## Supplementary information:

## Supplementary tables:

**Table S1:** Table listing the number of replicates per lineage per generation where fathers older than 0-1 days were used due to rearing constraints.

Lineage:	ZA♂xZA♀		AU♂rau		ZA 🗗 xAU 🕄		AU♂xZA♀	
Age (days):	0-1	1-3	0-1	1-3	0-1	1-3	0-1	1-3
F0	22	0	18	4	20	0	19	3
F1	19	1	17	1	20	2	18	1
F2	17	0	14	0	16	0	17	0

**Table S2:** Total successful replicates performed per lineage for each generation, and the number of replicates that failed to produce any offspring.

Lineage:	ZA 🗗 xZA 🕄		AU♂xAU♀		ZA 🗗 xAU 💡		AU <b>&amp;</b> xZA <b>?</b>	
Generation:	Total	Fail	Total	Fail	Total	Fail	Total	Fail
F0	22	1	22	2	20	0	22	1
F1	20	0	18	2	22	0	19	0
F2	17	1	14	1	16	1	17	2

**Table S3:** Results of Dunn's post-hoc tests of femur length of mother parasitoids of the F0 crosses, following on a significant Kruskal-Wallis test. Significant differences are indicated with an asterisk.

Pairwise comparison:	Z statistic:	Adjusted p-value:
AU♂xAU♀ - AU♂xZA♀	-0.746753	0.9104
AU♂xAU♀ - ZA♂xAU♀	1.941672	0.2087
AU♂xZA♀ - ZA♂xAU♀	2.711962	0.0334*
AU♂xAU♀ - ZA♂xZA♀	-0.883205	1.0000
AU♂xZA♀ - ZA♂xZA♀	-0.129659	0.8968
ZA♂xAU♀ - ZA♂xZA♀	-2.870570	0.0246*

**Table S4:** Results of Dunn's post-hoc tests of femur length of mother parasitoids of the F2 crosses, following on a significant Kruskal-Wallis test. Significant differences are indicated with an asterisk.

Pairwise comparison:	Z statistic:	Adjusted p-value:
AU♂xAU♀ - AU♂xZA♀	-2.673103	0.0451*
AU♂xAU♀ - ZA♂xAU♀	-1.000468	0.9513
AU♂xZA♀ - ZA♂xAU♀	1.718561	0.3428
AU♂xAU♀ - ZA♂xZA♀	-0.453958	0.6499
AU♂xZA♀ - ZA♂xZA♀	2.335002	0.0977
ZA♂xAU♀ - ZA♂xZA♀	0.580790	1.0000

**Table S5:** Pairwise Fisher post-hoc test with Holm correction comparing proportions of replicates failing to produce any offspring between lineages in the F1 generation. While the Fisher's exact test indicated a significant difference, the post-hoc test indicate there are no differences between lineages.

Pairwise comparison:	n:	p-value:	Adjusted p-value (Holm correction):
ZA♂xZA♀ - AU♂xAU♀	38	0.218	1.000
ZA♂xZA♀ - ZA♂xAU♀	42	1.000	1.000
$ZA \Im xZA $ - $AU \Im xZA $	39	1.000	1.000
AU♂xAU♀ - ZA♂xAU♀	40	0.196	1.000
AU♂xAU♀ - AU♂xZA♀	37	0.230	1.000
ZA♂xAU♀ - AU♂xZA♀	41	1.000	1.000

**Table S6:** Test statistics of separate Chi-squared tests to assess the significance of GLM model terms explaining sex ratio. Only in the F1 generation a significant effect of lineage on sex ratio was found, as indicated by the asterisk.

F0 generation							
Coefficient:	Df	Deviance	Resid. Df	Resid. Dev	Pr(>Chi)		
NULL			78	78.877			
lineage	3	4.218	75	74.659	0.239		
maternal femur length	1	0.163	74	74.496	0.686		
mating status of father	1	0.052	73	74.444	0.820		
F1 generation							
Coefficient:	Df	Deviance	Resid. Df	Resid. Dev	Pr(>Chi)		
NULL			75	112.880			
lineage	3	9.521	72	103.360	0.023*		
maternal femur length	1	3.225	71	100.140	0.073		
mating status of father	1	2.615	70	97.520	0.106		
		F2 gener	ration				
Coefficient:	Df	Deviance	Resid. Df	<b>Resid.</b> Dev	Pr(>Chi)		
NULL			58	66.986			
lineage	3	4.067	55	62.920	0.254		
maternal femur length	1	0.028	54	62.892	0.868		
mating status of father	1	0.161	53	62.731	0.688		

**Table S7:** Tukey adjusted p-values from a post-hoc analysis using the package 'emmeans' on sex ratio in the F1 generation following on a significant effect of lineage in the GLM analysis. The infinite degrees of freedom are produced by the 'emmeans' package, as it uses a z-test for calculating the Tukey adjusted p-values in the post-hoc analysis.

comparison	odds ratio	SE	df	null	z-ratio	p-value
ZA♂xZA♀ -	1.06	0.222	Inf	1	0.298	0.9908
AU∂xAU♀						
ZA∂xZA♀ -	1.32	0.251	Inf	1	1.483	0.4478
ZA♂xAU♀						
ZA∂xZA♀ -	1.68	0.342	Inf	1	2.548	0.0528.
AU∂xZA♀						
AU∂xAU♀ -	1.24	0.253	Inf	1	1.077	0.7038
ZA♂xAU♀						
AU∂xAU♀ -	1.58	0.346	Inf	1	2.086	0.1578
AU∂xZA♀						
ZA∂xAU♀ -	1.27	0.253	Inf	1	1.192	0.6319
AU∂xZA♀						

**Table S8:** Test statistics of Chi-squared tests to assess the significance of GLM model terms explaining male development time. In the F0 generation a significant effect of lineage on male development time was found, as indicated by the asterisk.

F0 generation								
Coefficient:	Df	Deviance	Resid. Df	Resid. Dev	Pr(>Chi)			
NULL			77	0.81276				
lineage	3	0.108934	74	0.70383	0.0143 *			
maternal femur length	1	0.000376	73	0.70345	0.8485			
F1 generation								
Coefficient:	Df	Deviance	Resid. Df	Resid. Dev	Pr(>Chi)			
NULL			72	0.49267				
lineage	3	0.0112354	69	0.48144	0.6693			
maternal femur length	1	0.0001696	68	0.48127	0.8782			
		F2 genera	tion					
Coefficient:	Df	Deviance	Resid. Df	Resid. Dev	Pr(>Chi)			
NULL			57	0.48112				
lineage	3	0.035051	54	0.44607	0.2518			
maternal femur length	1	0.010000	53	0.44097	0.4405			

**Table S9:** Tukey adjusted p-values from a post-hoc analysis using the package 'emmeans' on male development time in the F0 generation, following on a significant effect of lineage in the GLM analysis. Significant differences are indicated with an asterisk.

comparison	ratio	SE	df	null	t-ratio	p-value
ZA♂xZA♀ -	1.081	0.0357	73	1	2.361	0.0940
AU♂xAU♀						
ZA∂xZA♀ -	1.096	0.0375	73	1	2.663	0.0459 *
ZA♂xAU♀						
ZA∂xZA♀ -	1.022	0.0328	73	1	0.673	0.9070
AU♂xZA♀						
AU∂xAU♀ -	1.013	0.0349	73	1	0.383	0.9807
ZA♂xAU♀						
AU∂xAU♀ -	0.945	0.0313	73	1	-1.706	0.3280
AU♂xZA♀						
ZA∂xAU♀ -	0.933	0.0320	73	1	-2.027	0.1876
AU♂xZA♀						

**Table S10:** Test statistics of Chi-squared tests to assess the significance of GLM model terms explaining female development time. No significant effect of lineage or maternal femur length was found in any generation.

F0 generation							
Coefficient:	Df	Deviance	Resid. Df	Resid. Dev	Pr(>Chi)		
NULL			78	0.30019			
lineage	3	0.019941	75	0.28025	0.1474		
maternal femur length	1	0.009355	74	0.27090	0.1129		
		F1 genera	tion				
Coefficient:	Df	Deviance	Resid. Df	Resid. Dev	Pr(>Chi)		
NULL			75	0.40278			
lineage	3	0.004498	72	0.39828	0.8530		
maternal femur length	1	0.000921	71	0.39736	0.6885		
		F2 genera	tion				
Coefficient:	Df	Deviance	Resid. Df	Resid. Dev	Pr(>Chi)		
NULL			58	0.42339			
lineage	3	0.044522	55	0.37887	0.1019		
maternal femur length	1	0.000229	54	0.37864	0.8582		

Table S11: Test statistics of Chi-squared tests to assess the significance of linear model terms explaining
total offspring production. No significant effect of lineage was found in any generation. In the F1
generation there was a significant effect of maternal femur length (indicated with an asterisk).

F0 generation								
Coefficient:	Df	Sum Sq	Mean Sq	<b>F-value</b>	Pr(>F)			
lineage	3	217.89	72.632	2.1383	0.1021			
maternal femur length	1	47.10	47.100	1.3867	0.2425			
residuals	78	2649.39	33.967					
F1 generation								
Coefficient:	Df	Sum Sq	Mean Sq	<b>F-value</b>	<b>Pr(&gt;F)</b>			
lineage	3	82.65	27.55	0.9828	0.4057			
maternal femur length	1	481.54	481.54	17.1766	< 0.000 *			
residuals	73	2046.52	28.03					
		F2 gene	eration					
Coefficient:	Df	Sum Sq	Mean Sq	<b>F-value</b>	Pr(>F)			
lineage	3	89.56	29.853	0.7176	0.5454			
maternal femur length	1	0.05	0.052	0.0013	0.9718			
residuals	59	2454.39	41.600					

**Table S12**: Test statistics of Chi-squared tests to assess the significance of linear model terms explaining male offspring production. No significant effect of lineage, maternal femur length or mating status of the father was found in any generation.

F0 generation							
Coefficient:	Df	Sum Sq	Mean Sq	<b>F-value</b>	Pr(>F)		
lineage	3	34.93	11.6436	1.8282	0.1490		
maternal femur length	1	2.78	2.7816	0.4368	0.5107		
mating status of father	1	0.78	0.7811	0.1227	0.7271		
residuals	77	490.4	6.3688				

F1 generation								
Coefficient:	Df	Sum Sq	Mean Sq	F-value	Pr(>F)			
lineage	3	12.83	4.2774	0.638	0.5930			
maternal femur length	1	16.69	16.6935	2.4899	0.1190			
mating status of father	1	1.05	1.0453	0.1559	0.6941			
residuals	72	482.72	6.7045					
F2 generation								
Coefficient:	Df	Sum Sq	Mean Sq	F-value	Pr(>F)			
lineage	3	17.382	5.7939	1.2263	0.3084			
maternal femur length	1	0.003	0.0031	0.0007	0.9795			
mating status of father	1	4.576	4.5759	0.9685	0.3291			
residuals	58	274.039	4.7248					

**Table S13**: Test statistics of Chi-squared tests to assess the significance of linear model terms explaining female offspring production. No significant effect of lineage or mating status of the father was found in any generation. In the F1 generation there was a significant effect of maternal femur length (indicated with an asterisk).

F0 generation								
Coefficient:	Df	Sum Sq	Mean Sq	F-value	Pr(>F)			
lineage	3	108.21	36.071	2.0390	0.1154			
maternal femur length	1	26.99	26.990	1.5256	0.2205			
mating status of father	1	3.30	3.301	0.1866	0.6670			
residuals	77	1362.17 17.691						
F1 generation								
Coefficient:	Df	Sum Sq	Mean Sq	F-value	Pr(>F)			
lineage	3	107.58	35.86	2.2359	0.0913			
maternal femur length	1	318.92	318.92	19.8850	< 0.000 *			
mating status of father	1	36.41	36.41	2.2705	0.1362			
residuals	72	1154.74	16.04					
F2 generation								
Coefficient:	Df	Sum Sq	Mean Sq	<b>F-value</b>	<b>Pr(&gt;F)</b>			
lineage	3	56.82	18.9412	0.6838	0.5655			
maternal femur length	1	0.08	0.0811	0.0029	0.9570			
mating status of father	1	6.61	6.6118	0.2387	0.6270			
residuals	58	1606.48	27.6980					

**Table S14:** Some relevant Bioclim variables describing mean temperatures for important sites for this study. Boonah is the site where *Anaphes nitens* was collected from Australia in the current study, and the areas around Kwambonambi in Zululand, South Africa, are the target release sites. Penola is the original collection site of *A. nitens* in Australia, and Rooihoogte is the site where the South African parasitoids were collected for this study. Bioclim variables were obtained from WorldClim 2 (Fick & Hijmans, 2017) using the 'geodata' package in R, and averaged for an area with a 5km radius around the site.

site	latitude	longitude	bio1	bio5	bio6	bio10	bio11	remarks
Boonah (AU)	-27,9743	152,7208	19,4	30,3	5,8	24,0	13,9	AU population used in this study
Kwambonambi (ZA)	-28,5987	32,0904	21,5	29,3	12,0	24,7	18,0	Zululand, intended release area
Penola (AU)	-37,3755	140,8366	14,2	27,0	5,3	18,8	9,9	Original collection site of A. nitens
Rooihoogte (ZA)	-26,0613	30,2709	14,6	23,7	1,9	18,0	10,0	ZA population used in this study
Explanation of the Bioclim variables:								
Bio1	Annual Mean Temperature							
Bio5	Max Temperature of Warmest Month							
Bio6	Min Temperature of Coldest Month							
Bio10	Mean Temperature of Warmest Quarter							
Bio11 Mean Temperature of Coldest Quarter								

## Supplementary figures:



**Figure S1:** Mean femur length of the mother parasitoids used for the different lineages in each generation. Different letters indicate significant differences (for F0: Kruskal-Wallis test;  $\chi^2 = 10.2986$ , df = 3, p = 0.02; subsequent Dunn's post-hoc tests with p <0.05. For F2: Kruskal-Wallis test;  $\chi^2 = 8.6234$ , df = 3, p = 0.03; subsequent Dunn's post-hoc tests with p <0.05). Lineages were compared separately within each generation; the significance letters only apply within a generation. The error bars show the standard error.



**Figure S2:** The proportion of *A. nitens* fathers which mated prior to the experiment is shown per lineage for each generation. Since mating usually happens quickly after a pair is put together, males that emerged together with females were assumed to have mated. Due to limitations in parasitoid and host egg capsule availability, those males were still used, but their mating status was recorded for later analysis.



**Figure S3**: Average development time of male and female offspring for the whole dataset combined. The difference was significant according to the Wilcoxon signed rank test (V= 4944.5, p<0.001 and effect size = 0.304). Parasitoids were reared at a constant temperature of 23°C and 95%RH with a light:dark cycle of 14:10 hours. The error bars show the standard error.



**Figure S4**: Estimates for each model term in the GLMs for sex ratio, shown with the 95% confidence intervals. The ZA $\Im xZA \clubsuit$  lineage serves as the reference level for the effect 'lineage', and the model estimates of the other lineages indicate the estimated difference from this lineage. If there is no real difference between two lineages, the estimate of the model term is expected to be close to zero. Meanwhile, the 95% confidence intervals provide an indication for the level of certainty about the observed value. The graph shows that in the F1 generation for the AU $\Im xZA \clubsuit$  lineage the value zero does not fall within the 95% confidence interval. This could indicate a true difference with the ZA $\Im xZA \clubsuit$  lineage, which was also suggested by the Chi-squared test to assess the significance of the GLM model terms (Table S6), but not confirmed by the subsequent post-hoc test (Table S7). If sex ratio were to be truly lower for this lineage, it would still not be a concern as it is not an indication of reproductive incompatibility, but instead, a lower sex ratio means more daughters are produced and fertilization is succesfull.



**Figure S5**: Estimates for each model term in the GLMs for development time of daughters (top row) and sons (bottom row), shown with the 95% confidence intervals. The ZA $\Im xZA \square$  lineage serves as the reference level for the effect 'lineage', and the model estimates of the other lineages indicate the estimated difference with this lineage. In the F0 generation the sons of the ZA $\Im xAU\square$  lineage were found to develop faster compared to the ZA $\Im xZA\square$  lineage (Table S8 & S9) which is also supported by the model estimate and the 95% confidence intervals; all values are smaller than zero. Furthermore the 95% confidence intervals suggest that the sons of the AU $\Im xAU\square$  lineage also develop faster compared to the ZA $\Im xZA\square$  lineage, but this was not supported by the GLM analysis (Table S9). Note that the x-axes all have the same scale.



**Figure S6**: Estimates for each model term in the linear models for fecundity in terms of production of daughters (top row), sons (middle row) and total offspring (bottom row), shown with the 95% confidence intervals. The ZA $\Im xZA \square$  lineage serves as the reference level for the effect 'lineage', and the model estimates of the other lineages indicate the estimated difference of those lineages from the ZA $\Im xZA \square$  lineage by the model. No discrepancies with the other analyses for fecundity were found. Note that the x-axes have different scales.



**Figure S7**: Predicted relationship between the size of a mother parasitoid with total (A) and female (B) offspring production in the F1 generation. The estimated marginal means were calculated using the *emmeans* package from the linear models, and the 95% confidence intervals are shown with the shaded areas.