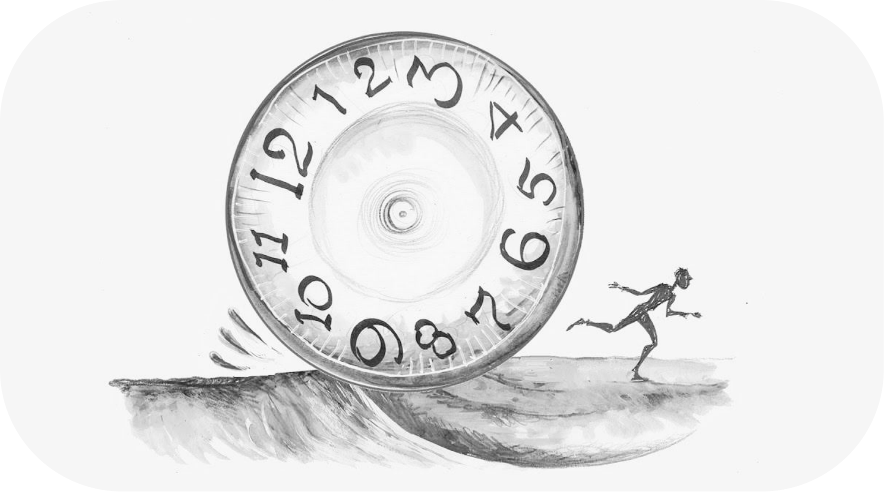
Skoltech

Transcriptomics-Based Screening for Pharmacological Treatments to Defer Aging

Kashuk E., Kubenko K., Vaulin. N.

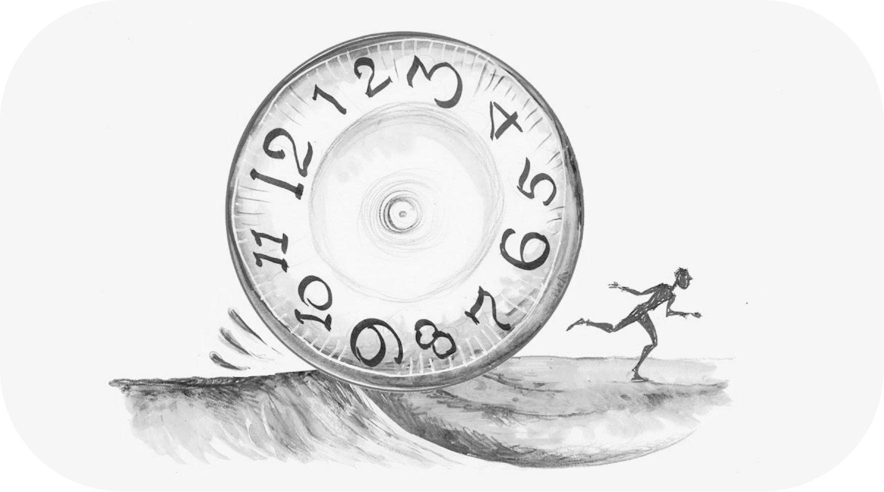
Background

- **Aging is** associated human **morbidity** and mortality and there are no way to revert it

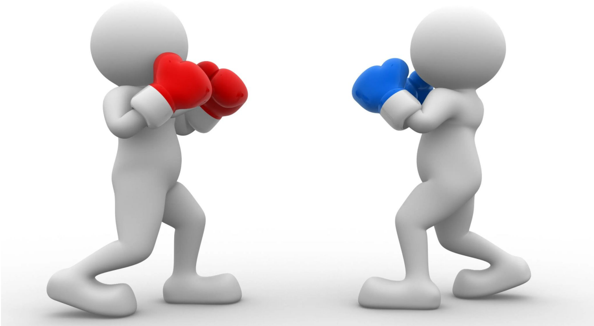


Background

- **Aging is** associated human **morbidity** and mortality and there are no way to revert it
- However, scientists search for treatments that can **slow down aging**



Magic pill **Aging**



Background

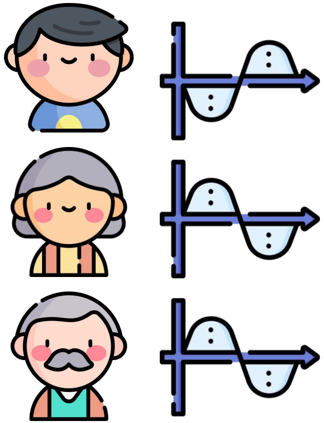
Cell Reports

Article

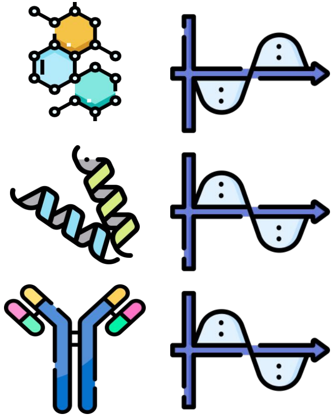
Transcriptomics-Based Screening Identifies Pharmacological Inhibition of Hsp90 as a Means to Defer Aging

2 transcriptomics datasets

Their own RNA-seq
~ Age



Connectivity Map (CMap)
~ Treatment



Janssens, Georges E et al. “Transcriptomics-Based Screening Identifies Pharmacological Inhibition of Hsp90 as a Means to Defer Aging.” Cell reports vol. 27,2 (2019): 467-480.e6. doi:10.1016/j.celrep.2019.03.044

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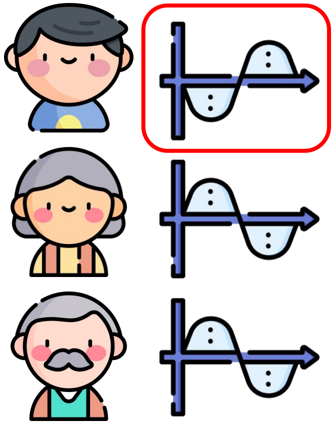
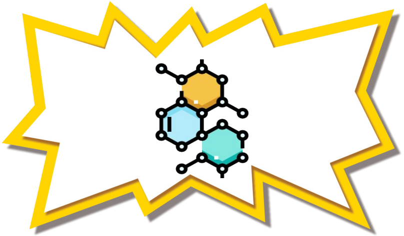
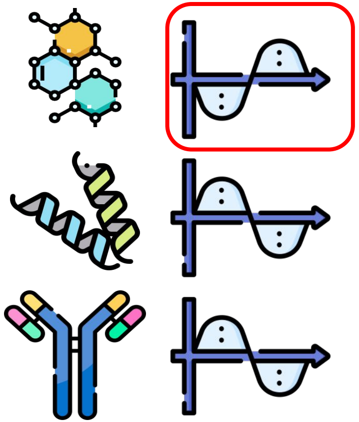


Anti-aging drug





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Spoiler

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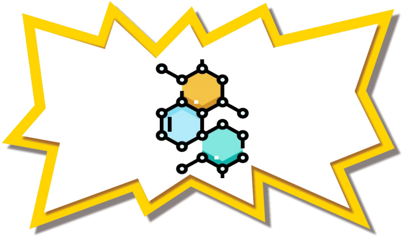
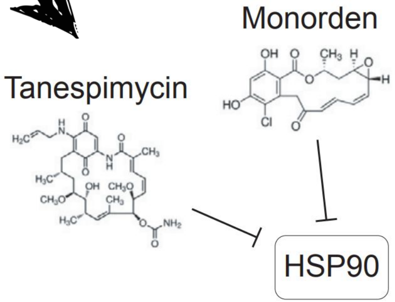
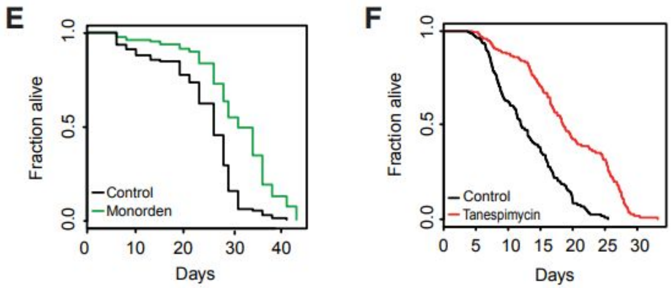




Authors identified several molecules as potential geroprotectors. Most promising of them (Hsp90 inhibitors) were tested in *C.elegans*



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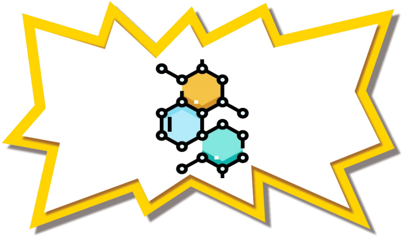

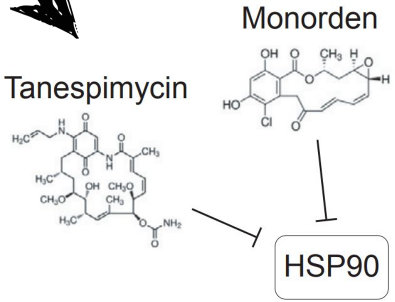

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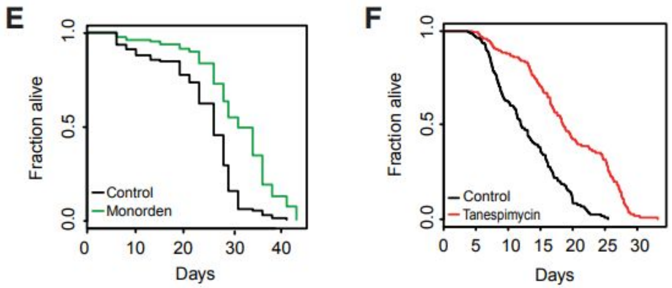
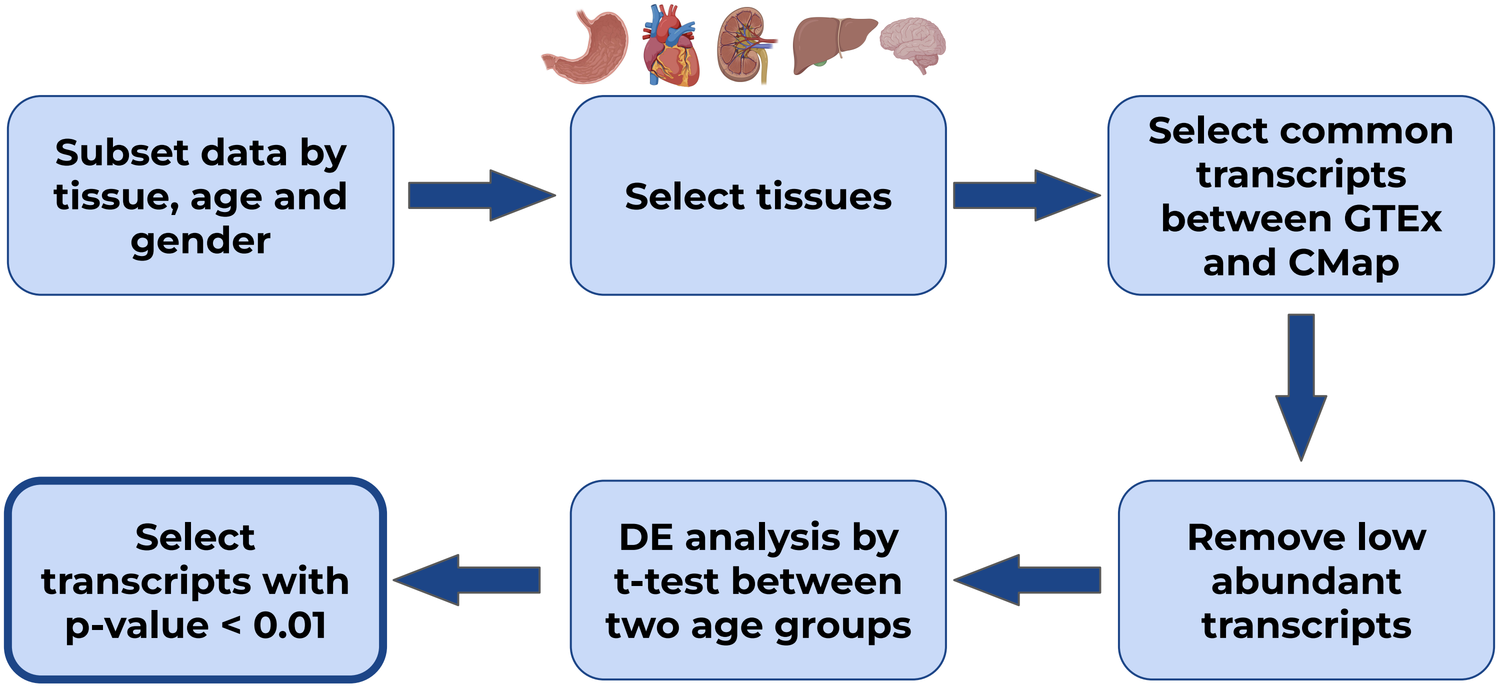


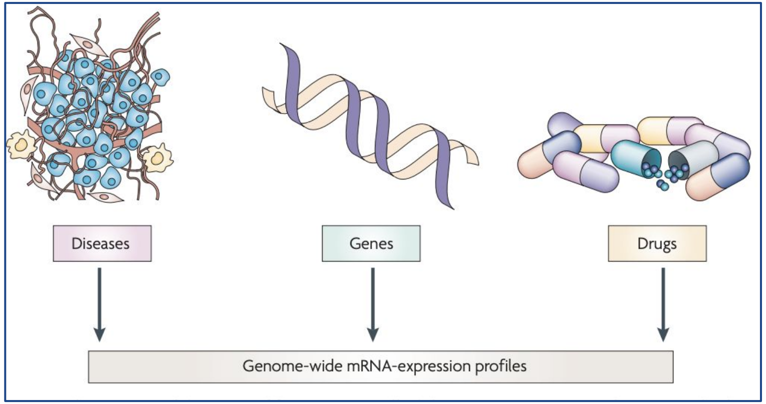
But we gonna do
our own research!

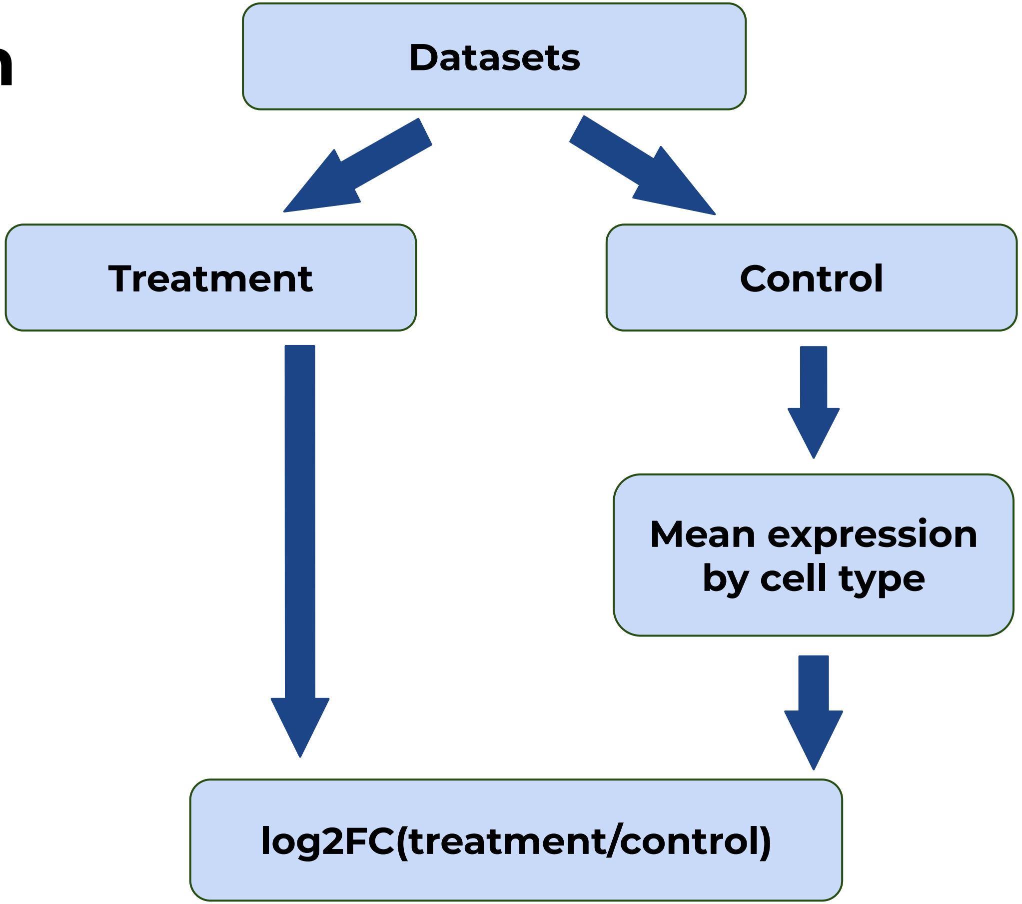
Janssens, Georges E et al. "Transcriptomics-Based Screening Identifies Pharmacological Inhibition of Hsp90 as a Means to Defer Aging." Cell reports vol. 27,2 (2019): 467-480.e6. doi:10.1016/j.celrep.2019.03.044

Data processing

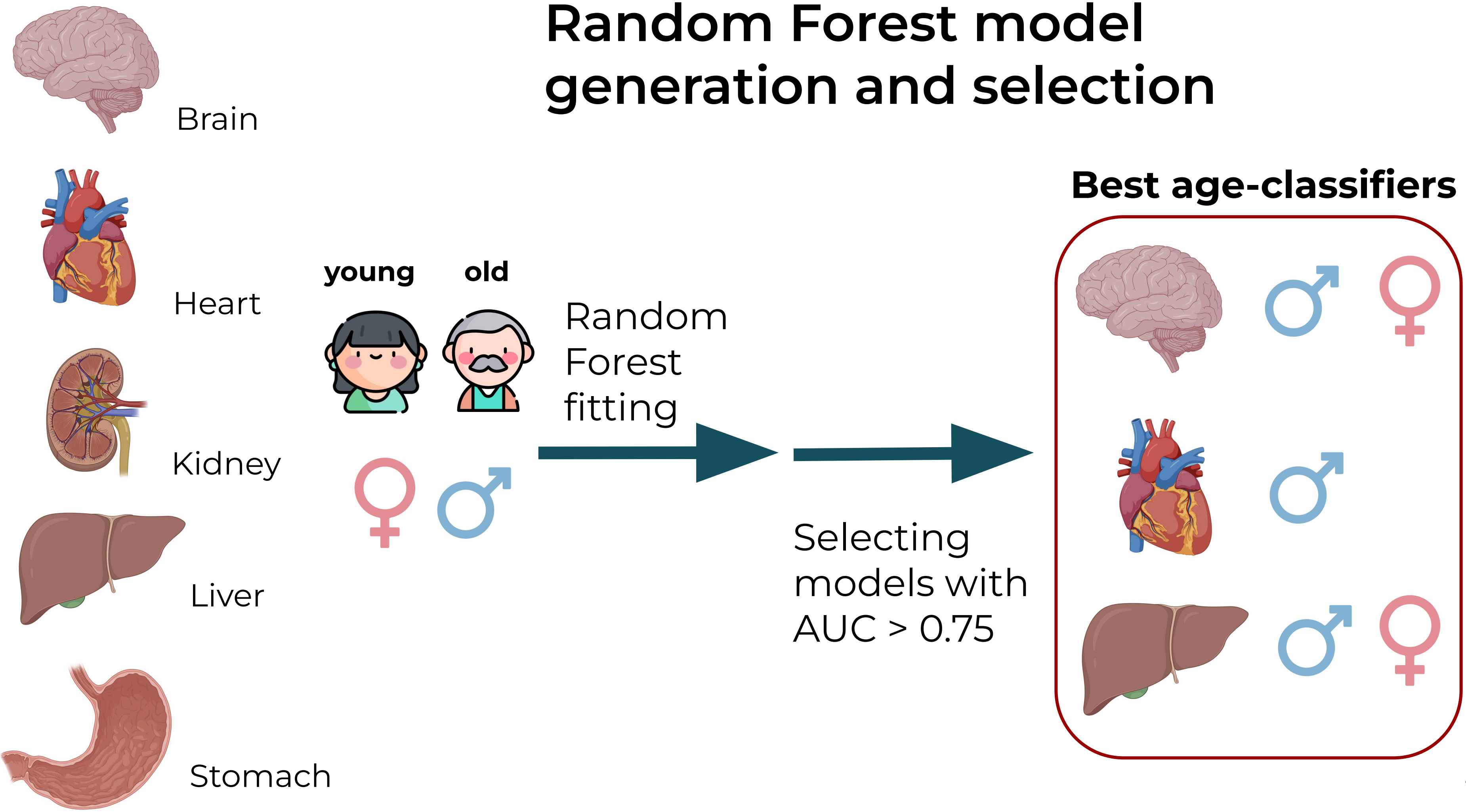


CMap data preparation

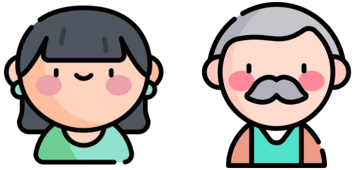




Random Forest model generation and selection



1 Generating ‘drug-induced’ transcriptomes for age classification



Average transcriptome between compared old and young datasets

→ Prototypical ‘middle-aged’ transcriptomes

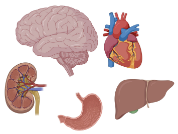
applying CMap fold changes →

‘Drug-induced’ transcriptomes

2 Application of age classifiers to ‘drug-induced’ transcriptomes

→ **Geroprotectors**

The list of geroprotectors



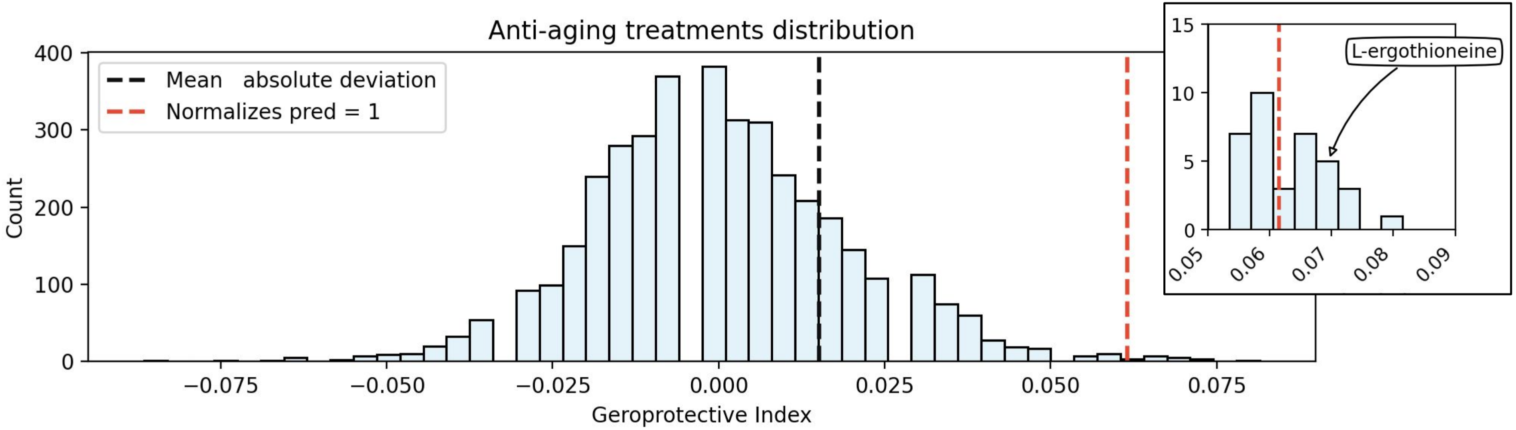
100 treatments
(out of 3546)

L-ergothioneine

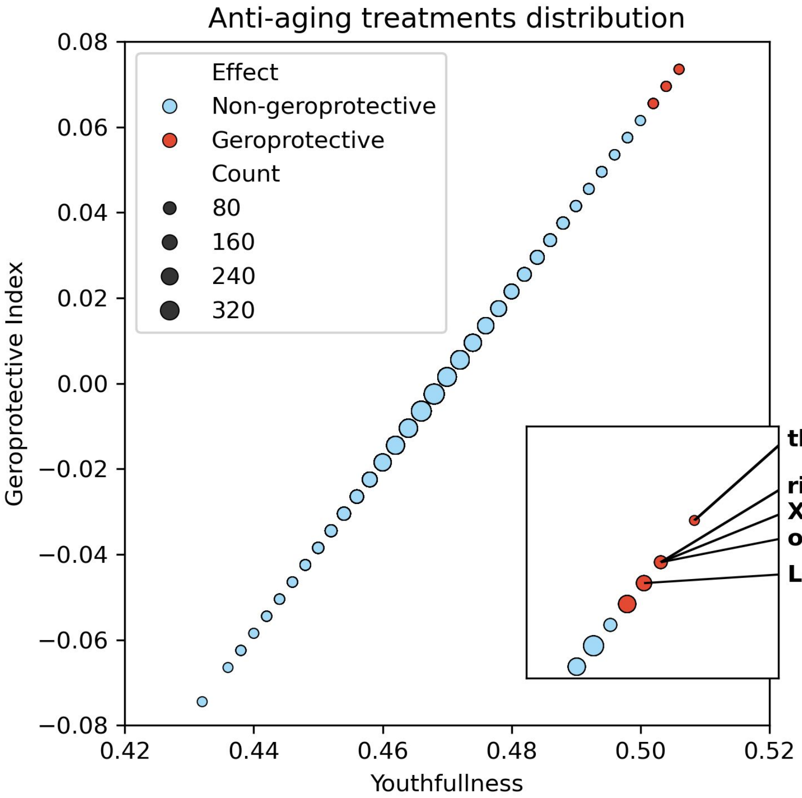
3546 treatments
(out of 3546)

11 compounds

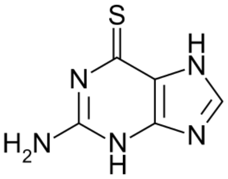
olanzapine
XL-888
ritonavir
thioguanine
L-ergothioneine
nafamostat
ochratoxin-a
etofylline-clofibrate
tebipenem
E-2012
EMF-sumo1-7



The list of geroprotectors



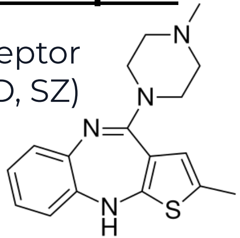
Thioguanine



Incorporates into DNA and **inhibits synthesis**. Used in the treatment of **leukaemia**.

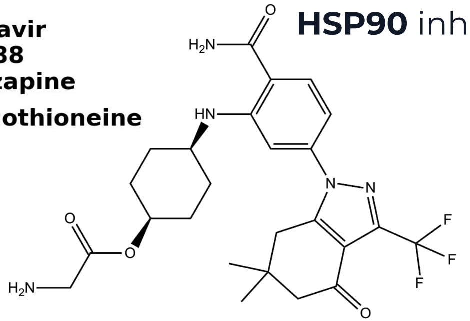
Olanzapine

Serotonin-dopamine-receptor antagonist, **antipsychotic** (BD, SZ)



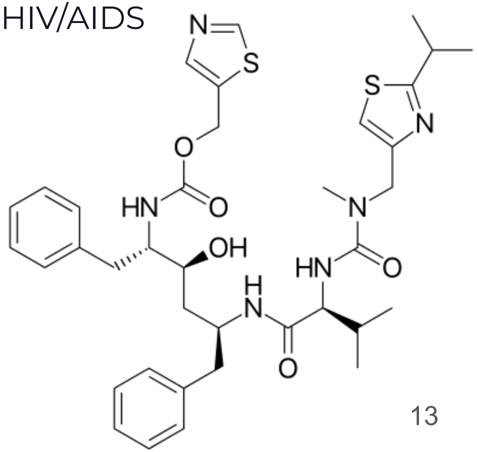
XL-888

HSP90 inhibitor



Ritonavir

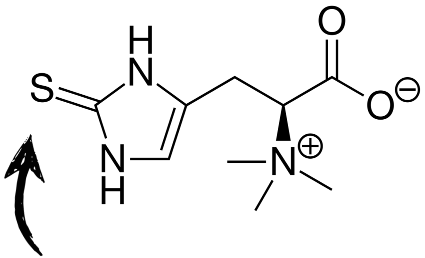
Proteases inhibitor, used for HIV/AIDS



Result validation: L-ergothioneine

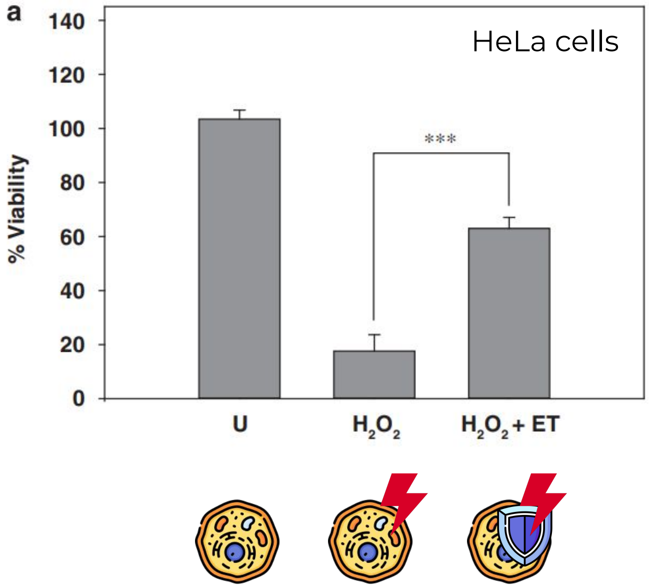
Is **L-ergothioneine** a real geroprotection candidate?

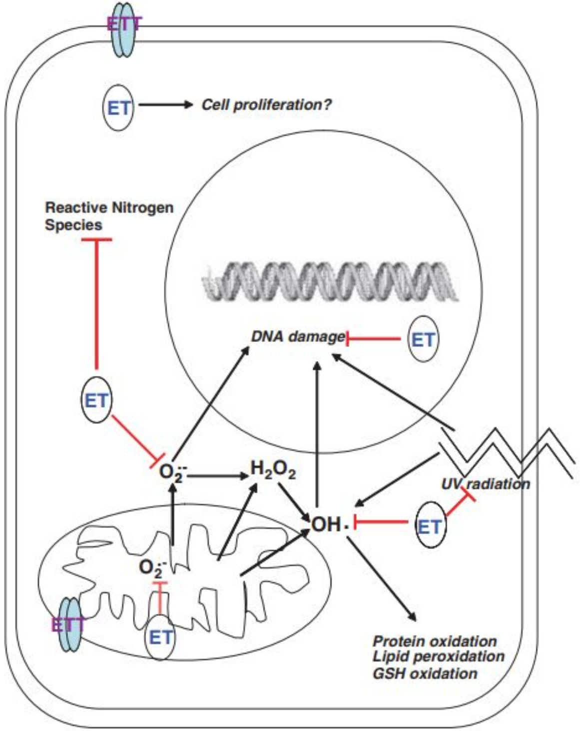
L-ergothioneine function



Sulphur group (can be oxidised and reduced)

May protect against reactive oxygen species





Paul, B D, and S H Snyder. "The unusual amino acid L-ergothioneine is a physiologic cytoprotectant." Cell death and differentiation vol. 17,7 (2010): 1134-40. doi:10.1038/cdd.2009.163

L-ergothioneine in the network of aging hallmarks

Protein Cell, 2023, XX, 1–16
https://doi.org/10.1093/procel/pwad048
Advance access publication 10 August 2023

Ergothioneine and its congeners: anti-ageing mechanisms and pharmacophore biosynthesis

Li Chen[1,2,‡], Liping Zhang[2,‡], Xujun Ye[1,*], Zixin Deng[1,2,*], Changming Zhao[1,2,*]

Antioxidant defense system

Intercellular **transport** (OCTN1)

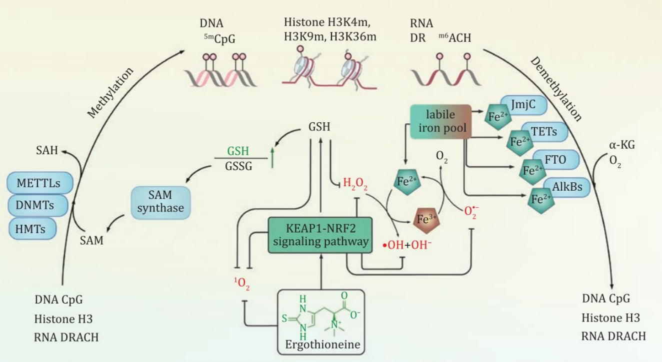
Signaling cascades (KEAP1 -NRF2 pathway)

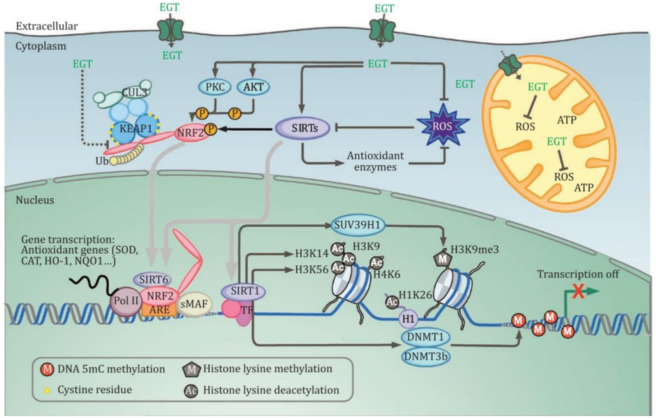
L-ergothioneine

Genome **stability**

Epigenome **stability** (DNA 5mC, sirtuins)

Replication

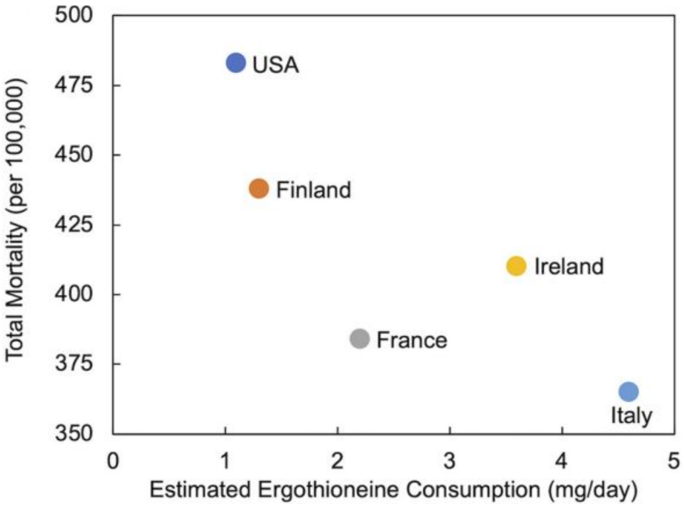


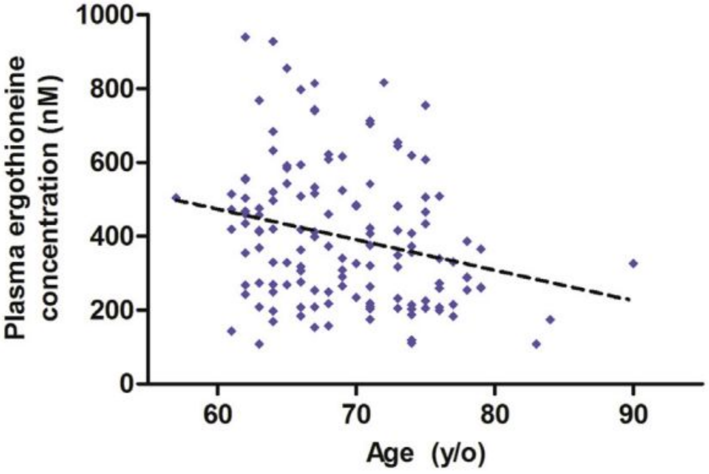


Apparoo, Yasaaswini et al. "Ergothioneine and its prospects as an anti-ageing compound." Experimental gerontology vol. 170 (2022): 111982. doi:10.1016/j.exger.2022.111982

L-ergothioneine in real life

- L-ergothioneine is correlated with mortality and life expectancy
- L-ergothioneine concentration in plasma decreases with aging

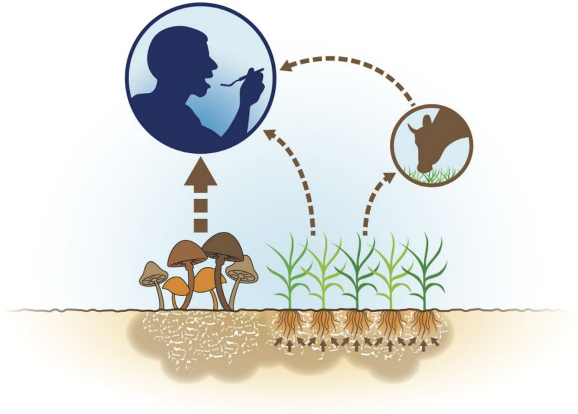


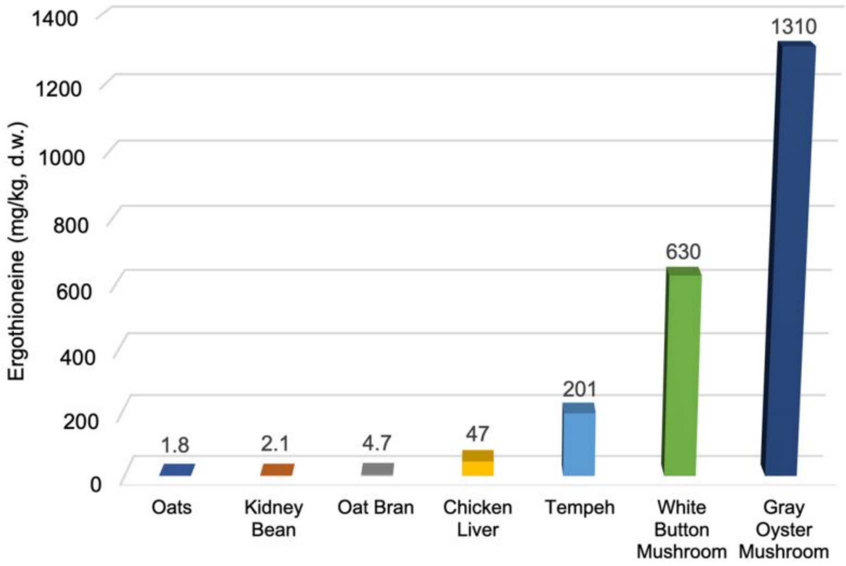


Beelman, Robert B et al. “Is ergothioneine a 'longevity vitamin' limited in the American diet?.” Journal of nutritional science vol. 9 e52. 11 Nov. 2020, doi:10.1017/jns.2020.44
Cheah, Irwin K et al. “Ergothioneine levels in an elderly population decrease with age and incidence of cognitive decline; a risk factor for neurodegeneration?.” Biochemical and biophysical research communications vol. 478,1 (2016): 162-167. doi:10.1016/j.bbrc.2016.07.074

L-ergothioneine sources

L-ergothioneine is mainly present in **mushrooms**





Beelman, Robert B et al. “Is ergothioneine a 'longevity vitamin' limited in the American diet?.” Journal of nutritional science vol. 9 e52. 11 Nov. 2020, doi:10.1017/jns.2020.44

Conclusions

1. We built transcriptomic **age-classification** model
2. We applied this model to **identify treatments** that result in a "young" transcriptome
3. We additionally investigated the **role of L-ergothioneine in the aging**

Drawbacks

Why do our results differ from the paper?

1. We used **reduced data** to save time. We selected some organs and ran a sample of 100 treatments (out of 3546 total) on them. We also ran our model for all treatments, but only for the brain.
2. Our compounds-**ranking** procedure differs from the paper due to lack of ran models
3. We suspect we used a slightly different **version of CMap** dataset. In our data we had 3546 treatments against 1309 in the paper.

Team

Ksenia Kubenko

Transcriptome data processing and CMap data preparation (mean expression by cell type calculation)

Ekaterina Kashuk

Random Forest model generation and selection, selection of geroprotectors

Nikita Vaulin

CMap data preparation (log2FC calculation), literature review on geroprotectors

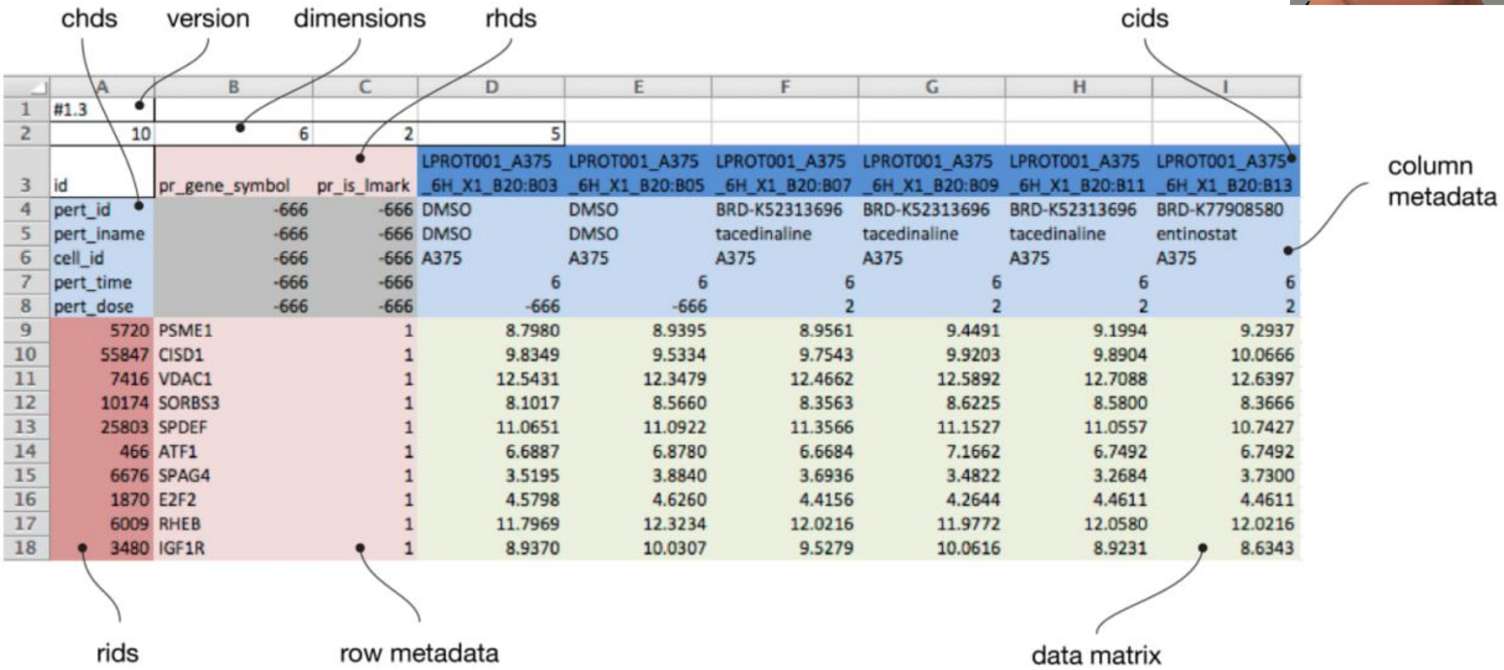
All authors contributed to the presentation and aging

Geroprotectors table

old	yng	treatment	normalized_pred	geroprotective_index
0.498	0.502	EMF-sumo1-7	1.004	0.0656
0.496	0.504	ochratoxin-a	1.008	0.0670
0.498	0.502	nafamostat	1.004	0.0656
0.496	*0.504*	*L-ergothioneine*	*1.008*	*0.0670*
0.494	**0.506**	**olanzapine**	**1.012**	**0.0740**
0.494	**0.506**	**XL-888**	**1.012**	**0.0740**
0.498	0.502	E-2012	1.004	0.0656
0.494	**0.506**	**ritonavir**	**1.012**	**0.0740**
0.49	**0.51**	**thioguanine**	**01.02**	**0.0815**
0.498	0.502	tebipenem	1.004	0.0656
0.496	0.504	etofylline-clofibrate	1.008	0.0670

CMap

Слайд про CMap　Работа с CMap



	A	B	C	D	E	F	G	H	I
1	#1.3								
2	10	6	2	5					
3	id	pr_gene_symbol	pr_is_lmark	LPROT001_A375_6H_X1_B20:B03	LPROT001_A375_6H_X1_B20:B05	LPROT001_A375_6H_X1_B20:B07	LPROT001_A375_6H_X1_B20:B09	LPROT001_A375_6H_X1_B20:B11	LPROT001_A375_6H_X1_B20:B13
4	pert_id	-666	-666	DMSO	DMSO	BRD-K52313696	BRD-K52313696	BRD-K52313696	BRD-K77908580
5	pert_iname	-666	-666	DMSO	DMSO	tacedinaline	tacedinaline	tacedinaline	entinostat
6	cell_id	-666	-666	A375	A375	A375	A375	A375	A375
7	pert_time	-666	-666	6	6	6	6	6	6
8	pert_dose	-666	-666	-666	-666	2	2	2	2
9	5720	PSME1	1	8.7980	8.9395	8.9561	9.4491	9.1994	9.2937
10	55847	CISD1	1	9.8349	9.5334	9.7543	9.9203	9.8904	10.0666
11	7416	VDAC1	1	12.5431	12.3479	12.4662	12.5892	12.7088	12.6397
12	10174	SORBS3	1	8.1017	8.5660	8.3563	8.6225	8.5800	8.3666
13	25803	SPDEF	1	11.0651	11.0922	11.3566	11.1527	11.0557	10.7427
14	466	ATF1	1	6.6887	6.8780	6.6684	7.1662	6.7492	6.7492
15	6676	SPAG4	1	3.5195	3.8840	3.6936	3.4822	3.2684	3.7300
16	1870	E2F2	1	4.5798	4.6260	4.4156	4.2644	4.4611	4.4611
17	6009	RHEB	1	11.7969	12.3234	12.0216	11.9772	12.0580	12.0216
18	3480	IGF1R	1	8.9370	10.0307	9.5279	10.0616	8.9231	8.6343

CMap

```
Ksusha's means
(2466225, 3)
```

	Transcript	sample	value
0	100017	AML001	6.847573
1	100019	AML001	6.603014
2	100037258	AML001	6.761893

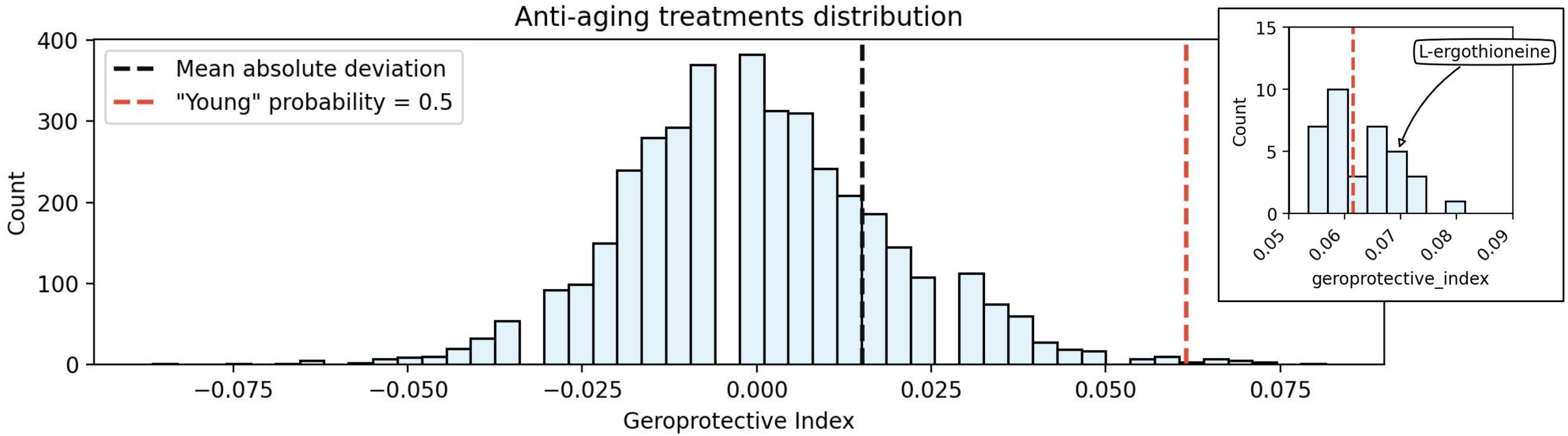
```
CMap
(214958468, 4)
```

	Transcript	variable	value	sample
0	12558	AML001_CD34_24H_X1_F1B10:E03	4.6286	AML001
1	11308	AML001_CD34_24H_X1_F1B10:E03	6.5425	AML001
2	12560	AML001_CD34_24H_X1_F1B10:E03	4.4462	AML001

```
%%time

cmap_means_merged = cmap_melted.merge(means_melted, on=['Transcript', 'sample']).compute()
```

```
CPU times: user 58.8 s, sys: 41.3 s, total: 1min 40s
Wall time: 59.3 s
```



Anti-aging treatments distribution
Mean absolute deviation
"Young" probability = 0.5
Count
Geroprotective Index
400
300
200
100
0
−0.075
−0.050
−0.025
0.000
0.025
0.050
0.075
L-ergothioneine
15
10
5
0
0.05
0.06
0.07
0.08
0.09
geroprotective_index