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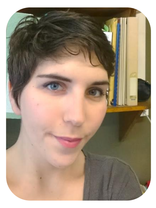
Testosterone enhances attractive sexual signals but no immunosuppression



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Background: Sexual selection

Female mate choice ➡ Male sexual signals = **honest** and **reliable** signals of male quality [1]

The **immunocompetence Handicap Hypothesis** (ICHH) [2] could explain the signal honesty

Testosterone

\- ↙ Immunity

\+ ↘ Sexual signal expression

The ICHH has never been tested in a **multimodal communication system**.

Aim: Testing the ICHH in *Hyla arborea*

Testosterone?

→ Acoustic signals (vocalize performance)

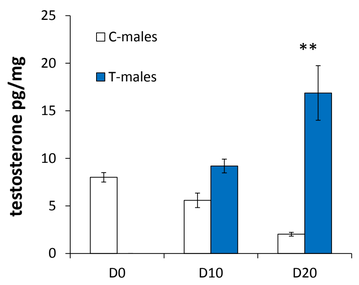
→ Visual signals (Vocal sac coloration)

→ Immune response capacity

1/ are acoustic signals testosterone-dependant?
2/ are visual signals testosterone-dependant?
3/ is testosterone immunosuppressive?

Material & methods:

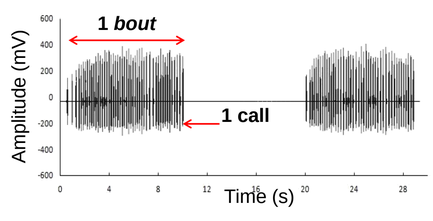
Testosterone supplementation: daily topical application (20 days). Measures of testosterone levels in saliva. N=48 T-males Vs N=48 C-males



*Variations during the 20 days of the experiment of the testosterone levels in saliva, ** P<0,01*

4h call recording per tree frogs:

Acoustic parameters:
- within-bout call rate (in call/s)
- Bout duration (in s)
- Dominant frequency (in Hz)



Coloration measurements:

Vocal sac colour parameters:
- Chroma (more or less saturation)
- Brightness (more or less darker)

Using a spectrophotometer with a optic fibre light (SpectraSuite®). Analyses with AVICOL®

Frog manipulation during the colour measure of the vocal sac. with the spectrophotometer.

Immunity test:

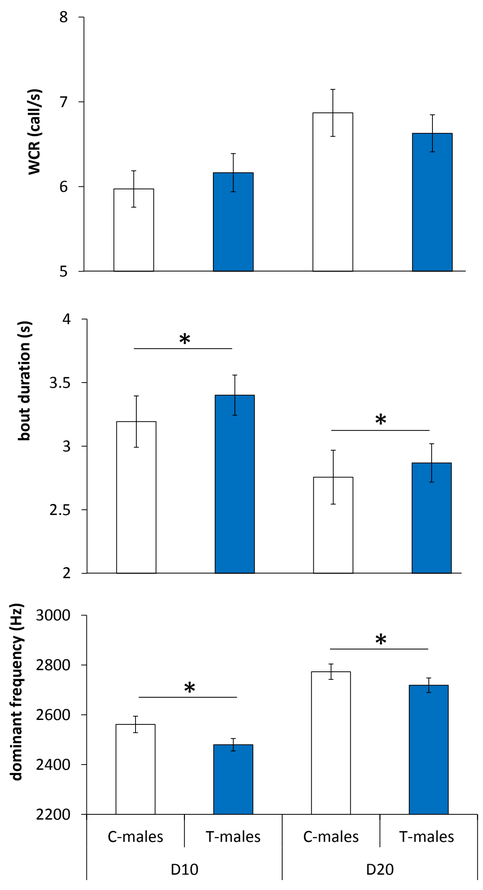
Injection of phytohaemagglutinin (PHA) in the leg. [3]

Measure of the leg thickness before and 18h after injection. Analyse of the leg swelling (mm)

Results:

1/ Testosterone effects on acoustic signals

GEE analysis: *Call parameter ~ body mass + day + treatment + body mass X treatment + day X treatment*

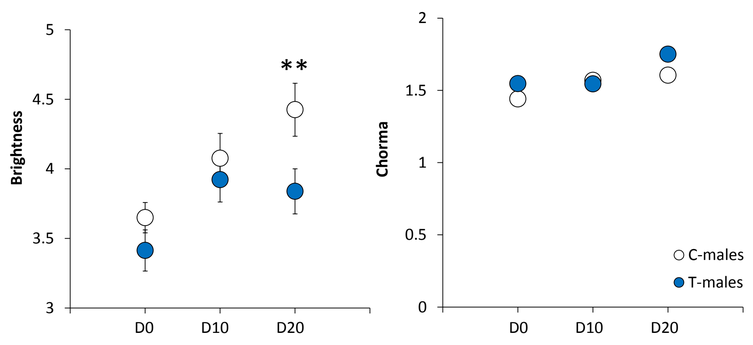


- **Testosterone** supplementation **increases the bout duration** *(pv<0,05)* and **decrease the dominant frequency** *(pv<0,05)*
- More the male is bigger more its **dominant frequency is lower** *(pv<0,001)* and its **bout duration is longer** *(pv<0,001)*
- There is a significant "Day effect" for all acoustic parameters *(pv<0,001)*

*Variations during the 20 days of the experiment of the call parameters WCR (within bout call rate) bout duration and dominant frequency, means +/- SEM, * P<0,05*

2/ Testosterone effects on visual signals

GEE analysis: *colour parameter ~ body mass + day + treatment + body mass X treatment + day X treatment*

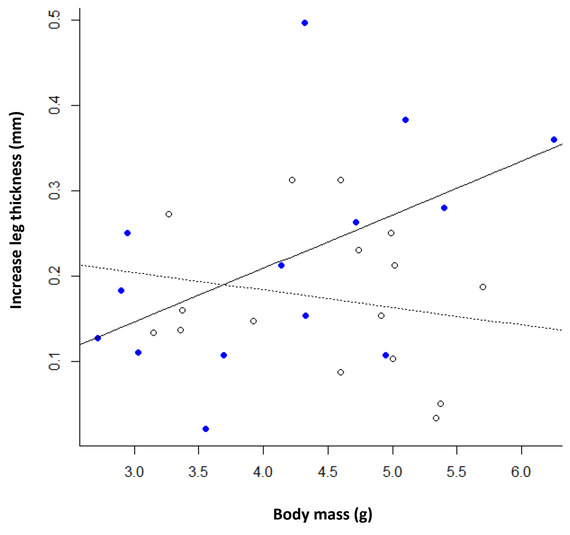


- **Testosterone** supplementation has no effect on chroma parameter but limits the leak of colour (brightening) in time. **After 20 days, T-males are darker than C-males** *(pv<0,01)*.
- More the male is bigger more its vocal sac **is darker** *(pv<0,01)*
- There is a significant "Day effect" for all colour parameters *(pv<0,05)*

*Variations during the 20 days of the experiment of the colour parameters the brightness and the chroma, means +/- SEM, * *P<0,01*

3/ Testosterone effects on immune response

Linear mixed model: *immune response ~ body mass + treatment + body mass X treatment*



- More the leg swelling is important more the immune response is important. **The immune response depends on the body mass of the male**.
- T-males with less body mass and C-males react in a similar manner.
- **Testosterone** supplementation: the strength of the swelling response **increased significantly for those T-males with a high body mass**. *(body mass X treatment, pv=0,05)*

Effect of the interaction between treatment and body mass on leg thickness (in mm) after PHA injection (N=30). Prediction of the linear model for the significant interaction between the two explanatory variables are represented (full and dotted lines for T-males (blue circles) and C-males (white circle), respectively)

Conclusions:

The originality of this study relies on the **measurements of multimodal signals** in the context of ICHH.

As predicted by the ICHH, our study reveals that several components of **acoustic signal and visual signal are testosterone-dependant.** Nevertheless, our results did **not reveal an immunosuppression effect of the testosterone** and thus, we did **not validate the ICHH** as a proximate mechanism explaining signal honesty in *Hyla arborea*. Surprisingly, the interaction between treatment and body mass on the immune response suggests **an immunostimulation in T-males greater than 4,5g**. The interaction of testosterone treatment and body mass effects **could be an indirect consequence of energy reallocation** from the immune system **to other metabolic demands** [4]
An integrative approach is now necessary to improve our understanding of the link between immunocompetence and sexual selection.

References:

[1] Zahavi A, 1977. Cost of Honesty - (Further Remarks on Handicap Principle). J Theor Biol 67:603-605.
[2] Folstad I, Karter AJ, 1992. Parasites, Bright Males, and the Immunocompetence Handicap. Am Nat 139:603-622.
[3] Josserand R, Brepson L, **Desprat JL.**, Lengagne T, Mondy N. 2015. A phytohaemaglutinin challenge test to assess immunocompetence of European tree frog *Hyla arborea*. *Amphibia-Reptilia*, DOI: 10.1163/15685381-00002983
[4] Alonso-Alvarez C, Bertrand S, Sorci G, 2007. Energetic reserves, leptin and testosterone: a refinement of the immunocompetence handicap hypothesis. Biol Lett 3:271-274.
Desprat JL, Lengagne T, Dumet A, Desouhant E, Mondy, N. 2015. Immunocompetence handicap hypothesis in tree frog: trade off between sexual signals and immunity? *Behavioural Ecology*. DOI: 10.1093/beheco/arv057

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