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*Publication date:*  
2018

*Document Version*  
Publisher's PDF, also known as Version of record

[Link back to DTU Orbit](#)

*Citation (APA):*  
Johansson, H. K. L., Nielsen, L. N., Vinggaard, A. M., Bahl, M. I., & Svingen, T. (2018). *Glyphosate alone does not adversely affect testicular androgen function in mature rats*. Poster session presented at 20th European Workshop on the Molecular and Cellular Endocrinology of the Testis, Obedos, Portugal.

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# Glyphosate alone does not adversely affect testicular androgen function in mature rats

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## Introduction

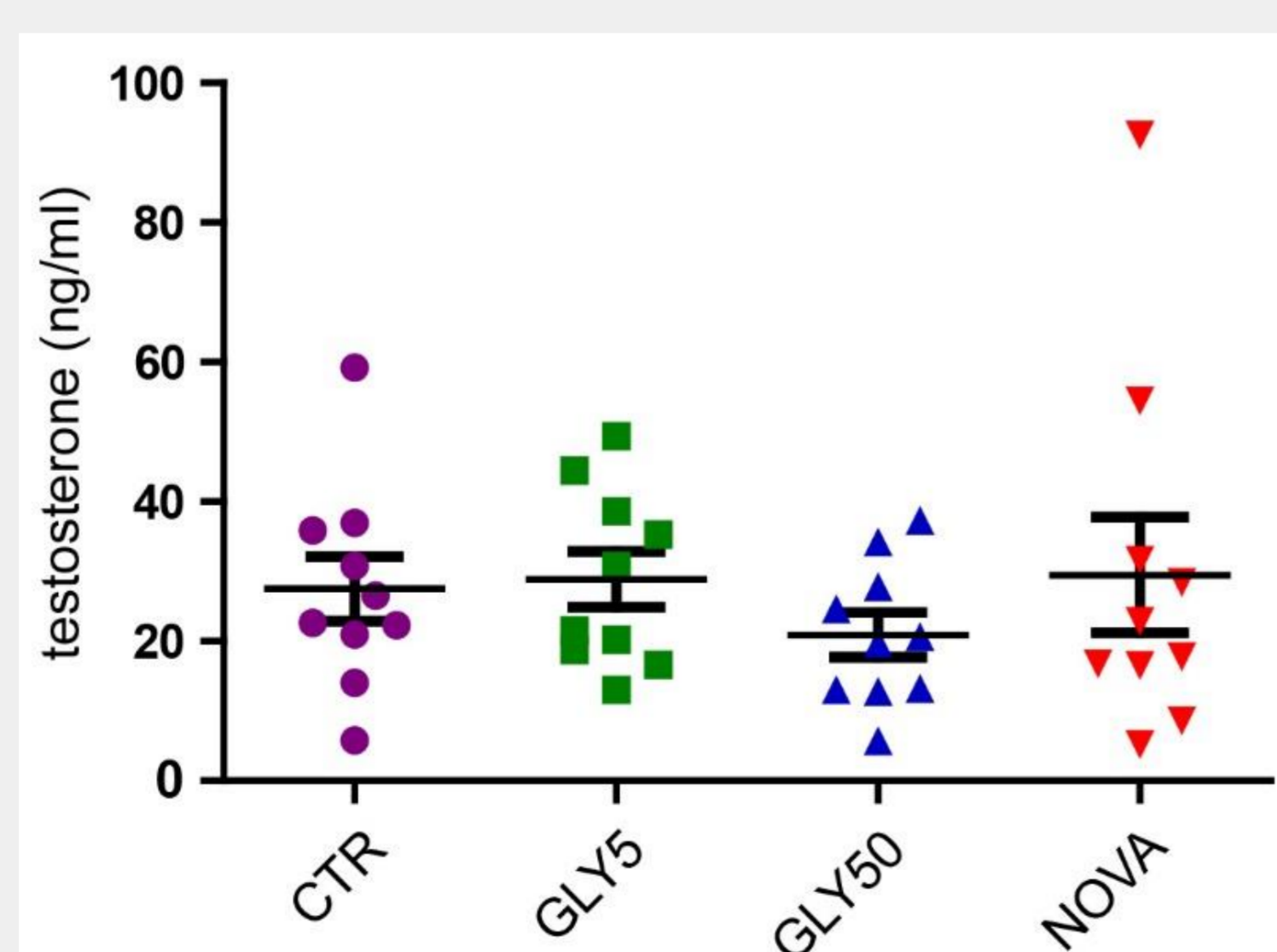
Glyphosate is the active ingredient in numerous herbicides used world-wide for weed control. Since its introduction to the market in the 1970s, it has grown to become one of the most used agricultural herbicide by volume. In USA alone, close to 100,000 tonnes was used in 2007 by agriculture, industry, and private users. There has also been an increased concern about potential harmful effects on humans and wildlife, including male reproductive health through disruption of endocrine signalling. Evidence are conflicting, however, with studies indicating disruption to testosterone synthesis, to no measurable effects. Here, we exposed male rats to glyphosate alone or in a commercial formulation (Glyphonova) and assessed if testosterone synthesis or testicular integrity was affected. Our data suggests that glyphosate itself has little to no effect on these parameters, but formulation adjuvants may affect testis function.

## Methods

- 4-week old male rats were orally exposed to 2.5 mg/kg/d (GLY5) or 25 mg/kg/day (GLY25) glyphosate, or 25 mg/kg/d Glyphonova-450 Plus (NOVA; 25 mg/kg/d equivalent dose of glyphosate) for two weeks.
- Exposure experiment was performed with 80 rats housed in pairs, with one rat per pair analysed as one biological unit (40 rats in total; N=10 per group).
- Testicles were collected from killed rats; one frozen for intra-testicular testosterone and gene expression analyses; one was fixed in formalin for histological assessments.

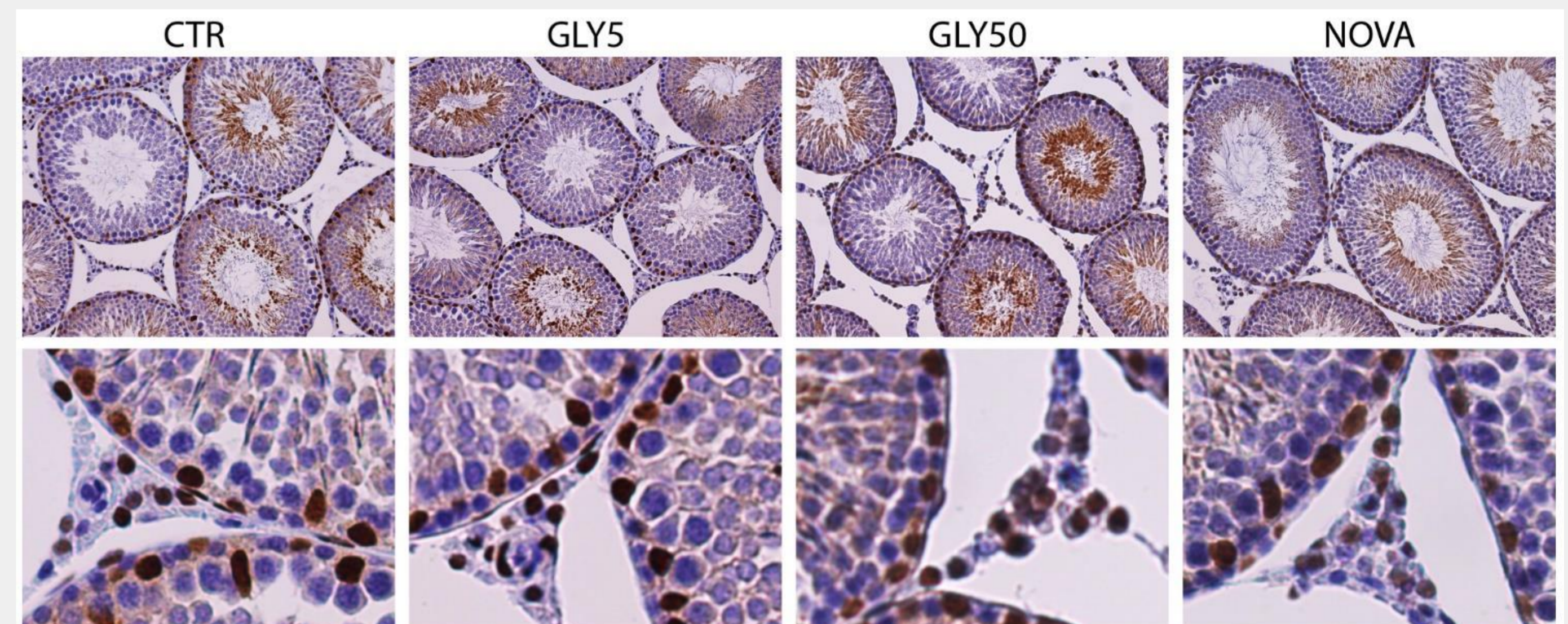


## Results – testosterone synthesis

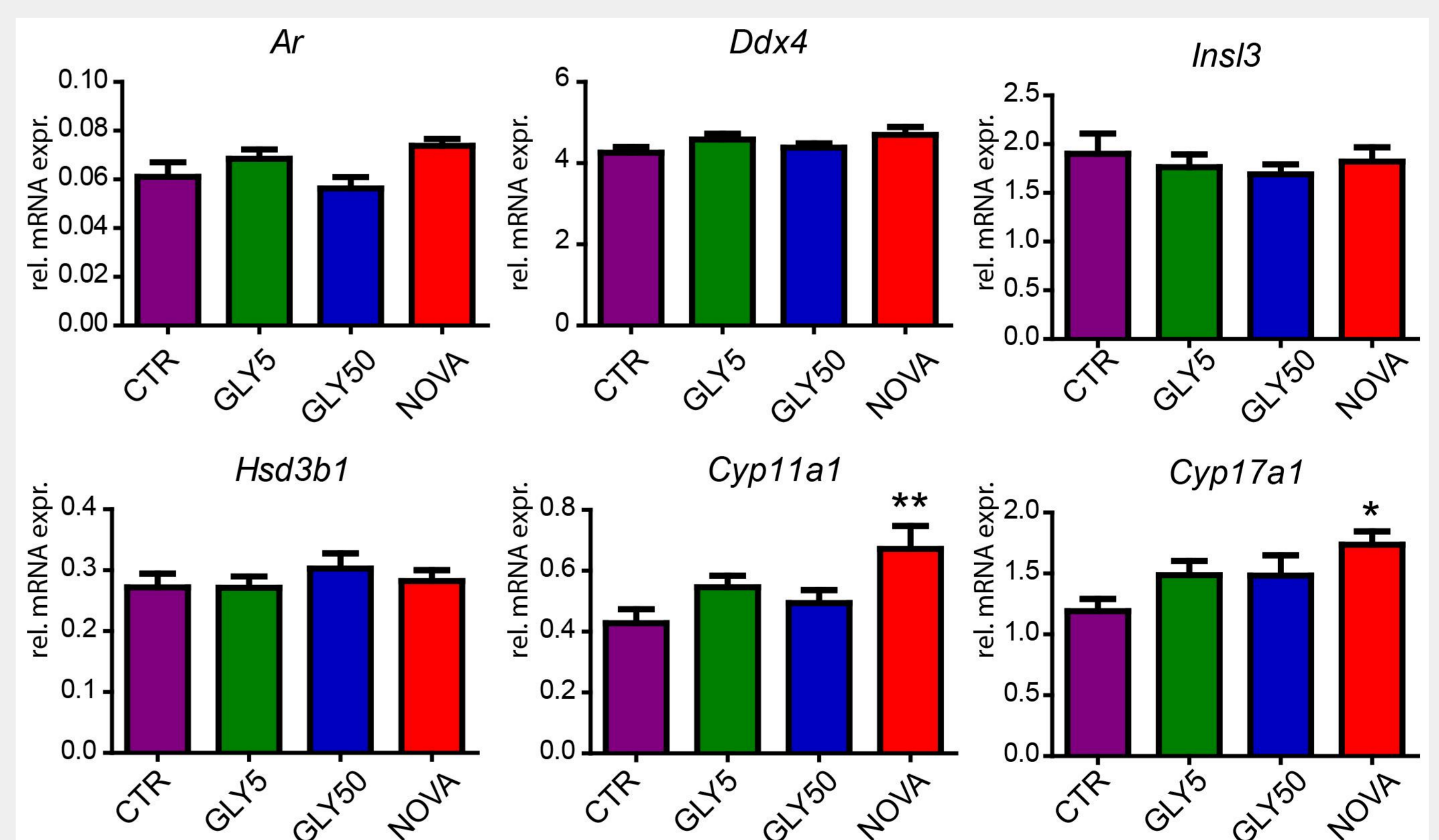


**Fig. 1:** Two weeks of exposure to 5x or 50x ADI glyphosate alone (GLY) or in Glyphonova formulation (NOVA) did not significantly alter intra-testicular testosterone synthesis. N=10 per group; Mean ± SEM.

## Results – histopathology & gene expression



**Fig. 2:** Exposure to glyphosate alone or in Glyphonova formulation (5 or 50x ADI), resulted in no obvious signs of histopathological changes or expression of the Androgen receptor (AR; brown) in adult rat testis. Tissue counterstained with haematoxylin. Representative images of N=4 per group.



**Fig. 3:** Exposure to 50x ADI glyphosate in Glyphonova formulation (NOVA), but not glyphosate alone (GLY), affected steroidogenic gene expression in adult Leydig cells. N=10 per group; Mean ± SEM; \*p<0.05, \*\*p<0.01 (ANOVA).

## Discussion

Exposure to glyphosate to 50x ADI for two weeks do not seem to adversely affect testicular function in young adult rats. Steroidogenic gene expression is affected following exposure to glyphosate in formulation only, indicating that it is the adjuvants, and not glyphosate, that are causative.