+00 | 2.85 | 8.78E-09 | discordant |
| 117 | 6 | 132617194 | 132722684 | MOXD1 | 12.85 | 3.59E-04 | 0.32 | 1.00E+00 | 2.60 | 2.00E-07 | discordant |
| 126 | 6 | **28227098** | **28228736** | **NKAPL** | 35.30 | 4.46E-09 | 0.24 | 1.00E+00 | 2.48 | 8.80E-07 | discordant |

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# HLA & Gender Effect

**Sex-differential in gene expression**

- In a preliminary study, we (Cakir & Dorak, unpublished) screened sex-specific transcriptomics studies to identify MHC-linked genes with differential expression by sex.
- In the **Sex-Associated Gene Database** (SAGD), only the histone gene ***HIST1H3G*** (*H3C8*) had a sex differential with log2 fold change >2.0 with more than 7-times difference in its expression between males and females in blood.
- Various transcriptomics studies (incl Guo et al, 2018) showed additional genes within xMHC with sex-associated expression such as ***HLA-DRB1, -DPB1, -F, IER3, MDC1, GTF2H4*** and ***AIF1***, but most consistently with MHC-linked **histone** genes (including *HIST1H3G*).

***HIST1H3G*: AR / SRY /MAZ TF binding site (ENCODE Chip-seq)**

SAGD
Sex-Associated Gene Database

SAGD: a comprehensive sex-associated gene database from transcriptomes

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# Conclusions

**There are examples of sex-specific or sex-dimorphic trait associations, gene expressions, DNA methylations, eQTL/pQTL effects and SNP functional impact in the HLA region**

**Despite this knowledge, there is persistence with traditional ways of analysing data which may obscure sex-specific results, especially sexually antagonistic ones**

**Histone genes within the extended MHC appear to be most consistent finding in sex-specific gene expression**

# *Thank You*


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