

Additional file 2 - Supplementary Note 1. Significant cross-sectional associations between phenotypes and epigenetic measures of ageing in both discovery and replication cohorts in a basic model adjusting for age and sex.

Cardiovascular Diseases

AgeAccelGrim alone was associated with heart disease (Odds Ratio (OR) per SD = 1.71, $P = 1.0 \times 10^{-7}$) but not stroke following replication.

In relation to continuous traits, AgeAccelGrim, DunedinPoAm, AgeAccelPheno and EEAA were positively associated with average heart rate ($\beta = [0.08, 0.18]$, $P = [1.1 \times 10^{-4}, 1.1 \times 10^{-19}]$), with AgeAccelGrim showing the strongest association in the replication set. AgeAccelPheno and IEAA were positively associated with body mass index ($\beta = [0.07, 0.11]$, $P = [5.5 \times 10^{-4}, 2.7 \times 10^{-8}]$). AgeAccelPheno was positively associated with higher diastolic blood pressure ($\beta = 0.10$, $P = 2.4 \times 10^{-7}$). AgeAccelGrim and DunedinPoAm were negatively associated with high-density lipoprotein cholesterol ($\beta = -0.11$ and -0.10 , $P = 1.9 \times 10^{-8}$ and 2.4×10^{-7} , respectively). AgeAccelGrim and AgeAccelPheno were positively associated with waist-to-hip ratio ($\beta = 0.15$ and 0.10 , $P = 1.1 \times 10^{-16}$ and 1.6×10^{-8} , respectively). DNAmTLadjAge was negatively associated with average heart rate ($\beta = -0.08$, $P = 1.7 \times 10^{-4}$) and waist-to-hip ratio ($\beta = -0.08$, $P = 7.6 \times 10^{-6}$).

Neurological and Psychiatric Diseases

AgeAccelGrim was associated with both self-reported depression (OR = 1.26, $P = 8.7 \times 10^{-5}$) and SCID Depression (OR = 1.18, $P = 6.6 \times 10^{-5}$). None of the measures were associated with either maternal or paternal history of Alzheimer's disease.

AgeAccelGrim and DunedinPoAm were both negatively associated with general cognitive ability ($\beta = -0.14$ and -0.13 , $P = 1.2 \times 10^{-11}$ and 1.5×10^{-11} , respectively) and a general factor of fluid intelligence (β

= -0.14 and -0.11, $P = 1.5 \times 10^{-12}$ and 6.1×10^{-9} , respectively). AgeAccelGrim and DunedinPoAm were also associated with neuroticism ($\beta = 0.11$ and 0.08 , $P = 4.8 \times 10^{-8}$ and 7.8×10^{-5} , respectively). DunedinPoAm, AgeAccelGrim and AgeAccelPheno were negatively associated with the Scottish Index of Multiple Deprivation (SIMD; lower indices correspond to more deprivation) ($\beta = [-0.12, -0.27]$, $P = [1.0 \times 10^{-9}, 1.7 \times 10^{-38}]$). DNAmTLadjAge was positively associated with SIMD ($\beta = 0.15$, $P = 4.6 \times 10^{-13}$).

Pulmonary Diseases

DunedinPoAm and AgeAccelGrim were associated with COPD (OR = 3.42 and 3.49, $P = 3.6 \times 10^{-11}$ and 1.4×10^{-13} , respectively). No measure of epigenetic ageing was associated with lung cancer (no. of events = 5).

AgeAccelGrim, AgeAccelPheno and DunedinPoAm were negatively associated with forced expiratory flow ($\beta = [-0.07, -0.08]$, $P = [1.7 \times 10^{-5}, 2.6 \times 10^{-23}]$) and forced expiratory volume ($\beta = [-0.06, -0.15]$, $P = [4.3 \times 10^{-5}, 5.7 \times 10^{-24}]$). AgeAccelGrim and DunedinPoAm were also negatively associated with forced vital capacity ($\beta = -0.08$ and -0.05 , $P = 6.5 \times 10^{-8}$ and 2.7×10^{-4} , respectively). DunedinPoAm was strongly associated with smoking pack years, representing the strongest association overall ($\beta = 0.38$, $P = 4.1 \times 10^{-91}$). AgeAccelPheno was also positively associated with smoking pack years ($\beta = 0.17$, $P = 1.4 \times 10^{-17}$). DNAmTLadjAge was positively associated with forced expiratory flow ($\beta = 0.08$, $P = 1.4 \times 10^{-5}$), forced expiratory volume ($\beta = 0.07$, $P = 5.4 \times 10^{-6}$) and negatively associated with pack years ($\beta = -0.20$, $P = 2.5 \times 10^{-23}$).

Diabetes Mellitus and Kidney Disease

AgeAccelGrim and AgeAccelPheno were associated with diabetes (OR: 1.47 and 1.48, $P = 1.1 \times 10^{-4}$ and 8.7×10^{-5} , respectively).

AgeAccelGrim and EEAA were positively associated with creatinine ($\beta = 0.08$ and 0.14 , $P = 4.6 \times 10^{-5}$ and 5.0×10^{-13} , respectively). DNAmTLadjAge was negatively associated with creatinine ($\beta = -0.11$, $P = 2.0 \times 10^{-8}$).

Cancer

None of the ageing measures were associated with either bowel or breast cancer following multiple testing correction.

Neck and Back Pain

DunedinPoAm and AgeAccelGrim were associated with back pain ($OR = 1.29$ and 1.34 , $P = 1.9 \times 10^{-4}$, 5.4×10^{-6} , respectively). No measures were associated with neck pain.

Table 1. Significant relationships between phenotypes and epigenetic measures of ageing present in both discovery and replication cohorts in a basic model. Those phenotypes which remained significant in both cohorts in a subsequent fully-adjusted model are emboldened.

		Discovery Cohort			Replication Cohort		
Categorical Phenotypes							
Measure	Variable	n event	OR	P	n event	OR	P
GrimAge	SCID Depression	825	1.45	1.8 x 10 ⁻²²	984	1.18	6.6 x 10 ⁻⁰⁵
GrimAge	Depression	371	1.62	2.5 x 10 ⁻²²	414	1.26	8.7 x 10 ⁻⁰⁵
GrimAge	COPD	48	2.37	1.4 x 10⁻¹²	32	3.49	1.4 x 10⁻¹³
GrimAge	Heart Disease	196	1.54	5.2 x 10 ⁻¹⁰	95	1.71	1.0 x 10 ⁻⁰⁷
GrimAge	Diabetes	147	1.58	1.4 x 10 ⁻⁰⁹	89	1.47	1.1 x 10 ⁻⁰⁴
PhenoAge	Diabetes	147	1.55	2.3 x 10 ⁻⁰⁸	89	1.48	8.7 x 10 ⁻⁰⁵
DunedinPoAm	COPD	48	1.89	3.4 x 10 ⁻⁷	32	3.42	3.6 x 10 ⁻¹¹
DunedinPoAm	Back Pain	480	1.23	4.2 x 10 ⁻⁵	293	1.29	1.9 x 10 ⁻⁰⁴
GrimAge	Back Pain	480	1.26	1.7 x 10 ⁻⁰⁵	293	1.34	5.4 x 10 ⁻⁰⁶
Continuous Phenotypes							
Measure	Variable	n	β	P	n	β	P
DunedinPoAm	Pack Years	4380	0.46	5.0 x 10⁻²³⁴	2522	0.38	4.1 x 10⁻⁹¹
DunedinPoAm	SIMD	4236	-0.24	8.7 x 10⁻⁵⁷	2457	-0.24	9.1 x 10⁻³⁴
GrimAge	SIMD	4236	-0.28	3.3 x 10⁻⁷²	2457	-0.27	1.7 x 10⁻³⁸
HannumAge	Creatinine	4427	0.24	2.8 x 10⁻⁶²	0.14	0.02	5.0 x 10⁻¹³
DunedinPoAm	Average Heart Rate	4444	0.21	4.0 x 10⁻⁴⁸	2572	0.13	2.1 x 10⁻¹¹
GrimAge	FEV	3750	-0.16	1.1 x 10⁻⁴²	2191	-0.15	5.7 x 10⁻²⁴
GrimAge	g	4291	-0.22	6.1 x 10 ⁻⁴²	2504	-0.14	1.2 x 10 ⁻¹¹
GrimAge	Average Heart Rate	4444	0.20	1.6 x 10⁻³⁷	2572	0.18	1.1 x 10⁻¹⁹

GrimAge	gf	4324	-0.19	5.9×10^{-37}	2529	-0.14	1.5×10^{-12}
DunedinPoAm	g	4291	-0.19	2.1×10^{-36}	2504	-0.13	1.5×10^{-11}
GrimAge	Waist:Hip Ratio	4383	0.15	6.5×10^{-33}	2535	0.15	1.1×10^{-16}
DunedinPoAm	gf	4324	-0.17	8.4×10^{-31}	2529	-0.11	6.1×10^{-9}
GrimAge	FEF	3750	-0.16	1.8×10^{-29}	2185	-0.18	2.6×10^{-23}
DunedinPoAm	FEV	3776	-0.12	7.8×10^{-29}	2191	-0.10	1.9×10^{-12}
DNAmTL	Pack Years	4380	-0.17	3.0×10^{-28}	2522	-0.20	2.5×10^{-23}
PhenoAge	Body Mass Index	4423	0.16	1.4×10^{-26}	2567	0.11	2.7×10^{-08}
PhenoAge	Pack Years	4380	0.15	2.4×10^{-24}	2522	0.17	1.4×10^{-17}
DunedinPoAm	HDL Cholesterol	4396	-0.14	2.8×10^{-23}	2543	-0.10	2.4×10^{-7}
GrimAge	FVC	3775	-0.12	1.1×10^{-22}	2191	-0.08	6.5×10^{-08}
DunedinPoAm	FEF	3750	-0.13	4.3×10^{-21}	2185	-0.13	8.3×10^{-14}
PhenoAge	Waist:Hip Ratio	4383	0.11	1.3×10^{-20}	2535	0.10	1.6×10^{-08}
GrimAge	HDL Cholesterol	4396	-0.14	1.4×10^{-20}	2543	-0.11	1.9×10^{-08}
GrimAge	Creatinine	4427	0.14	2.5×10^{-20}	2563	0.08	4.6×10^{-05}
PhenoAge	Average Heart Rate	4444	0.13	3.3×10^{-19}	2572	0.14	1.6×10^{-13}
DNAmTL	SIMD	4236	0.13	5.5×10^{-17}	2457	0.15	4.6×10^{-13}
DunedinPoAm	FVC	3775	-0.13	4.3×10^{-21}	2191	-0.05	2.7×10^{-04}
DNAmTL	Creatinine	4427	-0.11	3.3×10^{-14}	2563	-0.11	2.0×10^{-08}
PhenoAge	SIMD	4236	-0.11	7.7×10^{-14}	2457	-0.12	1.0×10^{-09}
HannumAge	Average Heart Rate	4444	0.11	9.2×10^{-13}	2572	0.08	1.1×10^{-04}
DunedinPoAm	Body Mass Index	4432	0.10	1.8×10^{-11}	22567	0.07	5.5×10^{-04}
PhenoAge	FEV	3776	-0.07	1.5×10^{-09}	2191	-0.06	4.3×10^{-05}

DNAmtL	Average Heart Rate	4444	-0.09	4.4×10^{-9}	2572	-0.08	1.7×10^{-4}
GrimAge	Neuroticism	4426	0.08	5.7×10^{-8}	2565	0.11	4.8×10^{-8}
DunedinPoAm	Neuroticism	4426	0.07	4.6×10^{-7}	2565	0.08	7.8×10^{-5}
DNAmtL	FEV	3776	0.05	3.9×10^{-6}	2191	0.06	5.4×10^{-6}
PhenoAge	Diastolic Pressure	4447	0.06	4.3×10^{-6}	2573	0.10	2.4×10^{-7}
DNAmtL	Waist:Hip Ratio	4383	-0.06	6.9×10^{-6}	2535	-0.08	7.6×10^{-7}
HorvathAge	Body Mass Index	4423	0.06	4.2×10^{-5}	2567	0.09	1.2×10^{-5}
PhenoAge	FEF	3750	-0.06	4.6×10^{-5}	2185	-0.07	1.7×10^{-5}
DNAmtL	FEF	3750	0.06	5.0×10^{-5}	2185	0.07	1.4×10^{-4}
<i>Mortality Analysis</i>							
Measure	Variable	n event	HR	P	n events	HR	P
GrimAge	All-Cause Mortality	182	1.87	$<2.0 \times 10^{-16}$	57	1.7	6.5×10^{-5}
DunedinPoAm	All-Cause Mortality	182	1.7	5.4×10^{-15}	57	1.69	8.1×10^{-5}

COPD (chronic obstructive pulmonary disease), FEF (forced expiratory flow), FEV (forced expiratory volume), FVC (forced vital capacity), g (general factor of cognitive ability), gf (general factor of fluid intelligence), HDL (high-density lipoprotein), HR (hazard ratio), OR (odds ratio), SCID (Structured Clinical Interview for DSM), SIMD (Scottish Index of Multiple Deprivation).