

Introducing the center for genetics, genomics, and epigenetics of substance use disorders in outbred rats

Abraham Palmer

Department of Psychiatry

University of California San Diego

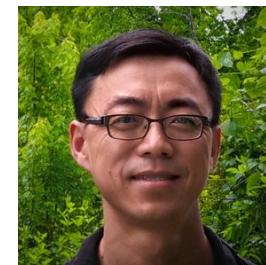


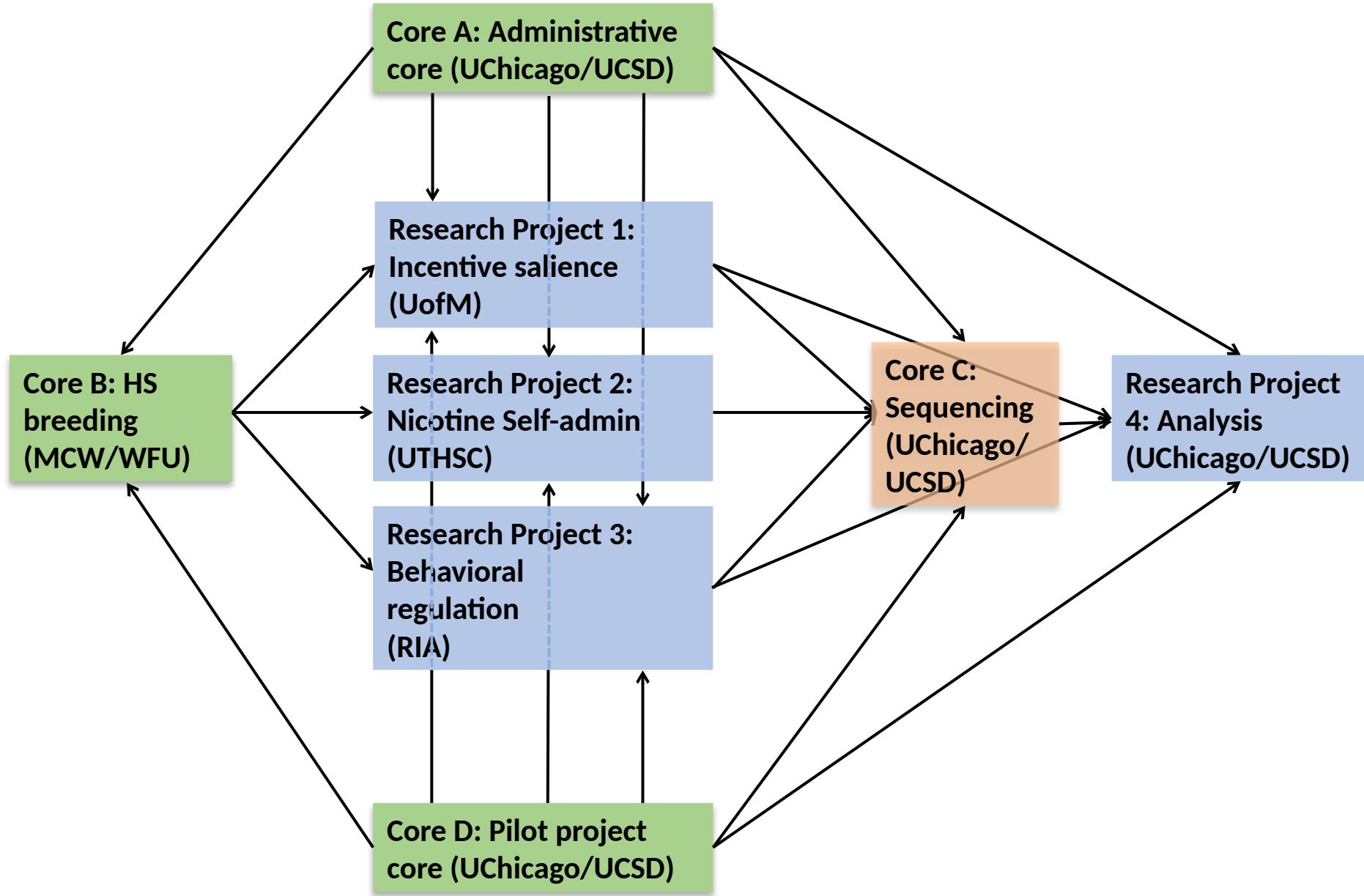
NIDA Center of Excellence for
Genetics, Genomics, and Epigenetics
of Substance Use Disorders in Outbred Rats

Original cast

History of the center

- P50 submitted September 25th, 2013
- Funded June 15th, 2014
- Abe moved to UCSD Jan 1st, 2016
- Leah moved to Wake Forest in 2018
- Renewed July 1st, 2019
- P50 in NCE until April 30th, 2025
- P30 funded Aug 1, 2024 to May 31, 2029





Cocaine Biobank
Oxycodone Biobank
Alcohol Biobank

Core B: HS
breeding
(WFU)

**Total sample size
is more than
15,000 rats**

Research Project 1:
Intermittent access
(Meyer, U Buffalo)

Research Project 2:
Nicotine Self-admin
(Chen, UTHSC)

Research Project 3:
Behavioral regulation
(Dietz, RIA)

U01: Compulsive
Cocaine Intake
(George, Palmer UCSD)

U01 Compulsive
Oxycodone Intake
(George, Palmer UCSD)

U01 Avoidance learning
in addiction (Jhou
MUSC)

U01 Heroine Self
Administration (Kalivas
et al, MUSC)

U01 Delay discounting
(Mitchell, Hitzemann
OHSU)

R01 Alcohol (Palmer,
Im, de Guglielmo,
UCSD)

R01 Bone Matrix
Composition and
Quality (Doug Adams;
U Colorado, Denver)

R01 Genetic
Modulators of
Glaucoma (Jablonski,
UTHSC)

Core C:
Genotyping,
database, GWAS,
eQTL, (Palmer,
UCSD)

Research Project 4:
Network Analysis
(Ideker, UCSD)

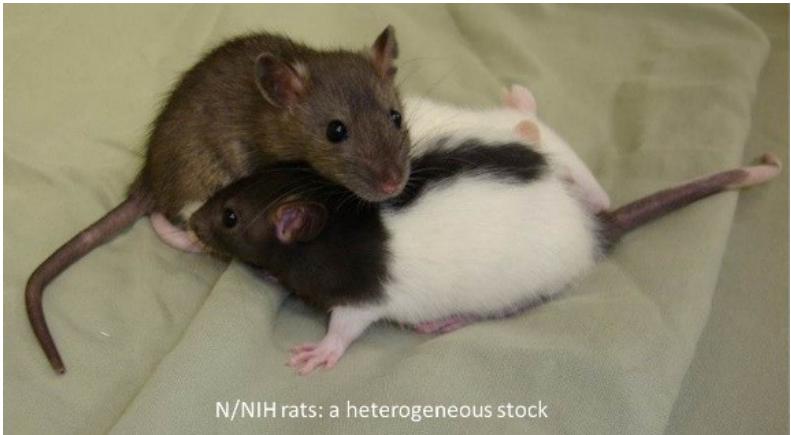
U01 SV and TRs in
mice and rats
(Palmer, Sebat and
Gymrek UCSD)

U01 Single Cell
RNASeq in Oxycodone
Exposed rats
(Telese, UCSD)

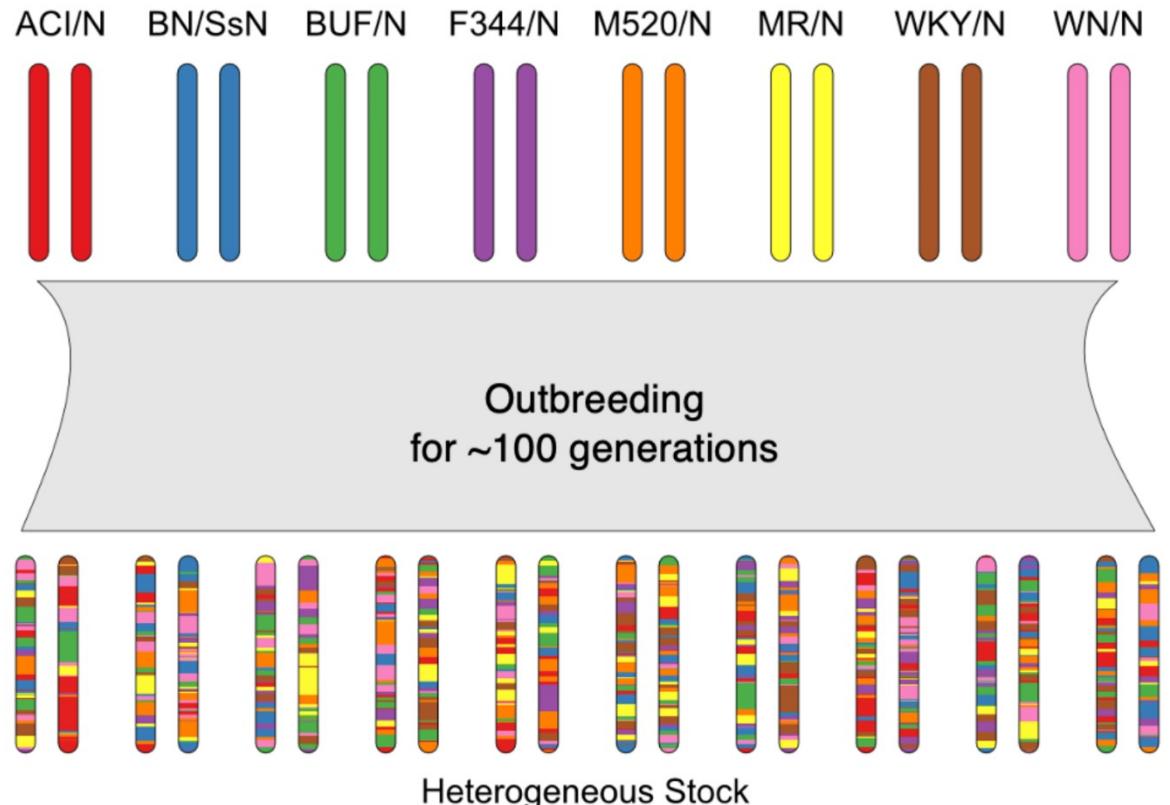
Physiological and other collaborations

	Collaborator	Institution	Area of Interest	Tissue	Grant
1	Arimantas Lionikas	University of Aberdeen, UK	muscle-tendon-bone	hind limb	None
2	Diethard Tautz	Max Planck Institute, Germany	evolution	heads	None
3	Cheryl Ackert-Bicknell Douglas Adams	University of Colorado, Denver	musculoskeletal research	carcass, limbs	Funded R01
4	Kevin Regner & Leah Solberg-Woods	Wake Forest University, US Medical College of Wisconsin	type 2 diabetes	serum, fat, liver, muscle	Funded State of Wisconsin Grant
5	Amelie Baud, Rob Knight	UCSD, Centre for Genomic Regulation	Gut Microbiome	cecum, fecal material	Funded pilot from CMI
6	Joel Leal & Pieter Dorrestein	UCSD	Gut Metabolome	cecum	Funded pilot from CMI
7	Matthew Dean	University of South California, US	evolutionary biology	baculum	None
8	Amelie Baud	University of Barcelona, Spain	indirect genetic effects	cage mate info	None
9	Francesca Telese	UC San Diego, US	epigenetics	brain tissue	Funded U01
10	Derek Daniels	University at Buffalo, US	feeding behaviors	food and water consumptions	None
11	Monica Jablonski	University of Tennessee, US	retinal biology	eyes	Funded R01
12	Chris Vulpe	University of Florida, US	copper and iron metab	liver, serum	None
13	Michael R. Garrett	University of Mississippi, US	Renal physiology	kidney	None
14	Joe Nadeau	Maine Med Cntr Res Institute, US	noise/signal structure	phenotypic data	Funded R01

The center is built around the HS rat

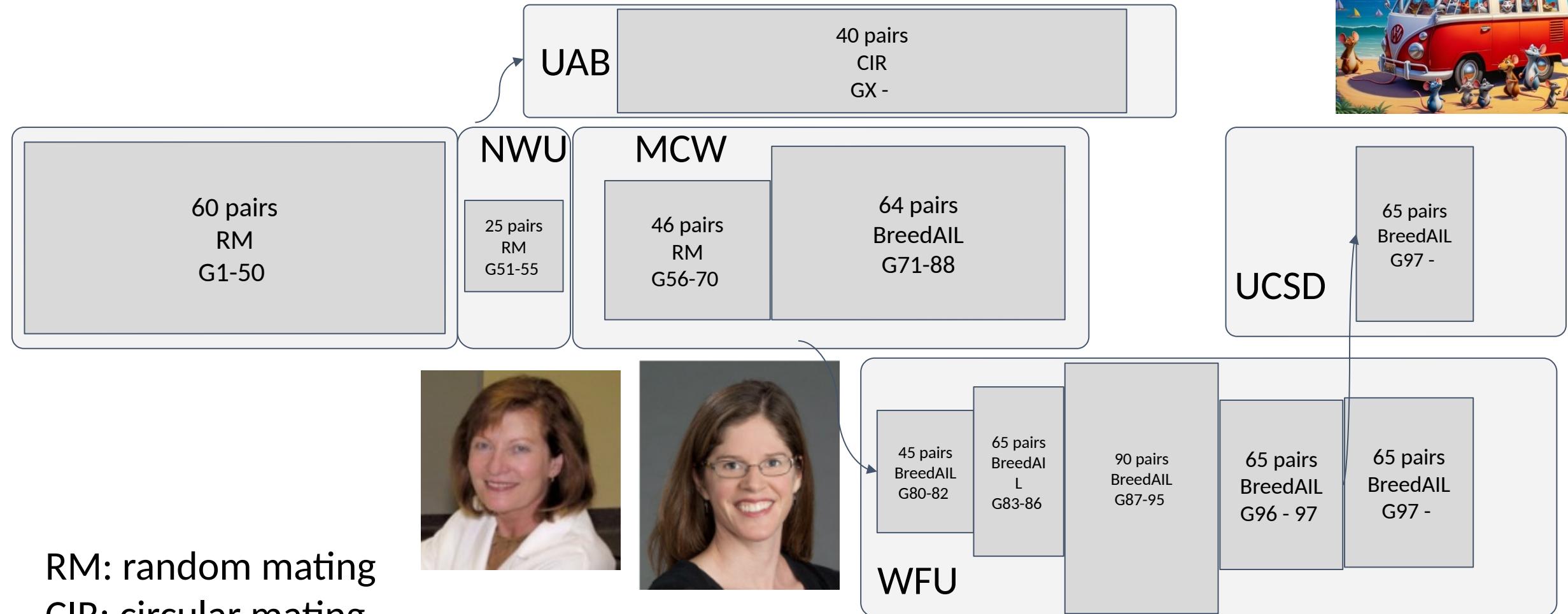


Leah Solberg Woods, PhD
Wake Forest University
School of Medicine



Happy 100th
Birthday!

What a long, strange trip its been

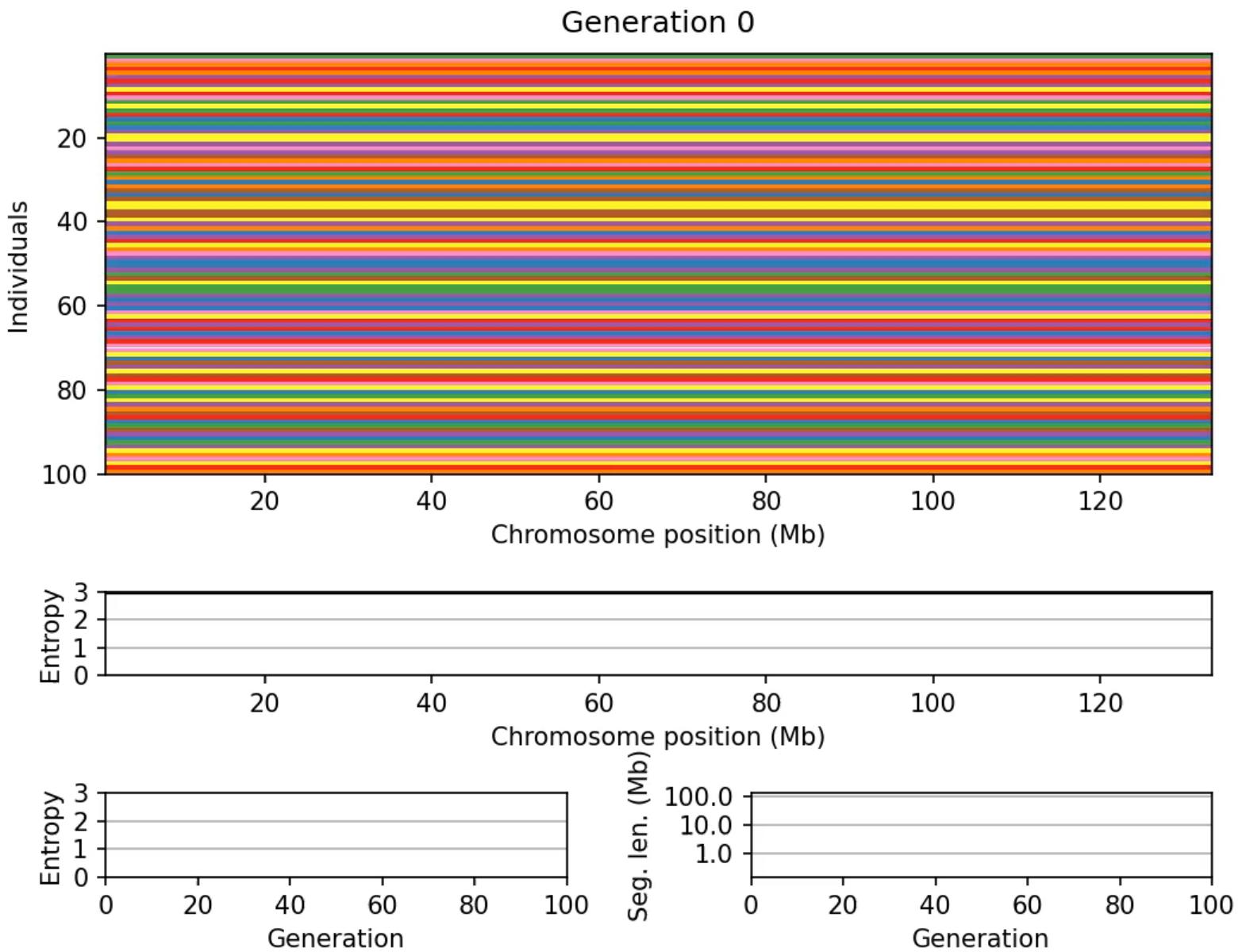


RM: random mating

CIR: circular mating

BreedAIL: max genetic distance mating





Sequencing of the 8 inbred founders of the HS

	Illumina WGS	PacBio HiFi	ONP ultralong
ACI/N	40.9x	39.7x	32.1 x
BN/SsN	37.0x	41.1x	
BUF/N	40.4x	40.2x	
F344/N	51.5x	39.8x	21.3 x
M520/N	40.5x	41.8x	25.8 x
MR/N	37.1x	39.9x	
WKY/N	42.8x	45.3x	28.1 x
WN/N	44.3x	40.6x	26.0 x



Denghui (Den)
Chen



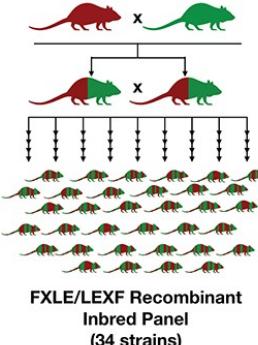
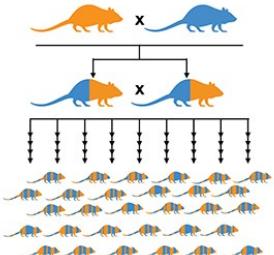
Jonathan Sebat



Melissa Gymrek

Availability of HS founders

HS founders	HRDP best match (MCW)	Genetic Similaratiy	Cost per rat (US\$)
ACI/N	ACI/EurMcwi	0.997	\$112.12
BN/N	BN/NHsdMcwi	0.999	\$112.12
BUF/N	BUF/Mna	0.736	\$112.12
F344/N	F344/DuCrl	0.995	\$112.12
M520/N	M520/NRrrcMcwi	0.997	\$112.12
MR/N	MR/NRrrc	0.998	\$112.12
WKY/N	WKY/NHsd	0.997	\$112.12
WN/N	WAG/RijCrl	0.720	\$112.12



Mindy Dwinell



Hao Chen

To obtain HRDP rats
HRDP@mcw.edu

One essential function of the center is maintaining and distributing the world's only colony of HS rats

Grant or Project	PI	Years	Rats
P50DA046346:NIDA center for genetic studies of drug abuse in outbred rats	Jerry Richards & David Dietz	2014-2023	3313
	Shelly Flagel	2014-2019	1652
	Hao Chen	2014-2024	5890
	Paul Meyer	2019-2024	1874
	Paul Meyer	2019	40
	Brittany Kuhn	2023	100
	Sade Spencer	2023	100
Pilot Projects for P50DA046346:NIDA center for genetic studies of drug abuse in outbred rats	Aron Geurts	2022-2023	67
	Giordano de Guglielmo	2021	64
	Matt Lattal	2023	64
	Hao Chen	2019-2020	70
	Francesca Telese	2019-2022	352
	Gregory Collins	2022	32
	Giordano de Guglielmo	2023	8
	Marsida Kallupi	2023	28
P50DA046346:Determining the role of adolescent sleep and circadian factors on risk for substance use in a rat model	Mary Torregrossa	2020-2023	440
U01DA043799:Identification of Genetic Variants that Contribute to Compulsive Cocaine Intake in Rats	Olivier George	2017-2024	1808
U01DA044451:Use of Next-Gen Sequencing to Identify Genetic Variants that Influence compulsive Oxycodone Intake in Outbred Rats	Olivier George	2019-2024	1622
U01DA044468:Genomic Analysis of Avoidance Learning in Addiction	Tom Jhou	2018-2023	1288
U01DA045300:The Genetic Basis of Opioid Dependence Vulnerability in a Rodent Model	Peter Kalivas	2018-2023	720
U01DA045300:The Genetic Basis of Opioid Dependence Vulnerability in a Rodent Model	Roberto Ciccocioppo	2018-2023	715
U01DA046077:Identification of genetic features of delay discounting using a heterogeneous stock rat model	Suzanne Mitchell	2018-2023	600
R01AA029688:A Framework for Translating Polygenic Findings Related to Alcohol Use Disorder Across Species	Giordano de Guglielmo	2022-2024	647
R21DA045146:Identification of Neurochemical Antecedents and Consequences of Distinct Learning Processes Relevant to Addiction Liability	Shelly Flagel	2018-2021	100
Fee For Service	Marcus Meinhardt	2021	282
Fee For Service	Anne Kwitek	2019	50
Fee For Service	Amelie Baud	2023	32
P30 Pilot project	Jared Young	2024	100
Total			~22,000 HS rats shipped

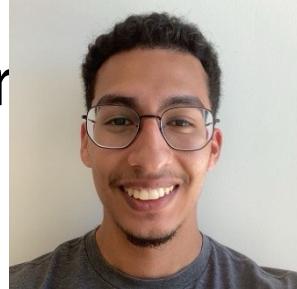
HS West --- we've got rats in San Diego!



- In the first 5 years, we did not have a single problem shipping rats
- Starting in ~2019, several shipments were mishandled by shippers
- Leah has increasingly shipped by ground instead of air
- Shipments from WFU to UCSD can't go via ground...
- There are now 3 GWAS projects at UCSD that use HS rats
- This led us to start HS West
 - Both WFU and HSWest maintain colonies
 - We just exchanged some breeders to reduce divergence
- Leah will discuss more in her Core B presentation



Yizhi Wang



Rudy Avila



Nina Suzuki

Another essential function of the center is to provide a centralized database and genetic analysis pipeline

Database/bioinformatics team

- Apurva started the database, I had originally budgeted 50% effort!
- The database has become one of the most valuable but also most labor-intensive parts of the center
 - Supports both projects, U01s and other collaborative grants and projects



Thiago Sanches



Ben Johnson



Elaine Keung



Gavriela ("Gavi") Ang

Database

- We will continue to use FAIR practices that were already established.
- RatGenes.org portal
- RatGTE.org portal

Database

- Intensive data cleaning/QC – Rat Viewer (see Montana's talk)
- Datasets are permanently stored at [UC Library Digital Collection](#).
- Raw reads are deposited to [SRA](#).
- Computation pipelines available on [GitHub](#);
- DOIs are generated for computational pipelines
- HS rats colonies are registered with [RGD](#); RRIDs are generated
- Use [protocols.io](#) for all protocols that were used to generate data.
- NEW: Internet Rat Server ([IRS](#)) - a cloud-hosted portal to access Center's HS rats database, including an archive of reports and data search tools

The regulatory landscape of multiple brain regions in outbred heterogeneous stock rats

Daniel Munro ^{ID}^{1,2}, Tengfei Wang ^{ID}³, Apurva S. Chitre ^{ID}¹, Oksana Polesskaya ^{ID}¹,
Nava Ehsan ^{ID}², Jianjun Gao¹, Alexander Gusev ^{ID}⁴, Leah C. Solberg Woods ^{ID}⁵,
Laura M. Saba ^{ID}⁶, Hao Chen ^{ID}³, Abraham A. Palmer ^{ID}^{1,7,*} and Pejman Mohammadi ^{ID}^{2,8,*}

¹Department of Psychiatry, University of California San Diego, La Jolla, CA, USA, ²Department of Integrative Structural and Computational Biology, Scripps Research, La Jolla, CA, USA, ³Department of Pharmacology, Addiction Science and Toxicology, University of Tennessee Health Science Center, Memphis, TN, USA, ⁴Division of Population Sciences, Dana-Farber Cancer Institute and Harvard Medical School, Boston, MA, USA, ⁵Section of Molecular Medicine, Department of Internal Medicine, Wake Forest University School of Medicine, Winston-Salem, NC, USA, ⁶Department of Pharmaceutical Sciences, Skaggs School of Pharmacy and Pharmaceutical Sciences, University of Colorado Anschutz Medical Campus, Aurora, CO, USA, ⁷Institute for Genomic Medicine, University of California San Diego, La Jolla, CA, USA and ⁸Scripps Research Translational Institute, Scripps Research, La Jolla, CA, USA

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Dan Munro



Hao Chen



Laura Saba



Pejman
Mohammadi

Project	Investigator	Tissue	Sample size
P50 DA037844 Y1-5, Project 2: Socially-acquired nicotine self-administration	Hao Chen, UTHSC	nucleus accumbens core	88
P50 DA037844 Y1-5, Project 2: Socially-acquired nicotine self-administration	Hao Chen, UTHSC	lateral habenula	88
P50 DA037844 Y1-5, Project 2: Socially-acquired nicotine self-administration	Hao Chen, UTHSC	orbitofrontal cortex	88
P50 DA037844 Y1-5, Project 2: Socially-acquired nicotine self-administration	Hao Chen, UTHSC	infralimbic cortex	88
P50 DA037844 Y1-5, Project 2: Socially-acquired nicotine self-administration	Hao Chen, UTHSC	prelimbic cortex	88
P50 DA037844 Y5-10, Project 1: Neurogenetic Substrates of Cocaine Addiction	Paul Meyer, U at Buffalo	nucleus accumbens, core	180
P50 DA037844 Y5-10, Project 1: Neurogenetic Substrates of Cocaine Addiction	Paul Meyer, U at Buffalo	insular cortex	180
P50 DA037844 Y5-10, Project 1: Neurogenetic Substrates of Cocaine Addiction	Paul Meyer, U at Buffalo	posterior ventral tegmental area	180
P50 DA037844 Y5-10, Project 2: Socially-acquired nicotine self-administration	Hao Chen, UTHSC	posterior ventral tegmental area	200
P50 DA037844 Y5-10, Project 2: Socially-acquired nicotine self-administration	Hao Chen, UTHSC	prelimbic cortex	200
P50 DA037844 Y5-10, Project 2: Socially-acquired nicotine self-administration	Hao Chen, UTHSC	nucleus accumbens, core	200
U01 DA050239:Single-cell Resolution analysis of chromatin accessibility and gene expression changes in a model of drug addiction	Francesca Telese, UCSD	nucleus accumbens	200
Pilot: Creating the dataset for TWAS in HS rats	Francesca Telese, UCSD	whole hemisphere	339
U01DA046077: Identification of Genetic Features of Delay Discounting Using a Heterogeneous Stock Rat Model	Suzanne Mitchell, OSHU	nucleus accumbens core	200
U01DA046077: Identification of Genetic Features of Delay Discounting Using a Heterogeneous Stock Rat Model	Suzanne Mitchell, OSHU	basolateral amygdala	200
U01DA046077: Identification of Genetic Features of Delay Discounting Using a Heterogeneous Stock Rat Model	Suzanne Mitchell, OSHU	infralimbic cortex	200
R01 EY021200: Genetic Modulators of Glaucoma	Monica Jablonsky, UTHSC	Eye	50
R01DK120667: Systems genetics identify neuronal genes for diet-induced obesity	Leah Solberg Woods, WFU	Adipose	400
R01DK120667: Systems genetics identify neuronal genes for diet-induced obesity	Leah Solberg Woods, WFU	Liver	400
R01DK120667: Systems genetics identify neuronal genes for diet-induced obesity	Leah Solberg Woods, WFU	Hippocampus	500
R01DK120667: Systems genetics identify neuronal genes for diet-induced obesity	Leah Solberg Woods, WFU	Hypothalamus	500

RatGTEx Portal

This portal provides gene expression, eQTL, and sQTL data for multiple rat tissues. It is managed by the [NIDA center for genetic studies of drug abuse in outbred rats \(P50DA037844\)](#).

[RRID:SCR_022145](#)

Tissues

The raw data for the tissues in this portal come from multiple studies. The processed data, however, may differ from the results reported in each study for a couple reasons:

1. Different processing decisions could be made for each study, while the data in this portal are processed uniformly to facilitate comparison across tissues.
2. The published study results are immutable, while new versions of data in this portal may be released, for example to improve quality or to use updated reference genome and annotations.

Nevertheless, we also host the original results from individual studies on the [Download](#) page when available.

Tissue	Abbreviation	Samples (rn6)	Samples (rn7)	Originating study
Infralimbic cortex	IL	81	83	P50 DA037844 Y1-5, Project 2
Lateral habenula	LHb	81	82	P50 DA037844 Y1-5, Project 2
Nucleus accumbens core	NAcc	75	77	P50 DA037844 Y1-5, Project 2
Orbitofrontal cortex	OFC	80	82	P50 DA037844 Y1-5, Project 2
Prelimbic cortex	PL	80	82	P50 DA037844 Y1-5, Project 2
Eye	Eye	51	53	R01 EY021200
Basolateral amygdala	BLA	185	191	U01DA046077
Nucleus accumbens core	NAcc2	188	193	U01DA046077
Prelimbic cortex	PL2	191	194	U01DA046077
Adipose	Adipose	411	-	R01DK106386
Liver	Liver	401	-	R01DK106386
Brain hemisphere	Brain	339	340	Pilot: Creating the dataset for TWAS in HS rats

Sample sizes for rn6 and rn7 versions differ due to availability of genotypes.

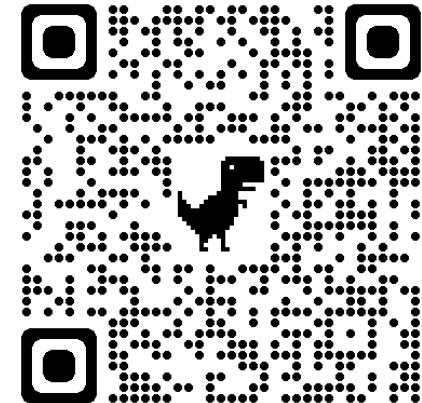
Data Use

For the tissues above whose originating study has been published (see below), you may use the data in your studies by citing the originating study. For the other tissues, contact us about whether, when, and how you may use them.

Associated Studies

More info on the [ratgenes.org eQTL page](#). Additional publications will be posted when available.

Tissues from the same study are from the same set of rats, though the final subsets per tissue differ slightly after quality control filtering. See the [sample info page](#) for details.



Another essential function of the center is to provide an inexpensive, high-throughput method for genotyping HS rats

Genotyping



Khai-Minh Nguyen

- In the early P50 projects performed (~2014-2019) we used genotyping by sequencing (GBS)
- Starting with U01s that were funded ~2017, we began using a newer method that we call “riptide”
- This created some problems for the joint analyses across projects
- Den and Ben have developed a ‘hybrid genotyping pipeline’ that uses the raw sequencing reads from both platforms; the results are very similar to prior iterations for both pipelines



Ben Johnson

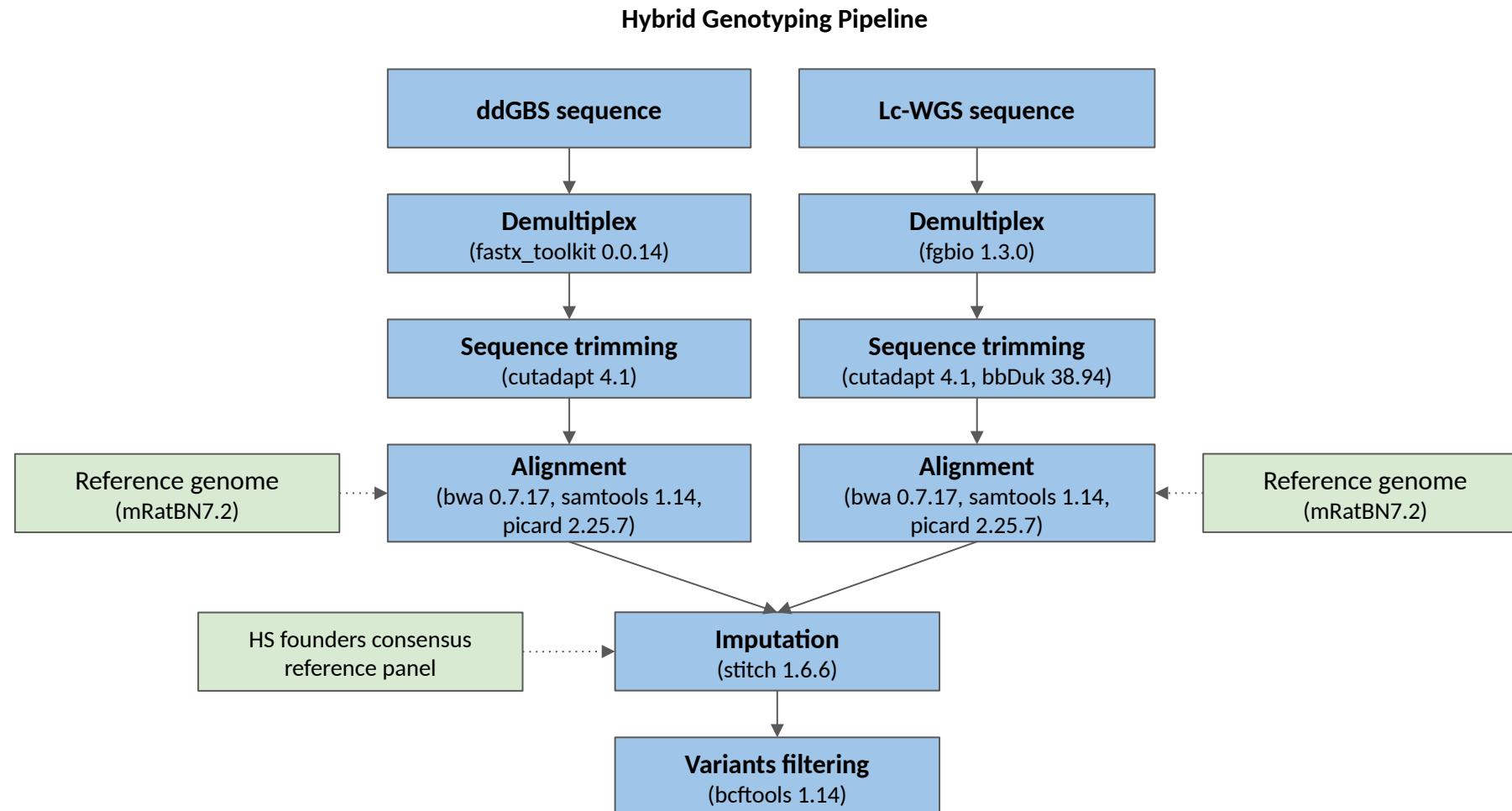


Denghui (“Den”) Chen



Gavrila (“Gavi”) Ang

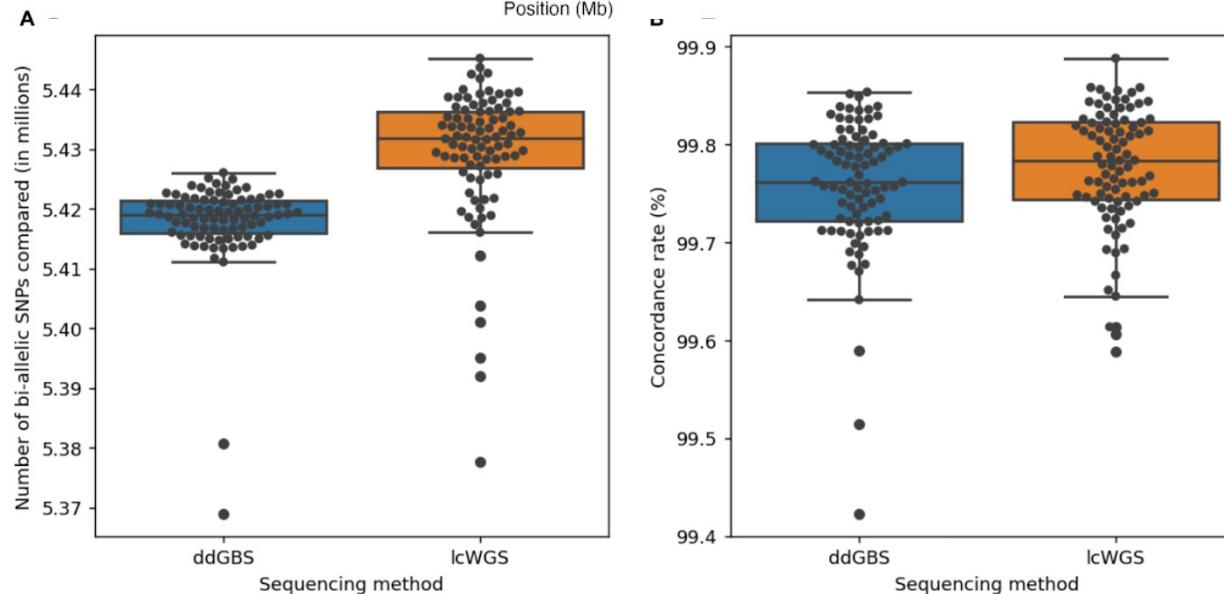
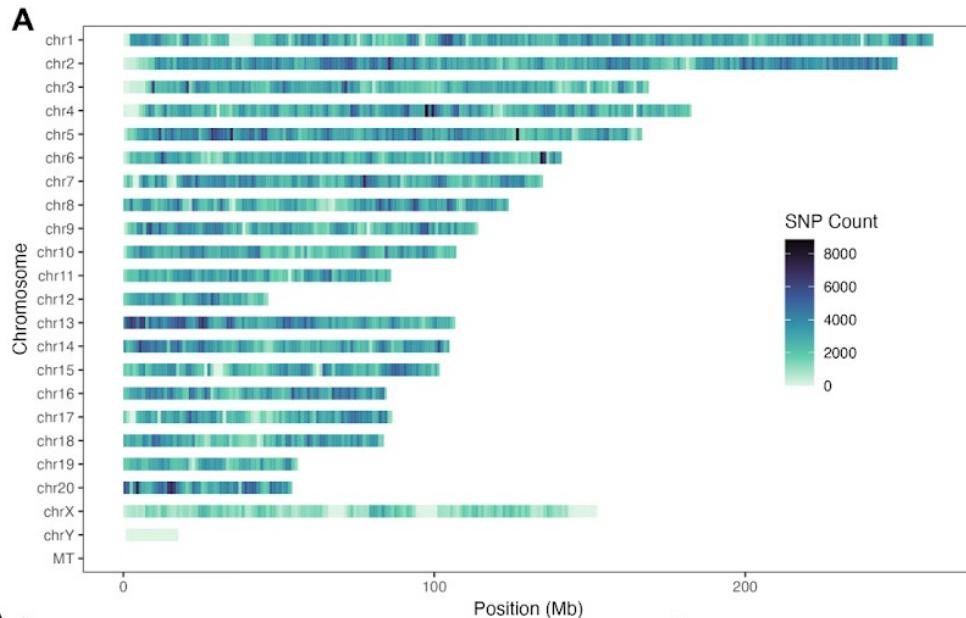
Hybrid Genotyping Pipeline Overview



Most recent set of genotypes (round 10.4)

- # SNPs post QC filtering: 7,348,512
- # rats before filtering: 20,031
- # rats after QC filtering: 19,117

Genotyping paper under review at G3



bioRxiv preprint doi: <https://doi.org/10.1101/2024.07.17.603984>; this version posted July 18, 2024. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY-NC-ND 4.0 International license.

A Cost-effective, High-throughput, Highly Accurate Genotyping Method for Outbred Populations

Denghui Chen¹, Apurva S. Chitre¹, Khai-Minh H. Nguyen², Katarina Cohen², Beverly Peng², Kendra S. Ziegler², Faith Okamoto², Bonnie Lin², Benjamin B. Johnson², Thiago M. Sanches², Riyan Cheng², Oksana Polesskaya², Abraham A. Palmer^{2, 3*}

¹Bioinformatics and System Biology Program, University of California San Diego, 9500 Gilman Dr, La Jolla, CA 92093

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*Corresponding author Abraham A Palmer (aap@ucsd.edu)

Running head: A Genotyping Method for HS Rats

Keywords: Low-coverage, Whole genome sequencing, Genotyping, Heterogeneous stock rat

The final essential part of
the center is our analysis
pipeline

General Information



Phenotype Info

- n = 874
- phenotype data: [here](#)
- covariate dropboxes: [here](#)
- phenotype statistical descriptions file: [here](#)

Genotype Info

- genotypes version: 10.2.1
- gwas pipeline version: v0.2.0-11-g990844d
- number of snps: before filter 7358643, after filter 5424892
- genotype missing rate filter: < 0.1 (23054 snps removed)
- minor allele frequency filter: > 0.005 (1908680 snps removed)

► Trait Descriptions

► Preprocessing

► Genomic PCA

► Genetic Correlation

► Heritability

► QTL

► Porcupine Plot

► Manhattan Plots

► Regional Association Plots

► Gene Enrichment

► FAQ

► References

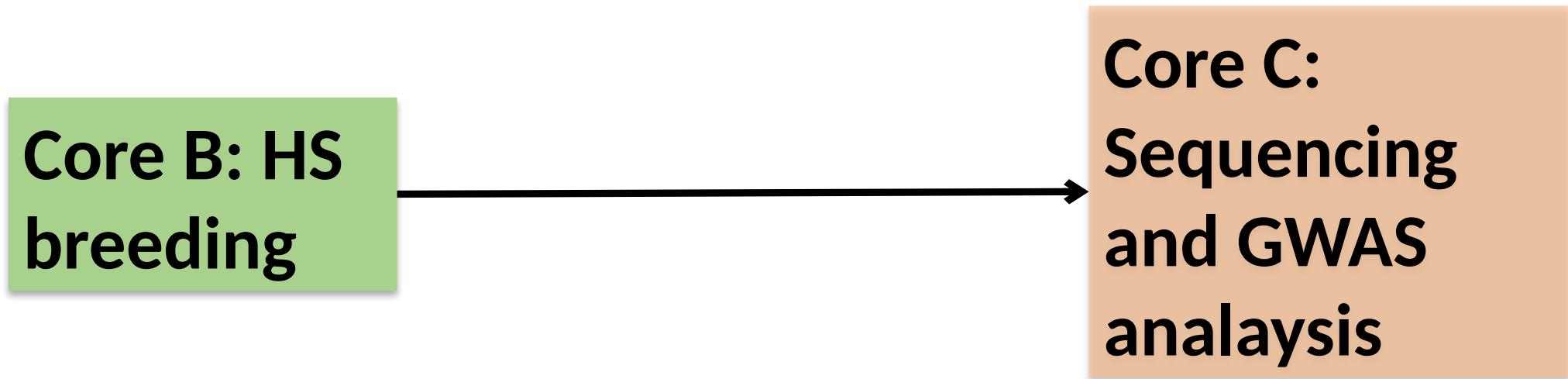


Apurva Chitre



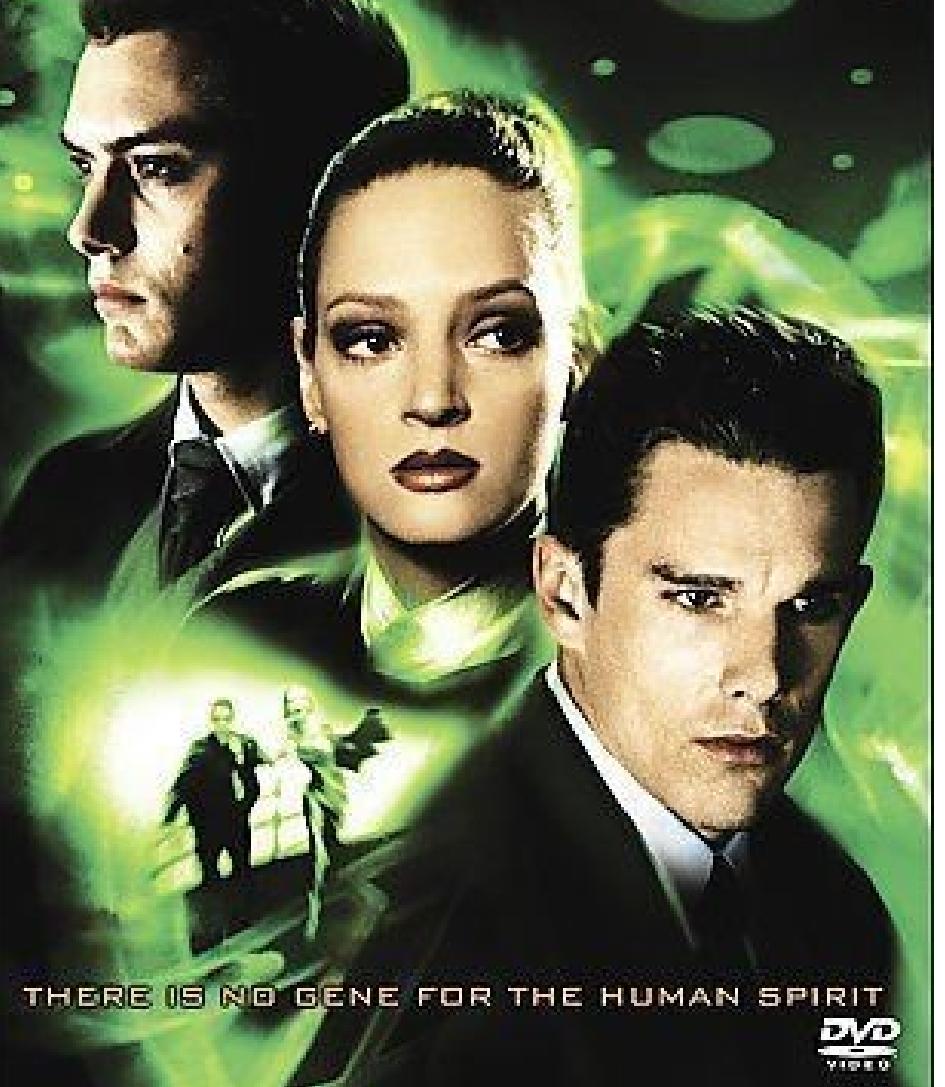
Thiago Sanches

These ability to generate HS rats, genotype HS rats and perform GWAS on HS rats allow us to support many different projects



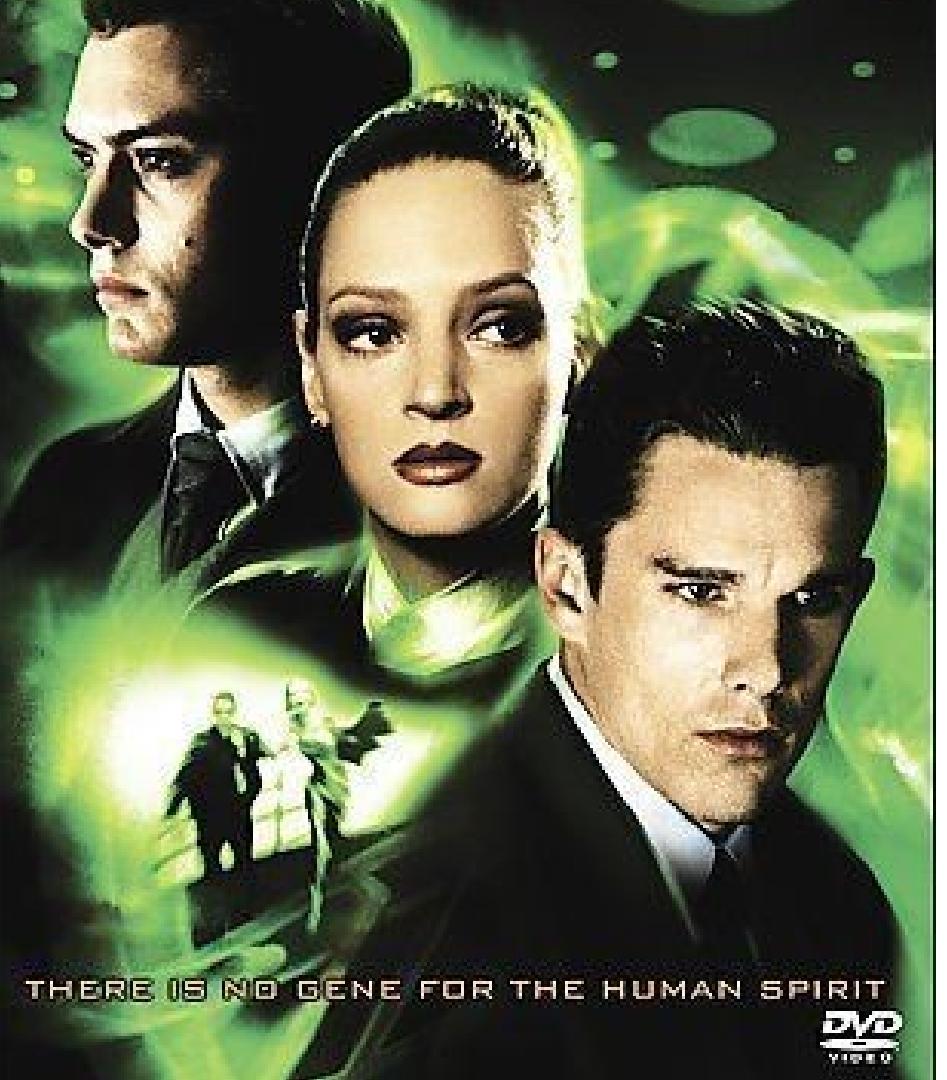
ETHAN HAWKE UMA THURMAN JUDE LAW
FROM ANDREW NICCOL, WRITER OF "THE TRUMAN SHOW"

GATTACA



ETHAN HAWKE UMA THURMAN JUDE LAW
FROM ANDREW NICCOL, WRITER OF "THE TRUMAN SHOW"

GATTACA



THERE IS NO GENE FOR THE HUMAN SPIRIT



ETHAN HAWKE UMA THURMAN JUDE LAW

RATTACA



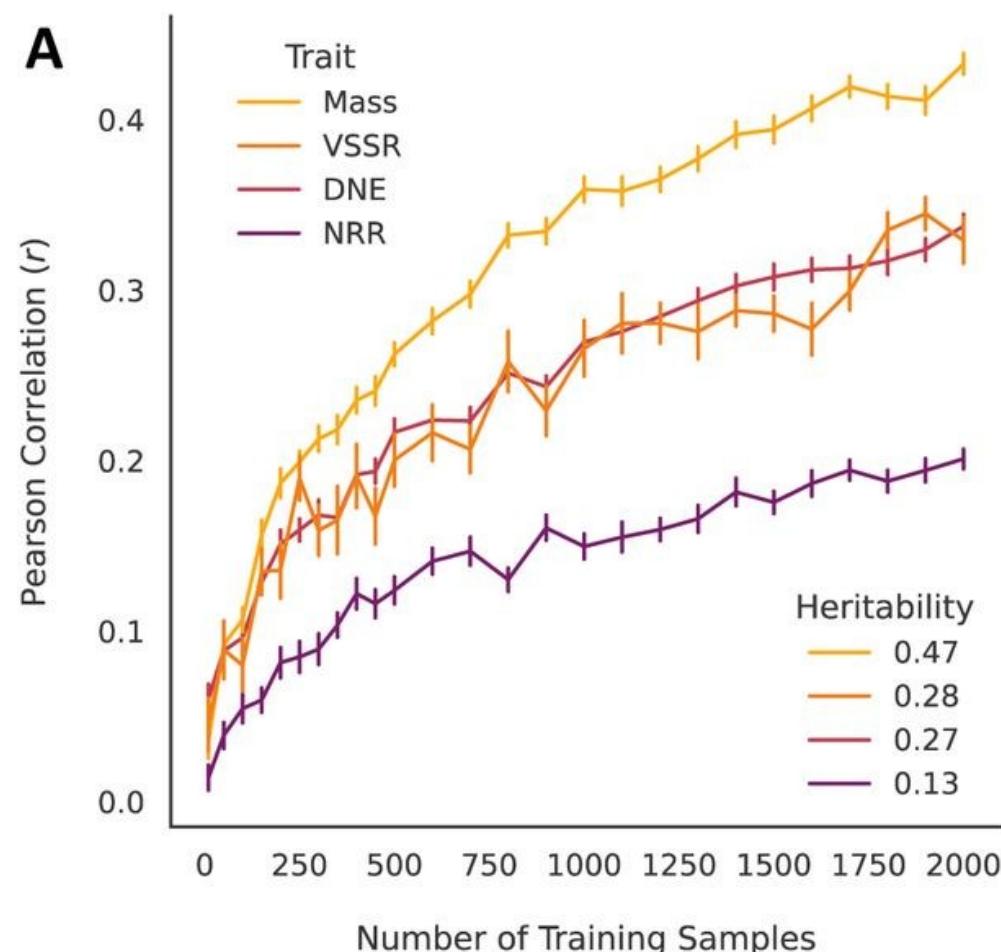
THERE IS NO GENE FOR THE HUMAN SPIRIT

...however we've gotten pretty good at predicting rat behavior.

RATTACA: A new paradigm for examining genetic correlations in outbred rats (Core B)

- We have phenotyped and genotyped more than 20,000 HS rats
- This allows us to use polygenic scores (**PGS**) a.k.a. polygenic risk scores (**PRS**), a.k.a. polygenic index (**PGI**) a.k.a. Genomic Estimated Breeding Values (**GEBV**) to predict phenotypes of newborn rats based only on their genotype

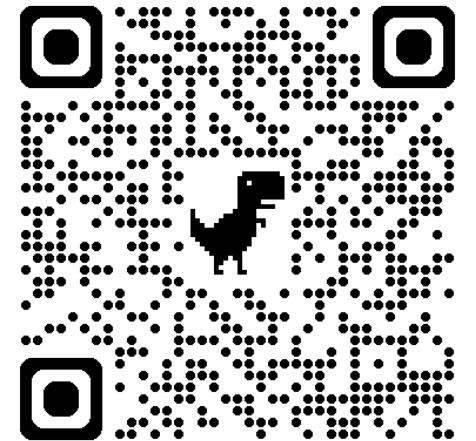
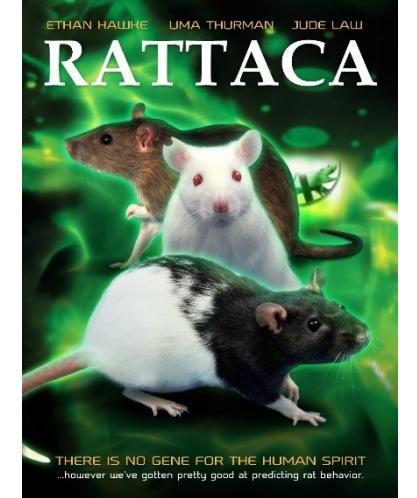
How good are the predictions?



What traits can we predict?

Trait	N	h^2
Locomotor activity	7,895	0.17
Body weight (grams)	5,667	0.44
Pavlovian conditioned approach	3,220	0.22
Choice reaction time	2,453	0.20
Social behavior (time with social partner)	1,844	0.23
Elevated plus maze (time in closed arms)	1,317	0.35
Nicotine, total infusions	1,422	0.21
cocaine, punishment resistance	982	0.30
cocaine, extended access, addiction index	771	0.08
cocaine, intermittent access, infusions	446	0.19
heroin, extended access, infusions	862	0.22
Oxycodone, extended access	530	0.12
Delay discounting	644	0.24

Thanks to the many members of the HS rat community



NIDA Center of Excellence for
Genetics, Genomics, and Epigenetics
of Substance Use Disorders in Outbred Rats