

Supplementary Material

1 Body size results



Figure S1: Principal Component Analysis (PCA) of body size metrics of *Neotermes chilensis* soldiers from unicolonial (yellow circles) and multicolonial (blue circles) scapes (See Table 1 for variable abbreviations). Bar charts at the right panel show the ten traits mostly contributing to each principal component. (A) PCA performed with 17 traits and (B) with only the 10 most contributed traits to the variation of body size metrics between unicolonial and multicolonial soldiers. The overall pattern of correlations between metrics showed that variations were concentrated along the first two PCA axis. The contributions of variables in accounting for the variability in a given principal component are expressed in percentage. The red line indicates the expected average

contribution of each variable. For each axis, a variable with a contribution higher than this threshold was considered as important to the component. The total contribution of a given variable to each axis was calculated using the function *fviz.contrib* of FactoMineR package (Le et al., 2008).

	PC1 PC 2		2	PC	23	РС	24	PC5		
Eigenvalues		7.9		2.2		1.9		1.4		0.8
Variance (%)		46.2		13.0		11.4		8.0		4.5
Cumulative variance (%)		46.2		59.2		70.6		78.7		83.2
Traits	Contr.	Corr.	Contr.	Corr.	Contr.	Corr.	Contr.	Corr.	Contr.	Corr.
RMA	6.83	0.73	12.47	0.53	0.50	-0.10	5.14	-0.26	0.27	-0.05
LMA	6.36	0.71	2.91	0.25	17.35	-0.58	0.70	-0.10	2.16	-0.13
RMP	5.67	0.67	16.74	0.61	1.65	-0.18	3.43	-0.22	0.01	0.01
LMP	5.03	0.63	3.02	0.26	20.45	-0.63	0.23	-0.06	2.73	-0.14
IDR	4.43	0.59	10.80	0.49	0.00	-0.01	13.33	0.43	0.01	0.01
IDL	2.94	0.48	0.61	0.12	1.00	-0.14	22.16	0.55	46.37	0.60
НА	9.58	0.87	9.07	-0.45	0.19	-0.06	1.37	0.14	0.66	-0.07
НР	9.44	0.86	9.58	-0.46	0.23	-0.07	1.05	0.12	0.99	-0.09
HL	8.83	0.83	9.83	-0.47	0.01	-0.02	1.10	0.12	0.73	-0.08
HMxW	7.60	0.77	7.34	-0.40	0.47	-0.10	0.82	0.11	5.64	-0.21
HMnW	4.35	0.58	7.65	-0.41	3.95	-0.28	0.01	-0.01	5.20	0.20
PmL	4.36	0.59	0.04	-0.03	3.62	0.27	14.31	-0.44	18.40	0.38
PmW	4.85	0.62	0.59	-0.11	0.81	0.13	20.35	-0.53	9.11	0.26
PnL	6.46	0.71	0.02	0.02	15.99	0.56	0.06	0.03	0.37	-0.05
PnW	6.67	0.72	0.01	0.01	9.42	0.43	3.50	-0.22	1.09	-0.09
MsL	3.38	0.52	7.46	0.41	8.64	0.41	11.96	0.40	0.02	-0.01
MtL	3.22	0.50	1.85	0.20	15.71	0.55	0.46	0.08	6.25	-0.22

Table S1. PCA statistics from the 17	7 traits to body size variation between soldiers from
unicolonial and multicolonial scapes	(Fig. S1A)

Contr.: trait contribution (%) calculated using the function *fviz.contrib* of FactoMineR package (Le et al., 2008). **Corr**.: correlation coefficient of traits with the principal components.

Table S2. PCA statistics from the 10 most contributing traits to body size variation between soldiers form unicolonial and multicolonial scapes, as selected from the PCA with 17 traits (Fig. S1b)

	РС	C1	РС	2	РС	23	РС	24	РС	5
Eigenvalues		5.81		1.90		1.09		0.60		0.23
Variance (%)		58.1		18.9		10.9		6.0		2.3
Cumulative variance (%)		58.1		77.0		87.9		93.9		96.2
	Contr.	Corr.								
RMA	8.80	0.71	20.52	0.62	0.13	-0.04	7.69	-0.21	9.89	-0.15
LMA	8.09	0.69	6.30	0.35	24.03	-0.51	1.64	-0.10	59.69	0.37
НА	13.91	0.90	8.61	-0.40	1.00	-0.10	0.09	0.02	1.31	-0.06
НР	13.64	0.89	9.26	-0.42	1.31	-0.12	0.00	0.01	0.81	-0.04
HL	12.76	0.86	10.03	-0.44	0.59	-0.08	0.04	0.02	0.98	-0.05
HMxW	11.30	0.81	8.53	-0.40	5.12	-0.24	0.00	0.00	2.40	-0.07
PnL	8.84	0.72	0.01	-0.01	37.56	0.64	1.55	0.10	7.48	0.13
PnW	9.46	0.74	0.19	0.06	29.59	0.57	10.60	-0.25	3.84	0.09
RMP	7.25	0.65	26.11	0.70	0.64	-0.08	3.23	-0.14	13.55	-0.18
IDR	5.95	0.59	10.45	0.45	0.04	0.02	75.15	0.67	0.05	-0.01

Contr.: trait contribution (%) calculated using the function *fviz.contrib* of FactoMineR package (Le et al., 2008). **Corr.**: correlation coefficient of traits with the principal components.

2 Fluctuating asymmetry results

Preliminary tests for fluctuating asymmetry analyses in order to assess occurrence of antisymmetry, directional asymmetry, body size dependence and measurement errors of data in four traits of soldiers of *Neotermes chilensis* from unicolonial and multicolonial scapes.

Table S3: Normality distribution test (Kolmogorov-Smirnov with Lilliefors correction), skewness and kurtosis for (R-L) distribution. P-values equal or higher than 0.05 indicate non-significant antisymmetry.

Coloniality	Trait	Skowpoor	Vuntosis	Normality			
	Iran	SKewness	KUITUSIS	K-S distribution	p-value		
unicolonial	femur length	-0.009	-0.209	0.110	0.413		
	femur width	-0.688	0.598	0.141	0.116		
	tibia length	0.366	0.228	0.083	0.766		
	femur area	0.124	1.265	0.087	0.716		
multicolonial	femur length	0.676	0.905	0.155	0.179		
	femur width	-0.046	-0.300	0.135	0.352		
	tibia length	-0.137	0.004	0.108	0.649		
	femur area	0.670	-0.043	0.177	0.072		

Table S4: One sample t-test to assess deviations from zero and linear regressions between R-L sides and trait size to discard body size dependence. Two tailed t-test and slope p-values equal or higher than 0.05 indicate non-significant directional asymmetry and body size dependence, respectively. df= degrees of freedom; se= standard error.

Coloniality	Trait	Trait One sample t-test			Linear regression			
		t _{df} value	p-value	slope ± se	p-value			
al	femur length	$t_{30} = 1.86$	0.072	0.248 ± 0.180	0.178			
loni	femur width	t ₃₀ = -0.54	0.596	0.179 ± 0.183	0.337			
unico	tibia length	$t_{30} = -0.51$	0.612	0.073 ± 0.185	0.696			
	femur area	$t_{30} = 0.84$	0.409	-0.165 ± 0.183	0.375			
lticolonial	femur length	$t_{21} = -0.59$	0.562	0.051 ± 0.223	0.821			
	femur width	$t_{21} = -0.36$	0.719	-0.06 ± 0.223	0.790			
	tibia length	$t_{21} = -0.69$	0.500	-0.151 ± 0.221	0.502			
mu	femur area	$t_{21} = -0.22$	0.827	-0.02 ± 0.224	0.930			

Table S5: Two-way ANOVA with soldiers and side (triplicate data for each side) as factors to discard measurement errors. P-values lower than 0.05 in the interaction soldier x side factors indicate non-significant measurement errors.

Coloniality	Trait	Soldiers	Side	Interaction
unicolonial	femur length	F _{30,185} = 1786.1 p< 0.001	F _{1,185} = 66.45 p< 0.001	F _{30,185} = 19.14 p< 0.001
	femur width	F _{30,185} = 218.0 p< 0.001	$F_{1,185} = 1.42$ p= 0.236	F _{30,185} = 4.93 p< 0.001
	tibia length	F _{30,185} = 1211.1 p< 0.001	F _{1,185} = 41.71 p< 0.001	F _{30,185} = 156.6 p< 0.001
	femur area	F _{30,185} = 208.8 p< 0.001	$F_{1,185}$ = 3.655 p= 0.058	F _{30,185} =5.23 p< 0.001
multicolonial	femur length	F _{21,131} = 2073.9 p< 0.001	F _{1,131} = 11.02 p< 0.001	F _{21,131} = 31.32 p< 0.001
	femur width	F _{21,131} = 503.2 p< 0.001	$F_{1,131} = 5.903$ p = 0.017	F _{21,131} = 45.11 p< 0.001
	tibia length	F _{21,131} = 662.7 p< 0.001	F _{1,131} = 14.35 p< 0.001	F _{21,131} = 30.67 p< 0.001
	femur area	F _{21,131} = 4469.4 p< 0.001	F _{1,131} = 6.89 p= 0.010	F _{21,131} = 145.7 p< 0.001



Figure S2: PCA of fluctuating asymmetry index 1 calculated for four traits of *N. chilensis* soldiers from unicolonial (yellow circles) and multicolonial (blue circles) scapes (See Table 1 for variable abbreviations). Bar charts at the right panel show the ten most contributed traits to each principal component. The overall pattern of correlations between metrics showed that variation was concentrated along the first two PCA axis. See legend of Fig. S1 for explanation of variable contribution

	РС	C1	PC	22	PC	23	PC	24
Eigenvalues		1.45		1.11		0.81		0.63
Variance (%)		36.2		27.7		20.3		15.8
Cumulative variance (%)		36.2		63.9		84.1		100
	Contr.	Corr.	Contr.	Corr.	Contr.	Corr.	Contr.	Corr.
FL	28.7	0.64	2.03	0.15	69.2	0.75	0.09	0.02
FW	28.7	0.65	25.9	0.54	5.5	0.21	39.8	0.50
TL	2.5	0.19	71.1	0.89	5.3	0.21	21.1	0.37
FA	40.1	0.76	1.00	0.10	20.0	0.40	38.9	0.50

Table S6. PCA of fluctuating asymmetry index 1 calculated for four traits of *N. chilensis* soldiers from unicolonial and multicolonial scapes.

Contr.: trait contribution (%) calculated using the function *fviz.contrib* of FactoMineR package (Le et al., 2008). **Corr.**: correlation of traits with the principal components.



Figure S3: Residual plots from DHARMa package (Hartig 2018) showing the observed fitted model residual values as a function of the expected residual values from GLMMs of (A) PC1 scores for body size measurements and (B) PC1 scores of fluctuating asymmetry variables size measurements. Residual points shown as black points in the QQ plot should align exactly with the red line. In the residuals vs. predicted plot, red lines should align horizontally with black dotted lines.