

**Sex-specific changes in autosomal methylation rate in ageing common terns**

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by

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**Table S1.** Information on the 74 samples collected from 34 individual common terns from 17 breeding pairs, including bird ID, year of sampling, sex, age at sampling, total number of methylation reads obtained, number of aligned reads obtained, mapping efficiency (in %), bisulfite conversion rate (in %), number of unfiltered CpGs and number of CpGs after filtering for >10x and <99.9th percentile coverage.

bird				total	aligned	mapping	conversion	CpG unfiltered	CpG filtered
ID	year	sex	age	reads	reads	efficiency	rate		
16	2014	2	13	54777086	31548630	57.59	99.21	7797103	688002
16	2017	2	16	67008519	41955885	62.61	99.41	4273340	1201348
8	2013	1	18	54767267	26238418	47.91	99.40	5800891	675152
8	2014	1	19	51863344	32293792	62.27	99.33	5686008	667405
12	2013	1	14	50813756	23581570	46.41	99.39	5409816	802832
12	2014	1	15	57378291	31663194	55.18	99.33	5579351	715575
12	2017	1	18	49245261	30346895	61.62	98.96	5529160	642686
27	2014	1	5	42892021	25986584	60.59	99.16	4954029	348814
27	2017	1	8	59278719	37547078	63.34	99.29	5122542	852908
3	2013	1	17	49551676	23533540	47.49	99.45	7131007	304770
3	2014	1	18	57897826	30379000	52.47	99.40	5614283	993432
5	2013	2	13	39951705	15914513	39.83	99.44	5491498	718390
5	2014	2	14	41847260	16450237	39.31	99.38	3874555	735391
18	2014	1	13	72326650	44663034	61.75	99.15	9135083	760787
18	2017	1	16	67348209	47510992	70.55	98.86	7680463	706584
9	2013	2	7	58044974	34440005	59.33	99.35	8656356	407377
9	2014	2	8	24942071	15866698	63.61	99.00	1911253	498140
26	2014	2	4	57239241	35252552	61.59	99.27	7340117	588774
26	2017	2	7	28307901	17976338	63.50	98.66	1464069	679344
29	2014	2	4	55400339	36241908	65.42	99.07	3265500	1163019
29	2017	2	7	40777189	25876662	63.46	99.05	6040359	378191
23	2014	1	4	78803246	55019176	69.82	99.05	1361472	1112660
23	2017	1	7	59149872	40432219	68.36	99.02	6779330	617986
34	2013	2	12	60023665	33027151	55.02	99.45	8325552	1011803
34	2017	2	16	38157280	25005040	65.53	98.57	6097280	261078
22	2013	2	12	33457109	10254303	30.65	99.43	3253239	256097

22	2014	2	13	42125465	24274415	57.62	99.26	5877438	334286
22	2017	2	16	44317724	27187356	61.35	99.06	5187049	457993
32	2014	2	4	42992411	26329522	61.24	98.87	5341475	552478
32	2017	2	7	52017038	30538273	58.71	99.20	4127165	846139
10	2014	2	7	38032755	17958580	47.22	99.31	3715593	498390
10	2017	2	10	61458457	37334663	60.75	98.23	7637488	474871
31	2014	1	3	57355267	36300007	63.29	99.32	5151980	772251
31	2017	1	6	48257844	31810506	65.92	98.77	5969976	569760
28	2014	2	4	71994197	47533588	66.02	98.41	1099343	757686
28	2017	2	7	58763104	37346153	63.55	98.97	6826871	653598
30	2014	1	4	43557575	24693715	56.69	99.27	5500641	452923
30	2017	1	7	45506638	27950342	61.42	99.06	7048571	487652
21	2014	1	6	30979667	17966442	57.99	99.26	5494908	277943
21	2017	1	9	51625588	31107383	60.26	99.31	6252722	483029
24	2014	1	4	69287030	43323814	62.53	99.20	8655737	852155
24	2017	1	7	51127096	32728222	64.01	99.24	6353028	530474
7	2013	1	12	51529741	20239308	39.28	99.42	4982777	635192
7	2014	1	13	52594196	27862996	52.98	99.39	4714086	722434
11	2013	2	14	34806822	9647441	27.72	99.43	4159247	542808
11	2014	2	15	51104181	17299884	33.85	99.40	3779860	726385
11	2017	2	18	32780045	17617203	53.74	99.47	3224271	635251
4	2013	2	19	34915343	10640666	30.48	99.41	3533478	320103
4	2014	2	20	35465505	13106425	36.96	99.41	3637525	674374
2	2013	2	10	69780306	40432799	57.94	99.30	7798402	699174
2	2014	2	11	54182101	32598451	60.16	99.34	5305365	700119
17	2014	2	6	55141490	31269762	56.71	99.31	7726477	897338
17	2017	2	9	70446371	39946939	56.71	99.44	3555459	1487064
13	2014	2	13	50605752	24173905	47.77	99.32	5119304	628289
13	2017	2	16	58449090	37104983	63.48	98.22	6701658	505201
19	2013	1	15	58271307	31207343	53.56	99.29	6123172	599398
19	2014	1	16	53391308	30605374	57.32	99.20	7641306	735559
19	2017	1	19	82760425	50218876	60.68	98.80	8652556	905387
20	2013	1	5	42736396	20070619	46.96	99.42	3041406	940815
20	2014	1	6	43669575	21507206	49.25	99.34	6316423	673655
20	2017	1	9	52822526	29760937	56.34	98.88	6660278	529165

6	2013	1	10	65457412	28896915	44.15	99.41	7696664	614548
6	2014	1	11	50422031	23410387	46.43	99.40	4689912	753516
14	2014	1	13	59899049	33507686	55.94	99.34	5793552	851990
14	2017	1	16	56712154	34144112	60.21	98.91	7291024	493699
25	2014	2	4	51340048	33919009	66.07	99.20	6608459	480020
25	2017	2	7	60654555	37960138	62.58	98.71	3761318	1099406
33	2013	1	13	52785750	29867983	56.58	99.41	8242606	470503
33	2017	1	17	37616309	19712482	52.40	99.53	2414913	1145111
15	2014	1	8	40506842	19779762	48.83	99.25	6240175	445186
15	2017	1	11	35769540	21991871	61.48	99.36	3687276	433576
1	2013	2	5	45333405	20016916	44.15	99.20	5189812	419008
1	2014	2	6	61743501	32382636	52.45	99.26	6837951	856112
1	2017	2	9	45327429	25015606	55.19	99.14	4776749	536500

**Table S2.** Average ( $\pm$  SD) number of sequenced and aligned reads with their mapping efficiency, bisulfite conversion rate, and number of (un)filtered CpGs for the 74 samples collected from 34 individual common terns from 17 breeding pairs.

Total reads	51,482,011 ( $\pm$ 11,868,120)
Aligned reads	29,044,662 ( $\pm$ 9,591,384)
Mapping efficiency	55.57 % ( $\pm$ 9.62)
Conversion Rate	99.19 ( $\pm$ 0.28)
CpG unfiltered	5,536,718 ( $\pm$ 1,845,420)
CpG (10x min coverage)	661,447 ( $\pm$ 240,806)

**Table S3.** Results from a Generalised Linear Mixed Model with a binomial error distribution testing whether variation in autosomal methylation rate of 17 male common terns is explained by the between- (average age) and within-individual (delta age) components of age. Estimates and 95% confidence intervals (CI) are provided for each fixed (mean) and random (standard deviation) effect. The data set includes 475,067 observations on 15,700 CpG positions.

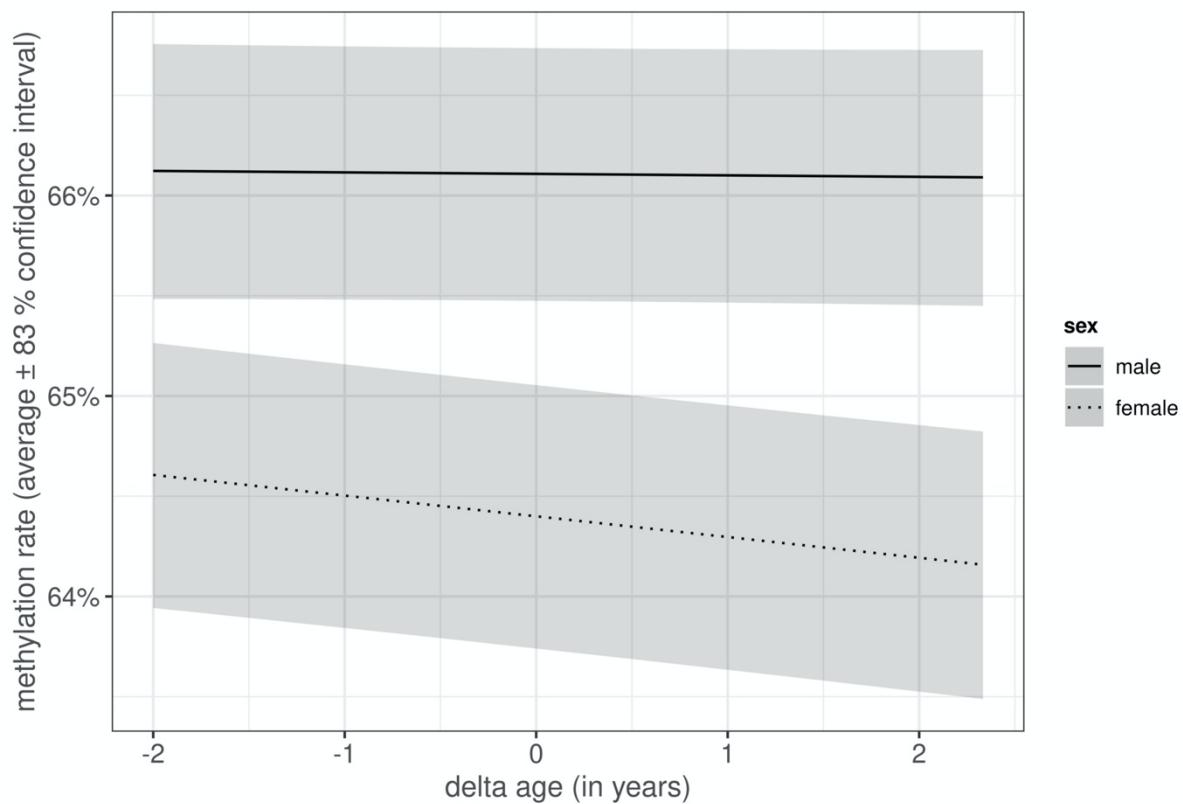
parameter	estimate	95% CI	z	p
<b>fixed effects</b>				
intercept	0.6187	0.4457, 0.7927	6.97	<0.001
average age	0.0040	-0.0105, 0.0184	0.535	0.593
delta age	-0.0008	-0.0034, 0.0019	-0.576	0.565
<b>random effects</b>				
bird identity	0.1424	0.0687, 0.2951		
position identity	1.3471	1.3317, 1.3626		
observation	1.1622	1.1586, 1.1658		

**Table S4.** Results from a Generalised Linear Mixed Model with a binomial error distribution testing whether variation in autosomal methylation rate of 17 female common terns is explained by the between- (average age) and within-individual (delta age) components of age. Estimates and 95% confidence intervals (CI) are provided for each fixed (mean) and random (standard deviation) effect. Significant fixed effects (p-value <0.05) are presented in bold. The data set includes 452,423 observations on 15,700 CpG positions.

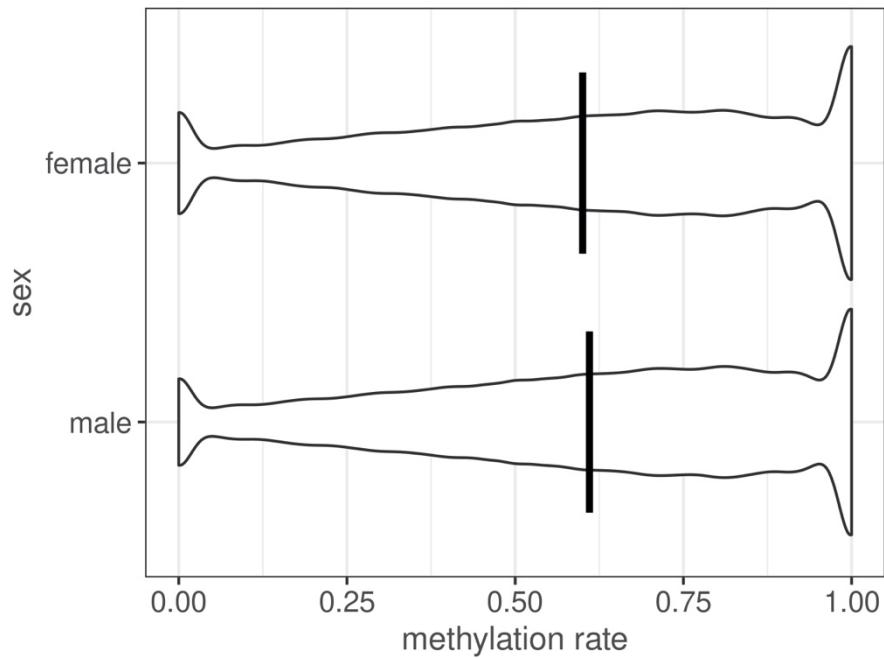
parameter	estimate	95% CI	z	p
<b>fixed effects</b>				
intercept	0.6732	0.5803, 0.7661	14.203	<0.001
average age	-0.0052	-0.0133, 0.0030	-1.248	0.212
delta age	<b>-0.0044</b>	<b>-0.0073, -0.0014</b>	<b>-2.925</b>	<b>0.003</b>
<b>random effects</b>				
bird identity	0.0750	0.0540, 0.1041		
position identity	1.3503	1.3347, 1.3660		
observation	1.2645	1.2605, 1.2685		

**Table S5.** Results from a Generalised Linear Mixed Model with a binomial error distribution testing whether variation in W chromosome methylation rate of 17 female common terns is explained by the between- (average age) and within-individual (delta age) components of age. Estimates and 95% confidence intervals (CI) are provided for each fixed (mean) and random (standard deviation) effect. The data set includes 2,936 observations on 103 CpG positions (sequenced in at least 70 % of the female samples).

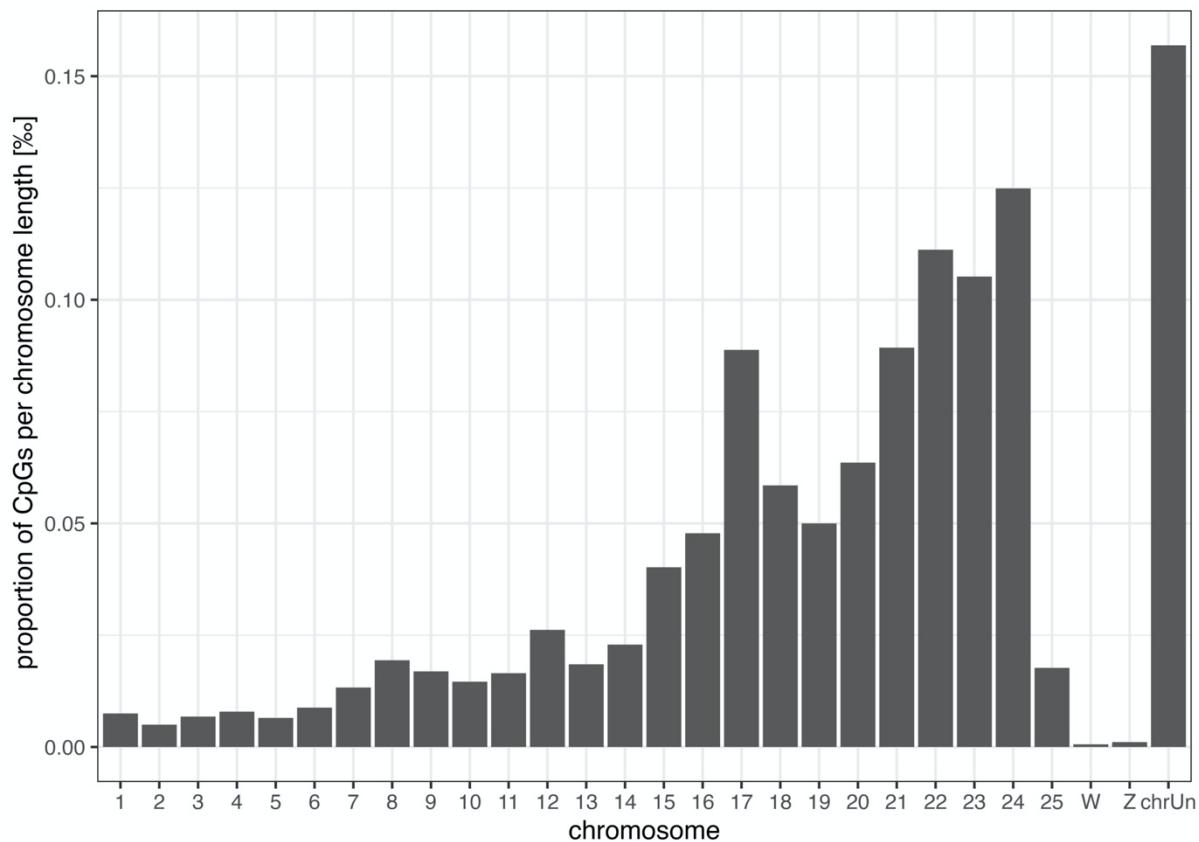
parameter	estimate	95% CI	z	p
<b>fixed effects</b>				
intercept	<b>1.2199</b>	<b>0.7459, 1.6938</b>	<b>5.045</b>	<b>&lt;0.001</b>
average age	-0.0092	-0.0283, 0.0098	-0.95	0.342
delta age	0.0049	-0.0329, 0.0428	0.254	0.799
<b>random effects</b>				
bird identity	0.1338	0.068, 0.2630		
position identity	2.1851	1.8924, 2.5232		
observation	1.2287	1.1766, 1.2831		



**Figure S1.** Autosomal DNA methylation rate (estimated across 15,700 genomic positions) declines with age within individual female (dotted line), but not male (solid line), common terns. Lines represent the model-predicted within-individual change in methylation rate with age, grey areas the 83% confidence interval around this change.



**Figure S2.** Violin plots showing the mean autosomal methylation rates (across 15,700 genomic positions) for male and female common terns.



**Figure S3.** Proportion (in permille) of 15,816 CpGs per chromosome length sequenced in 74 samples obtained from 34 individual common terns across the chromosomes. Smaller scaffolds for which the chromosome was unknown are merged and named ‘chrUn’.