<u>Supplementary material for:</u> "Non-linear selection and the evolution of variances and covariances for continuous characters in an anole."

Appendix S1: Methods and Results for canonical analyses of multivariate selection

To canonically analyses our estimated multivariate selection surface we use the approach described in Blows & Brooks (2003; also see references therein). We first computed  $\Lambda$  and  $\mathbf{M}$  such that  $\Lambda = \mathbf{M}' \gamma \mathbf{M}$ . Here  $\mathbf{M}$  and  $\Lambda$  contain the normalized eigenvectors (in columns) and eigenvalues of  $\gamma$ , respectively. Now the eigenvectors of  $\mathbf{M}$  are independent dimensions of maximum curvature on the selection surface, and the eigenvalues of  $\Lambda$  are standardized coefficients of quadratic selection on each independent dimension given by  $\mathbf{M}$ . These coefficients ( $\lambda_1, \lambda_2, \ldots, \lambda_m$  for *m* traits) can be interpreted in the same way as standard quadratic coefficients (i.e.,  $\lambda_i < 0$  indicates disruptive selection on the trait axis given by the *i*th column of  $\mathbf{M}$ , henceforward  $\mathbf{m}_i$ ; and vice versa).

We evaluated the type I error probability of each diagonal coefficient in  $\Lambda$  by first rotating the data for each individual (z) into the eigenspace defined by **M**, using  $\mathbf{y} = \mathbf{Mz}$ . We then permuted the fitness values for individuals randomly 9,999 times. For each permutation we fit the model  $w = \alpha + \mathbf{\theta}\mathbf{y} + \frac{1}{2}\mathbf{y}'\Lambda\mathbf{y} + \varepsilon$  using least-squares. We computed the type I error probability (i.e., significance) by counting the fraction of  $\lambda_{ii}$  (diagonal of the permutation estimate of  $\Lambda$ ) in the randomization distribution that had an absolute value larger than our estimated value for  $\lambda_{ii}$  obtained from the original data.

Alternatively, we used the selection differential method. In this case, to obtain P-values we also rotated our size-corrected morphological measures into the space defined by **M**, i.e., for

1

each individual we computed  $\mathbf{y} = \mathbf{Mz}$ . We then randomized fitnesses among individuals 9,999 times, and each time computed rotated variance covariance matrices  $\mathbf{P}_{rot}$  (from all individuals) and  $\mathbf{P}_{rot}^*$  (calculated from only individuals with fitness values of 1.0 in the permutation). We then estimated the selection differential for  $\mathbf{P}$ ,  $\mathbf{C} = \mathbf{P}_{rot}^* - \mathbf{P}_{rot} + \mathbf{ss'}$ . Finally, we estimated the rotated curvature matrix using  $\mathbf{\Lambda} = \mathbf{P}_{rot}^{-1}\mathbf{CP}_{rot}^{-1}$  (Lande & Arnold, 1983). As before, we estimated the type I error as the fraction of the diagonal elements of  $\mathbf{\Lambda}$  in the permutation distribution with larger absolute values than their analogs in the original data.

Tables S1.1 to S1.4 give the eigenvectors (**M**) and eigenvalues of the  $\gamma$  matrices reported in the main text, as well as the type I error probabilities (P values) for each eigenvalue of  $\gamma$ , in which we estimated the P values using the permutation procedure described in the preceding paragraphs. In general we found relatively few independent multidimensional trait axes with significant stabilizing or disruptive selection. For example, in the regression analysis of external measurements with fitness assessed from mark-recapture information, we found no significant independent dimensions of disruptive or stabilizing selection. By contrast, in the regression model based on body condition and internal measures, we found significant disruptive selection on eigenvector  $\mathbf{m}_1$ . In this analysis the trait axis  $\mathbf{m}_1$  represents positive changes in jaw length and head width (or vice versa), associated with negative changes in forelimb length and pelvis width (or vice versa). This analysis also revealed marginally non-significant stabilizing selection on eigenvector  $\mathbf{m}_5$  (Table S1.2).

The canonical analysis of the selection differential method results was similarly ambiguous. The selection differential analysis performed with the external measures yield no significant axes of quadratic selection, although eigenvector  $\mathbf{m}_1$  (mostly head width) was marginally non-significant for weak disruptive selection. There was one significant independent axis of quadratic selection in the differential analysis based on internal measures, this one ( $\mathbf{m}_2$ : positive changes on hindlimb associated with negative changes on all other characters, or vice versa) indicated significant disruptive selection.

**Table S1.1** Canonical analysis of  $\gamma$  from the quadratic regression selection analysis on external data.  $\mathbf{m}_i$  is the *i*th column of  $\mathbf{M}$ , the matrix of eigenvectors in the canonical analysis of the curvature of the selection surface,  $\gamma$  (Table S2.3). The values for  $\lambda$  are the diagonal elements the eigenvalue matrix  $\mathbf{\Lambda}$ . P values were computed as described in the supplementary Appendix S1 text.

| Eigenvector \<br>Trait | $\mathbf{m}_1$ | $\mathbf{m}_2$ | <b>m</b> <sub>3</sub> | $\mathbf{m}_4$ |
|------------------------|----------------|----------------|-----------------------|----------------|
| hindlimb               | 0.3403         | 0.7705         | -0.1292               | 0.5234         |
| forelimb               | -0.5997        | 0.5028         | -0.4246               | -0.4552        |
| jaw length             | -0.2248        | -0.3857        | -0.7154               | 0.5374         |
| head width             | -0.6885        | 0.0687         | 0.5396                | 0.4797         |
| λ                      | 0.3994         | 0.0148         | -0.0896               | -0.4169        |
| P(permutation)         | 0.2182         | 0.8863         | 0.8911                | 0.1450         |

**Table S1.2** Canonical analysis of  $\gamma$  from the quadratic regression selection analysis on internal (x-ray) data. The quadratic selection matrix,  $\gamma$ , for this analysis is given in Table 3 of the main text.  $\mathbf{m}_i$  and  $\lambda$  are as in Table S1.1.

| Eigenvector \<br>Trait | $\mathbf{m}_1$ | $\mathbf{m}_2$ | <b>m</b> <sub>3</sub> | $\mathbf{m}_4$ | <b>m</b> <sub>5</sub> |
|------------------------|----------------|----------------|-----------------------|----------------|-----------------------|
| hindlimb               | 0.0277         | 0.4948         | -0.8600               | -0.1009        | -0.0683               |
| forelimb               | 0.1933         | -0.7150        | -0.4720               | 0.3491         | 0.3266                |
| jaw length             | -0.5675        | -0.4532        | -0.1932               | -0.3550        | -0.5561               |
| head width             | -0.6836        | 0.1963         | 0.0018                | 0.6969         | 0.0921                |
| pelvis width           | 0.4154         | 0.0037         | 0.0161                | 0.5062         | -0.7556               |
| λ                      | 0.2172         | 0.0097         | -0.0030               | -0.1664        | -0.2249               |
| P(permutation)         | 0.0283         | 0.8787         | 0.7308                | 0.1090         | 0.0903                |

**Table S1.3** Canonical analysis of  $\gamma$  from the selection differential analysis on external data. The quadratic selection matrix,  $\gamma$ , for this analysis is given in Table 4 of the main text.  $\mathbf{m}_i$  and  $\lambda$  are

| Eigenvector \<br>Trait | $\mathbf{m}_1$ | $\mathbf{m}_2$ | <b>m</b> <sub>3</sub> | $\mathbf{m}_4$ |
|------------------------|----------------|----------------|-----------------------|----------------|
| hindlimb               | 0.1436         | -0.0224        | 0.6757                | 0.7227         |
| forelimb               | -0.0801        | -0.0465        | 0.7345                | -0.6722        |
| jaw length             | 0.3202         | -0.9447        | -0.0592               | -0.0375        |
| head width             | 0.9330         | 0.3237         | -0.0206               | -0.1561        |
| λ                      | 0.9685         | 0.1554         | -0.0176               | -0.1167        |
| P(permutation)         | 0.0908         | 1.0000         | 0.9644                | 0.4875         |

**Table S1.4** Canonical analysis of  $\gamma$  from the selection differential analysis on internal data. The quadratic selection matrix,  $\gamma$ , for this analysis is given in Table 5 of the main text.  $\mathbf{m}_i$  and  $\lambda$  are

| Eigenvector \<br>Trait | $\mathbf{m}_1$ | <b>m</b> <sub>2</sub> | <b>m</b> <sub>3</sub> | $\mathbf{m}_4$ | <b>m</b> <sub>5</sub> |
|------------------------|----------------|-----------------------|-----------------------|----------------|-----------------------|
| hindlimb               | 0.2443         | -0.2379               | 0.2478                | 0.8830         | -0.2067               |
| forelimb               | -0.8237        | 0.3576                | -0.2238               | 0.3577         | -0.1251               |
| jaw length             | 0.4006         | 0.4741                | -0.5393               | 0.2837         | 0.4933                |
| head width             | 0.0669         | 0.6687                | 0.7317                | -0.0173        | 0.1130                |
| pelvis width           | 0.3113         | 0.3790                | -0.2495               | -0.1078        | -0.8280               |
| λ                      | 3.7093         | 0.9927                | 0.8039                | -0.0133        | -0.8863               |
| P(permutation)         | 0.4225         | 0.0013                | 0.3879                | 0.3984         | 0.5555                |

as in Tables S1.1, S1.2, and S1.3.

as in Tables S1.1 and S1.2.

## Appendix S2: Supplementary tables

**Table S2.1** Eigenvectors  $(\mathbf{p}_i)$  and eigenvalues  $(\lambda_i)$  of  $\mathbf{P}_{ex}$ , the phenotypic variance covariance

|             | Eigenvectors of $\mathbf{P}_{ex}$ |                       |                       |                |  |  |  |
|-------------|-----------------------------------|-----------------------|-----------------------|----------------|--|--|--|
| Traits      | $\mathbf{p}_{max}$                | <b>p</b> <sub>2</sub> | <b>p</b> <sub>3</sub> | $\mathbf{p}_4$ |  |  |  |
| hindlimb    | 0.8729                            | 0.3888                | 0.2939                | 0.0201         |  |  |  |
| forelimb    | 0.4854                            | -0.7291               | -0.4694               | -0.1119        |  |  |  |
| jaw length  | 0.0199                            | 0.5584                | -0.7786               | -0.2857        |  |  |  |
| head width  | 0.0446                            | 0.0737                | -0.2952               | 0.9516         |  |  |  |
| $\lambda_i$ | 3.3410                            | 0.6234                | 0.4369                | 0.1482         |  |  |  |
| % variance  | 73.44                             | 13.70                 | 9.60                  | 3.26           |  |  |  |

matrix calculated from external measurements.

**Table S2.2** Eigenvectors  $(\mathbf{p}_i)$  and eigenvalues  $(\lambda_i)$  of  $\mathbf{P}_{in}$ , the phenotypic variance covariance

matrix calculated from internal x-ray measurements.

|              |                    | Eig                   | genvectors of         | $\mathbf{P}_{ex}$ |                       |
|--------------|--------------------|-----------------------|-----------------------|-------------------|-----------------------|
| Traits       | $\mathbf{p}_{max}$ | <b>p</b> <sub>2</sub> | <b>p</b> <sub>3</sub> | $\mathbf{p}_4$    | <b>p</b> <sub>5</sub> |
| hindlimb     | 0.8805             | -0.2835               | 0.3768                | 0.0469            | 0.0139                |
| forelimb     | 0.4477             | 0.2414                | -0.8533               | -0.0614           | -0.0967               |
| jaw length   | 0.1206             | 0.7942                | 0.2481                | 0.5414            | 0.0080                |
| head width   | 0.0786             | 0.4454                | 0.2613                | -0.7856           | -0.3317               |
| pelvis width | 0.0599             | 0.1798                | -0.0033               | -0.2893           | 0.9383                |
| $\lambda_i$  | 1.8068             | 0.2762                | 0.1488                | 0.1038            | 0.0629                |
| % variance   | 75.33              | 11.51                 | 6.20                  | 4.33              | 2.62                  |

 Table S2.3 Quadratic regression selection analysis; external data.

| <b>γ-</b> matrix | hindlimb | forelimb | jaw length | head width | <b>β-</b> vector |
|------------------|----------|----------|------------|------------|------------------|
| hindlimb         | -0.0607  | 0.0186   | -0.1605    | -0.1912    | -0.0617          |
| forelimb         |          | 0.0448   | 0.1258     | 0.2770     | 0.0437           |
| jaw length       |          |          | -0.1439    | -0.0115    | -0.3418*         |
| head width       |          |          |            | 0.0674     | 0.3196           |

\*<0.05

**Table S2.4** Eigenvectors and eigenvalues of  $-\gamma^{-1}$  and  $\gamma$ . Left-right rank order of the

eigenvectors is based on  $-\gamma^{-1}$ .  $\omega_i$  is the *i*th eigenvector of  $-\gamma^{-1}$  due to  $\omega \approx -\gamma^{-1}$  under weak stabilizing selection (Lande, 1979). External data; least-squares regression selection analysis.

|                              | E                           | Eigenvectors of $-\gamma^{-1}$ and $\gamma$ |                           |                       |  |  |  |
|------------------------------|-----------------------------|---|---------------------------|-----------------------|--|--|--|
| Traits                       | $\omega_{\rm max}/\gamma_3$ | $\omega_2^{}/\gamma_4^{}$                   | $\omega_3 / \gamma_{max}$ | $\omega_4 / \gamma_2$ |  |  |  |
| hindlimb                     | -0.1292                     | 0.5234                                      | 0.3403                    | 0.7705                |  |  |  |
| forelimb                     | -0.4246                     | -0.4552                                     | -0.5997                   | 0.5028                |  |  |  |
| jaw length                   | -0.7154                     | 0.5374                                      | -0.2248                   | -0.3857               |  |  |  |
| head width                   | 0.5396                      | 0.4797                                      | -0.6885                   | 0.0687                |  |  |  |
| $\lambda_i(-\gamma^{-1})$    | 11.158                      | 2.3984                                      | -2.5039                   | -67.724               |  |  |  |
| $\lambda_i(\mathbf{\gamma})$ | -0.0896                     | -0.4169                                     | 0.3994                    | 0.0148                |  |  |  |

**Table S2.5** Eigenvectors and eigenvalues of  $-\gamma^{-1}$  and  $\gamma$ . Rank order of the eigenvectors is

based on  $-\gamma^{-1}$ . Internal data; least-squares regression selection analysis.

|                              | Eigenvectors of $-\gamma^{-1}$ and $\gamma$ |                       |                     |                           |                     |  |
|------------------------------|---|-----------------------|---------------------|---------------------------|---------------------|--|
| Traits                       | $\omega_{\rm max}/\gamma_3$                 | $\omega_2 / \gamma_4$ | $\omega_3/\gamma_5$ | $\omega_4 / \gamma_{max}$ | $\omega_5/\gamma_2$ |  |
| hindlimb                     | -0.8600                                     | 0.1009                | 0.0683              | -0.0277                   | -0.4948             |  |
| forelimb                     | -0.4720                                     | -0.3491               | -0.3266             | -0.1933                   | 0.7150              |  |
| jaw length                   | -0.1932                                     | 0.3550                | 0.5561              | 0.5675                    | 0.4532              |  |
| head width                   | 0.0018                                      | -0.6969               | -0.0921             | 0.6836                    | -0.1963             |  |
| pelvis width                 | 0.0161                                      | -0.5062               | 0.7556              | -0.4154                   | -0.0037             |  |
| $\lambda_i(-\gamma^{-1})$    | 331.79                                      | 6.0083                | 4.4468              | -4.6048                   | -103.45             |  |
| $\lambda_i(\mathbf{\gamma})$ | -0.0030                                     | -0.1664               | -0.2249             | 0.2172                    | 0.0097              |  |

-0.0176

 $\lambda_i(\mathbf{\gamma})$ 

| based on $-\gamma^{-1}$   | . External d                | ata; selectior                              | n differential            | analysis.             |  |  |
|---------------------------|-----------------------------|---|---------------------------|-----------------------|--|--|
|                           | F                           | Eigenvectors of $-\gamma^{-1}$ and $\gamma$ |                           |                       |  |  |
| Traits                    | $\omega_{\rm max}/\gamma_3$ | $\omega_2 / \gamma_4$                       | $\omega_3 / \gamma_{max}$ | $\omega_4 / \gamma_2$ |  |  |
| hindlimb                  | -0.6757                     | -0.7227                                     | -0.1436                   | -0.0224               |  |  |
| forelimb                  | -0.7345                     | 0.6722                                      | 0.0801                    | -0.0465               |  |  |
| jaw length                | 0.0592                      | 0.0375                                      | -0.3202                   | -0.9447               |  |  |
| head width                | 0.0206                      | 0.1561                                      | -0.9330                   | 0.3237                |  |  |
| $\lambda_i(-\gamma^{-1})$ | 56.889                      | 8.5713                                      | -1.0325                   | -6.4358               |  |  |

-0.1167

**Table S2.7** Eigenvectors and eigenvalues of  $-\gamma^{-1}$  and  $\gamma$ . Rank order of the eigenvectors is

0.9685

0.1554

**Table S2.6** Eigenvectors and eigenvalues of  $-\gamma^{-1}$  and  $\gamma$ . Rank order of the eigenvectors is

based on  $-\gamma^{-1}$ . Internal data; selection differential analysis.

|                              |                         | Eigenvectors of $-\gamma^{-1}$ and $\gamma$ |                           |                     |                         |  |  |
|------------------------------|-------------------------|---|---------------------------|---------------------|-------------------------|--|--|
| Traits                       | $\omega_{max}/\gamma_4$ | $\omega_2/\gamma_5$                         | $\omega_3 / \gamma_{max}$ | $\omega_4/\gamma_2$ | $\omega_{5}/\gamma_{3}$ |  |  |
| hindlimb                     | -0.8830                 | -0.2067                                     | -0.2443                   | -0.2379             | 0.2478                  |  |  |
| forelimb                     | -0.3577                 | -0.1251                                     | 0.8237                    | 0.3576              | -0.2238                 |  |  |
| jaw length                   | -0.2837                 | 0.4933                                      | -0.4006                   | 0.4741              | -0.5393                 |  |  |
| head width                   | 0.0173                  | 0.1130                                      | -0.0669                   | 0.6687              | 0.7317                  |  |  |
| pelvis width                 | 0.1078                  | -0.8280                                     | -0.3113                   | 0.3790              | -0.2495                 |  |  |
| $\lambda_i(-\gamma^{-1})$    | 74.986                  | 1.1282                                      | -0.2696                   | -1.0074             | -1.2440                 |  |  |
| $\lambda_i(\mathbf{\gamma})$ | -0.0133                 | -0.8863                                     | 3.7093                    | 0.9927              | 0.8039                  |  |  |

| Eigenvectors of <b>P</b> | Eigenvalues of $\mathbf{P}(\lambda_i)$ | Selection on eigenvector $(\mathbf{p}'_i \mathbf{\gamma} \mathbf{p}_i)$ |              |  |
|--------------------------|--|---|--------------|--|
|                          |  | Least-squares   | Differential |  |
| <b>p</b> <sub>max</sub>  | 3.3410                                 | -0.0259   | -0.0081      |  |
| <b>p</b> <sub>2</sub>    | 0.6234                                 | -0.2542   | 0.0971       |  |
| <b>p</b> <sub>3</sub>    | 0.4369                                 | 0.1881  | 0.2158       |  |
| $\mathbf{p}_4$           | 0.1482                                 | -0.0005   | 0.6849       |  |

**Table S2.8** Analysis using the method of Hunt *et al.* (2007); external data.

Table S2.9 Analysis using the method of Hunt et al. (2007); internal (x-ray) data.

| Eigenvectors of <b>P</b> | Eigenvalues of <b>P</b> $(\lambda_i)$ | Selection on eigenvector $(\mathbf{p}'_i \gamma \mathbf{p}_i)$ |              |
|--------------------------|---------------------------------------|--|--------------|
|                          |                                       | Least-squares  | Differential |
| <b>p</b> <sub>max</sub>  | 1.8068                                | -0.0049  | -0.0162      |
| <b>p</b> <sub>2</sub>    | 0.2762                                | 0.0405   | 0.8334       |
| <b>p</b> <sub>3</sub>    | 0.1488                                | 0.0055   | 3.1517       |
| $\mathbf{p}_4$           | 0.1038                                | -0.1425  | 0.5587       |
| <b>p</b> <sub>5</sub>    | 0.0629                                | -0.0661  | 0.0785       |

**Table S2.10** Variability of **P** evaluated over the eigenvectors of  $\gamma$ ; external data.

| Eigenvectors of $\gamma$ | Least-squares                   |   | Selection differential          |   |
|--------------------------|---------------------------------|---|---------------------------------|---|
|                          | $\gamma\left(\lambda_{i} ight)$ | $\mathbf{\gamma}_i' \mathbf{P} \mathbf{\gamma}_i$ | $\gamma\left(\lambda_{i} ight)$ | $\mathbf{\gamma}_i' \mathbf{P} \mathbf{\gamma}_i$ |
| $\gamma_{\rm max}$       | 0.3994                          | 0.3911  | 0.9685                          | 0.3253  |
| $\gamma_2$               | 0.0148                          | 2.8616  | 0.1554                          | 0.3878  |
| $\gamma_3$               | -0.0896                         | 0.5481  | -0.0176                         | 3.0423  |
| $\gamma_4$               | -0.4169                         | 0.7487  | -0.1167                         | 0.7941  |

| Eigenvectors $_{\rm of  \gamma}$ | Least-squares       |   | Selection differential |   |
|----------------------------------|---------------------|---|------------------------|---|
|                                  | $\gamma(\lambda_i)$ | $\mathbf{\gamma}_i' \mathbf{P} \mathbf{\gamma}_i$ | $\gamma(\lambda_i)$    | $\mathbf{\gamma}_i' \mathbf{P} \mathbf{\gamma}_i$ |
| $\gamma_{\rm max}$               | 0.2172              | 0.1708  | 3.7093                 | 0.1504  |
| $\gamma_2$                       | 0.0097              | 0.1970  | 0.9927                 | 0.2541  |
| $\gamma_3$                       | -0.0030             | 1.7754  | 0.8039                 | 0.1288  |
| $\gamma_4$                       | -0.1664             | 0.1345  | -0.0133                | 1.6864  |
| $\gamma_5$                       | -0.2249             | 0.1206  | -0.8863                | 0.1787  |

**Table S2.11** Variability of **P** evaluated over the eigenvectors of  $\gamma$ ; internal (x-ray) data.