# A PATERNAL LEGACY: THE TRANSMISSION OF EDC EPIMUTATIONS TO THE FUTURE GENERATIONS

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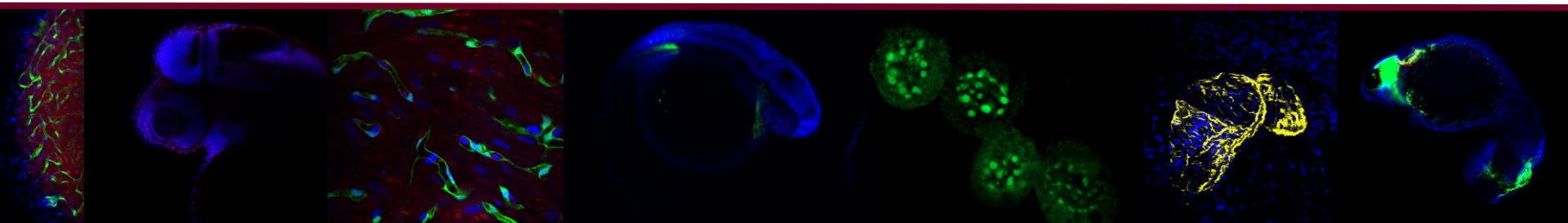
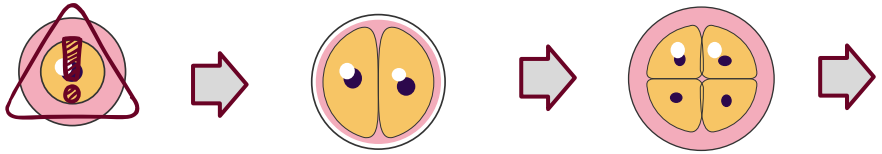
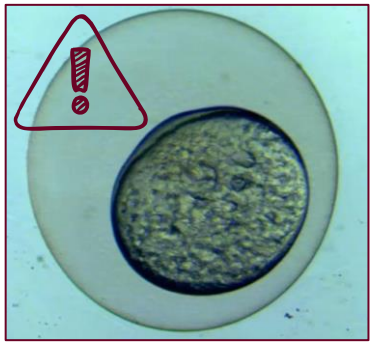
**Universidad de León, Spain**

**Rome, 17th November 2022**

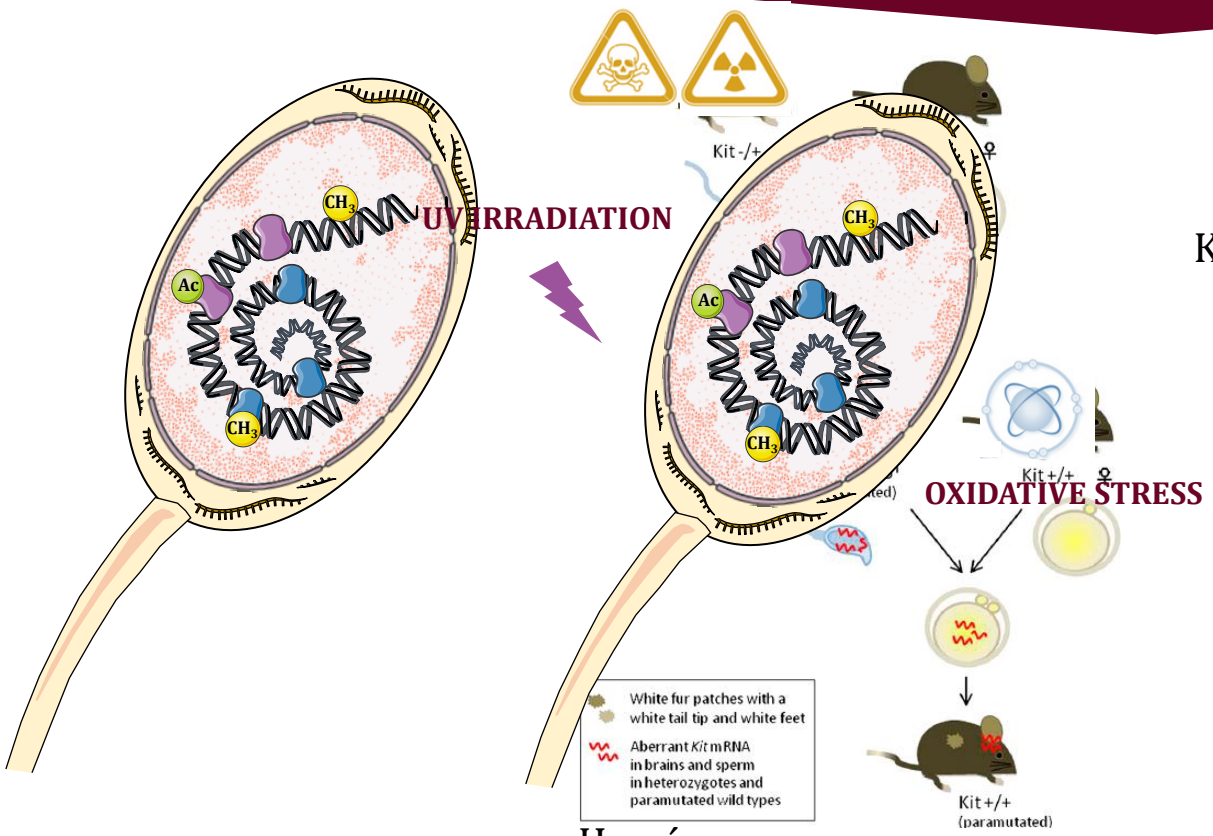


# INTRODUCTION



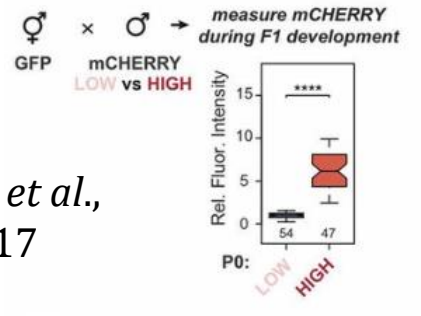


## Paternal contribution to embryo development

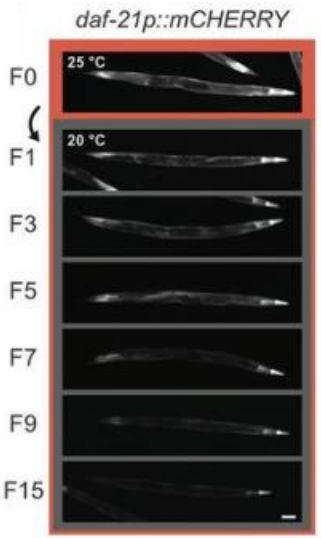



Herráez *et al.*, 2017

Hamatani *et al.*, 2011

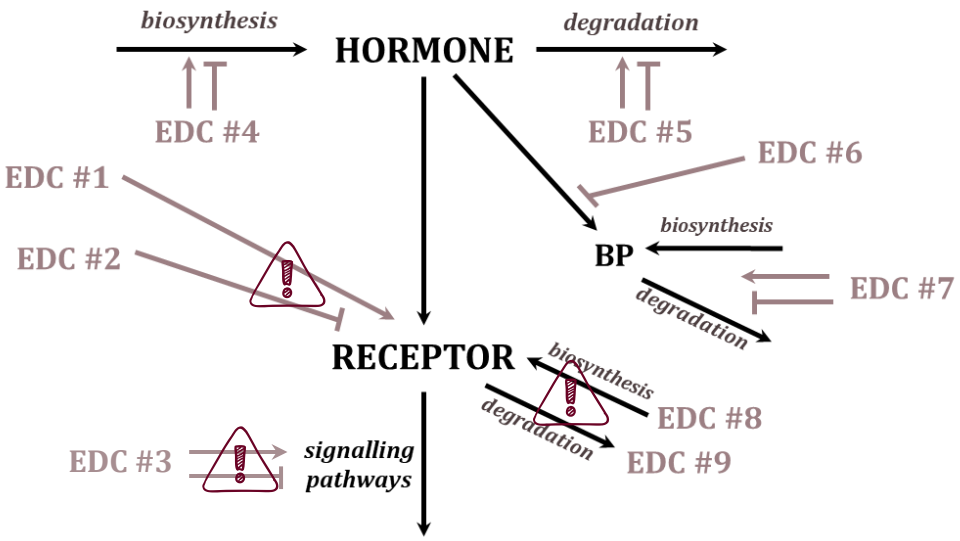



Klosin *et al.*, 2017

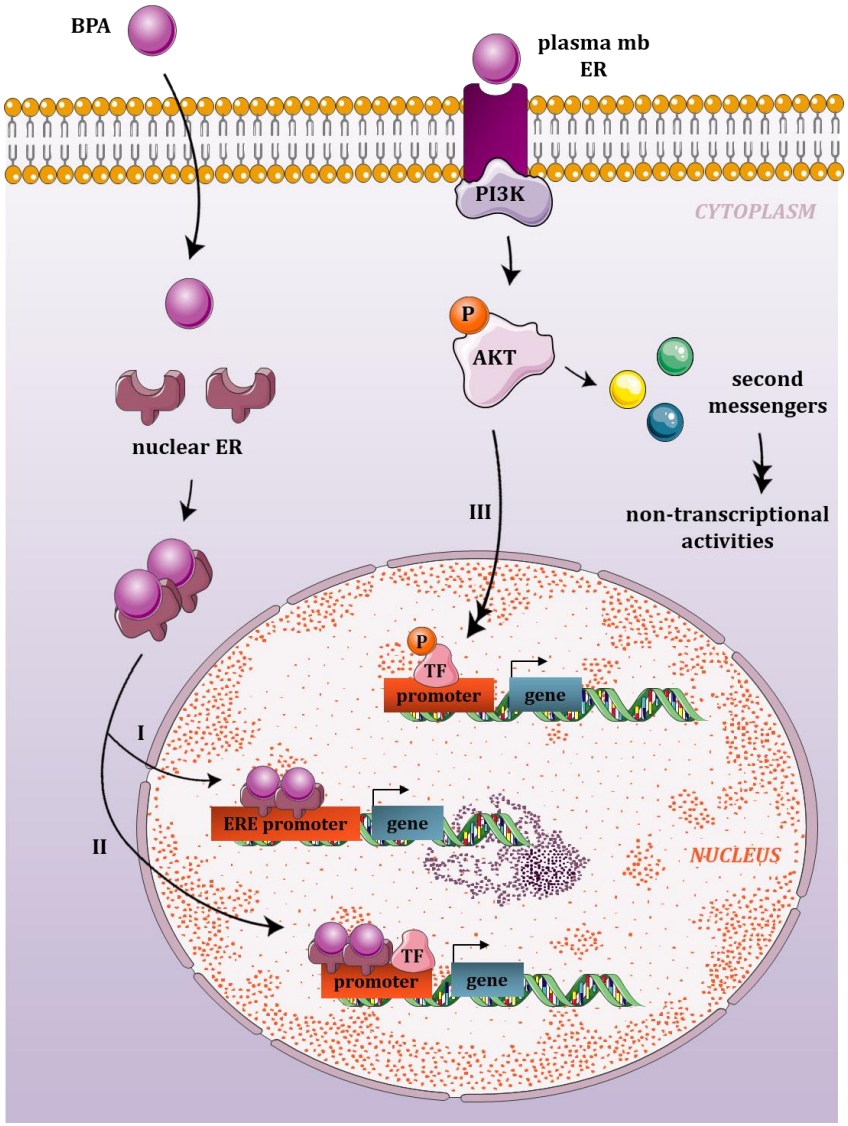


# INTRODUCTION

## BPA: ENDOCRINE DISRUPTOR



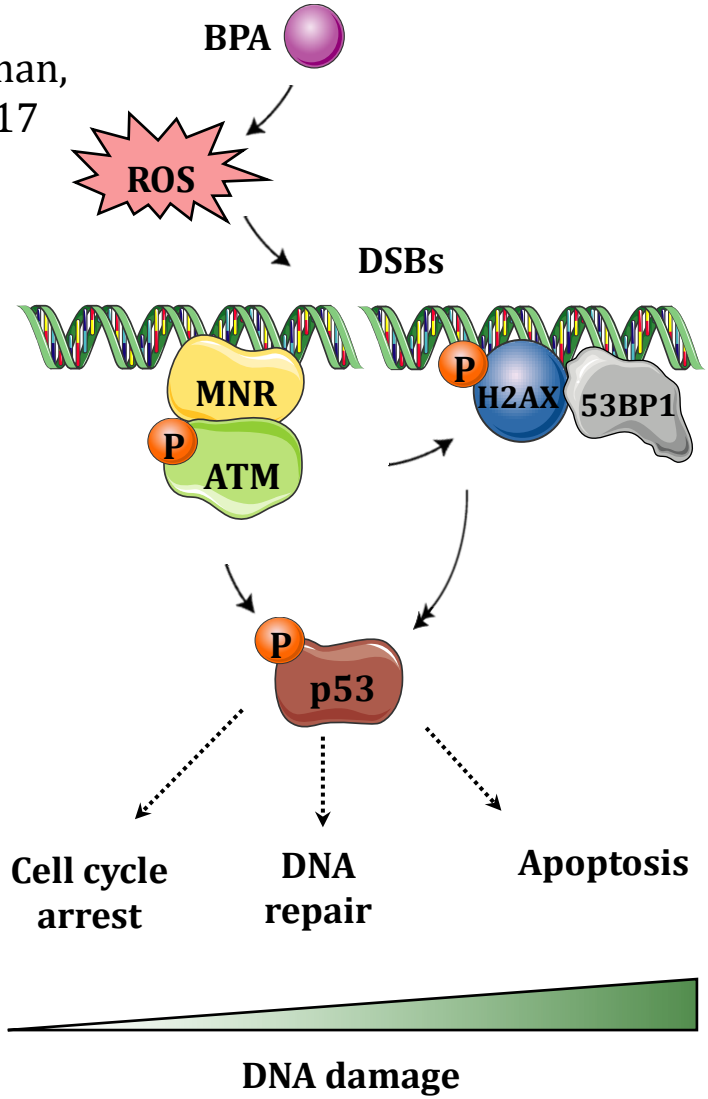

Combarnous and Nguyen, 2019



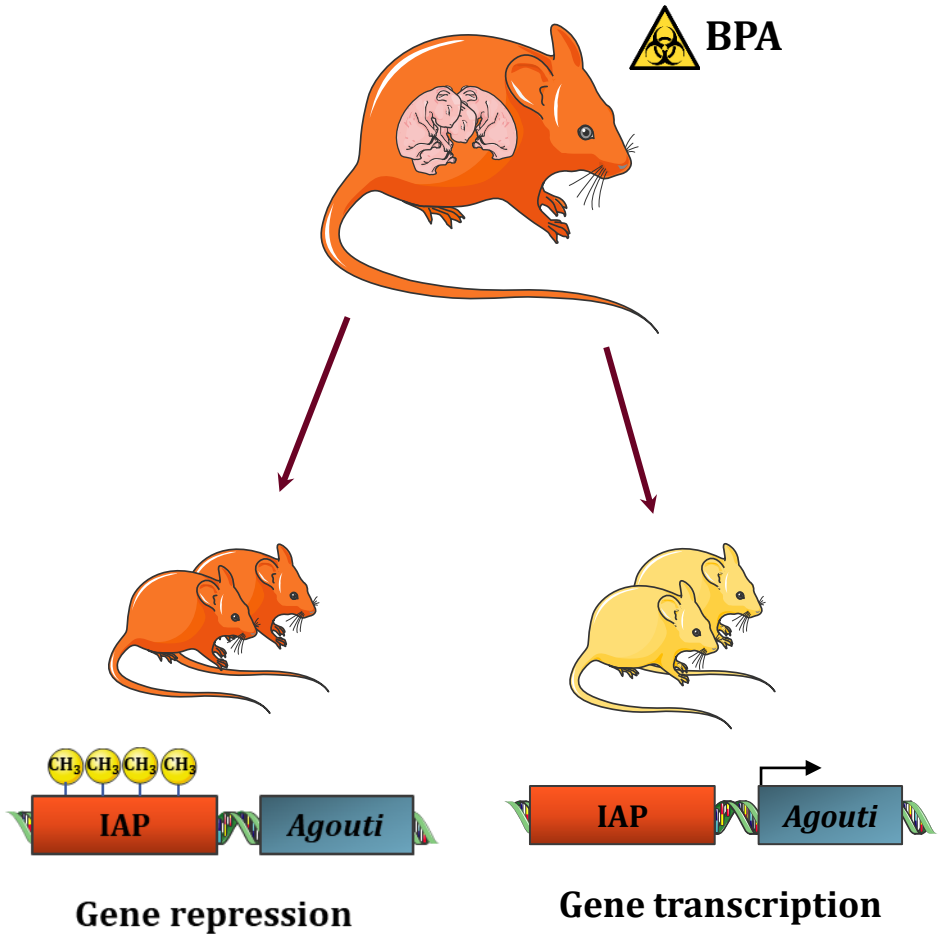
# INTRODUCTION

## BPA GENOTOXICITY

Gassman, 2017

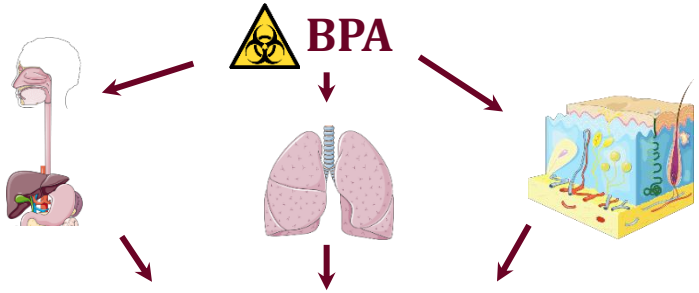



## BPA EPIGENOTOXICITY




Dolinoy, 2007

# INTRODUCTION




Hypothalamus

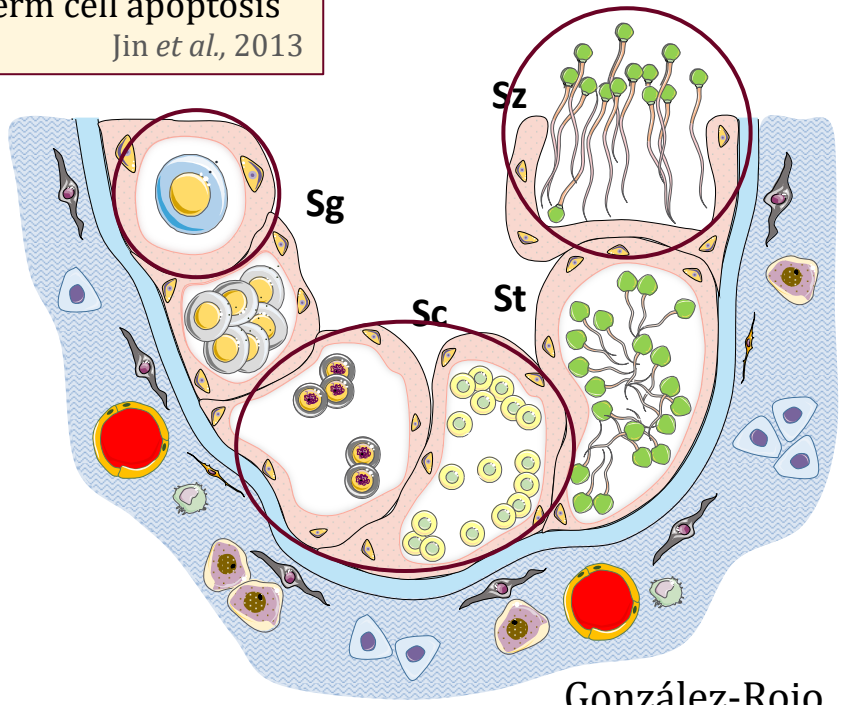
Pituitary gland

Testicles

Testosterone

Reduced sperm count
Manfo *et al.*, 2013

Germ cell apoptosis
Jin *et al.*, 2013




González-Rojo, 2018

Altered spermatogenesis

Meiotic arrest
Reduced spermatocyte count
González-Rojo *et al.*, 2019
Liu *et al.*, 2013

# HYPOTHESIS

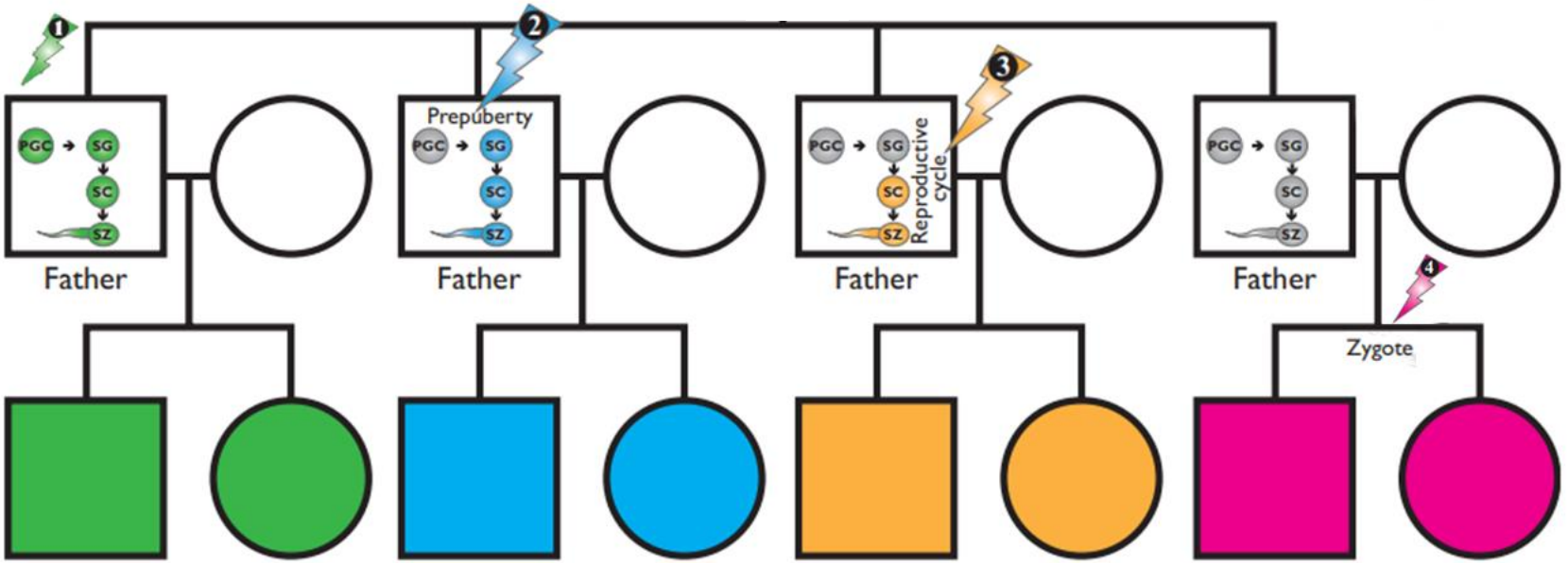
Given that the **information** contained in these cells is crucial **for the embryo development,** paternal exposure to bisphenol A could jeopardize health of future generations, even to those which are not directly exposed to the toxicant

# MAIN OBJETIVE

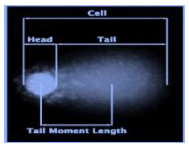
To determine how bisphenol A alters the paternal contribution to embryo development

# EXPERIMENTAL DESIGN

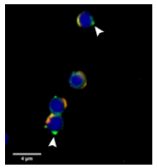
## Bisphenol A male exposure




Soubry *et al.*, 2014

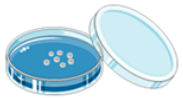



**Comet assay (Fernández-Díez *et al.*, 2018)**

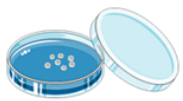


**Cell immunostaining (González-Rojo *et al.*, 2018)**

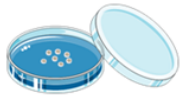
**F1**



**F1 control**



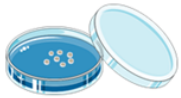
**F1 100 µg/L BPA**



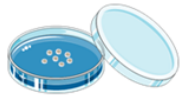
**F1 2000 µg/L BPA**

**F2**

**F1 control**



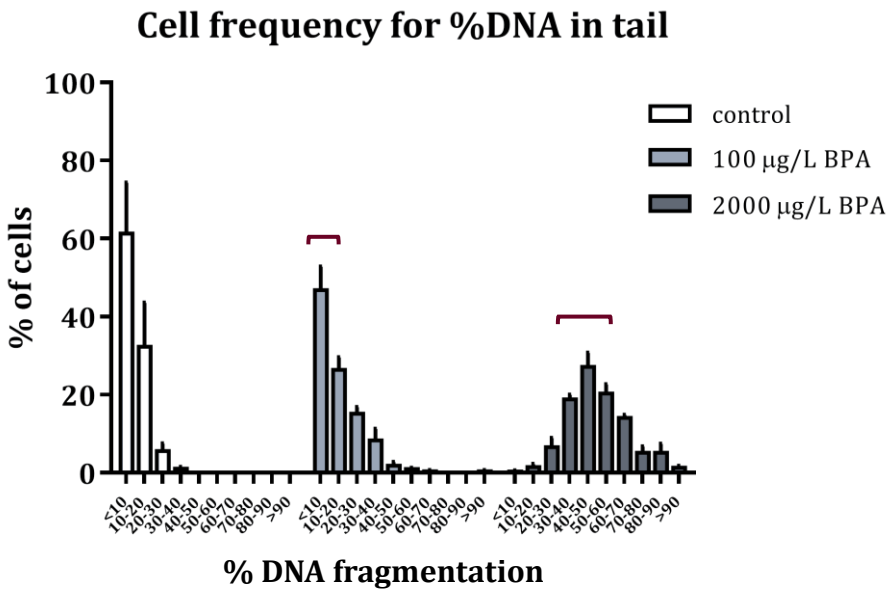
**F1 100 µg/L BPA**

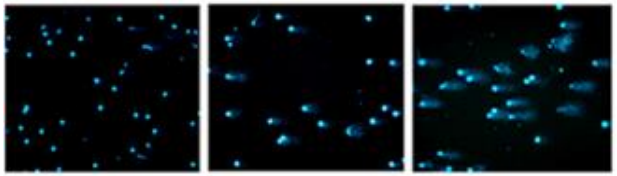


**F1 2000 µg/L BPA**

# GENOTOXIC EFFECTS

## Sperm DNA fragmentation

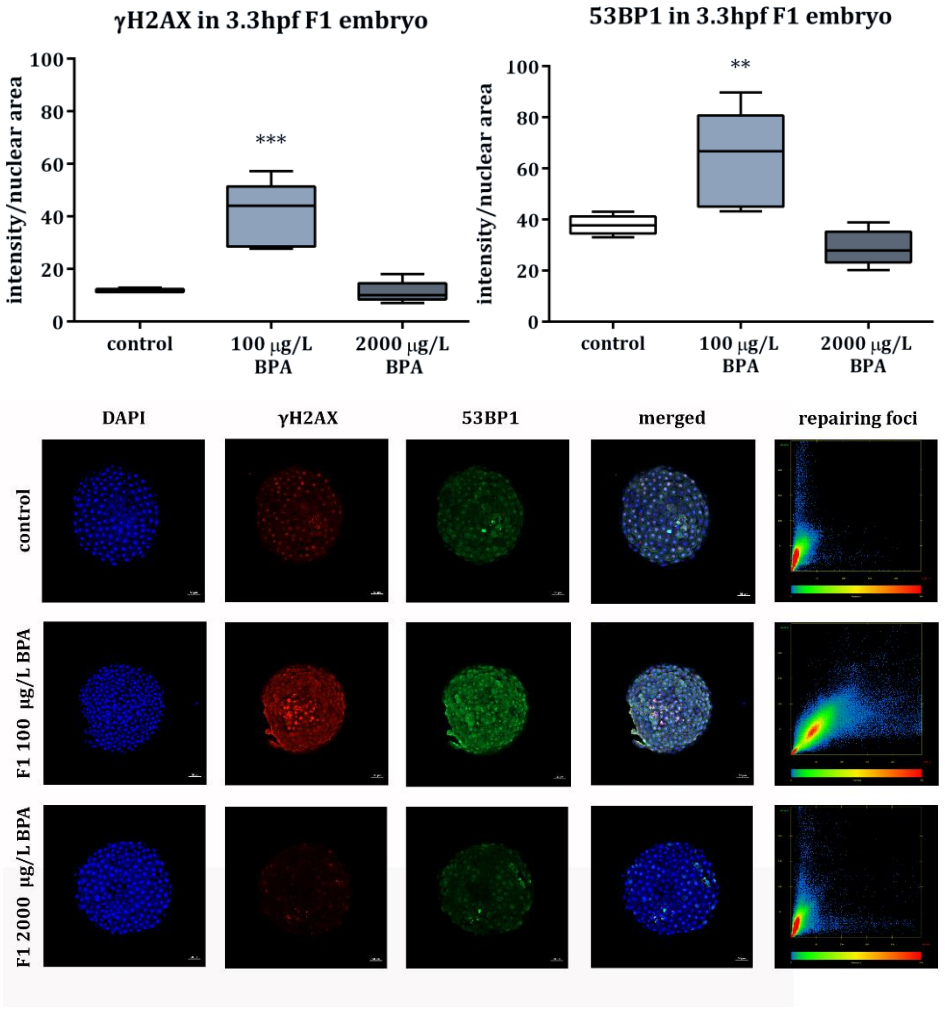


Mature spermatozoa lack repairing mechanisms and their extremely compacted chromatin blocks the access of the DNA repairing machinery.

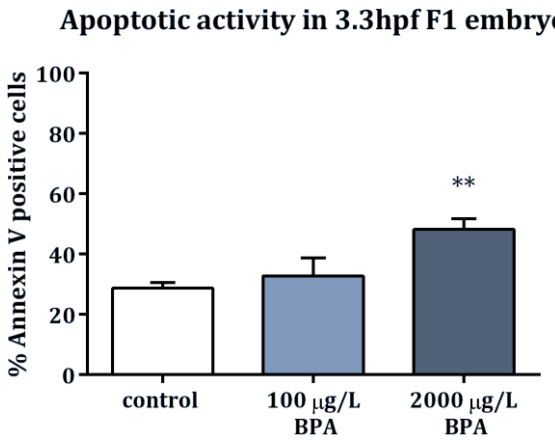
Herráez *et al.*, 2017

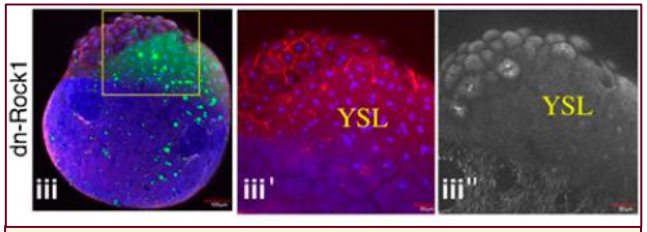
## Activation of DNA repair



# GENOTOXIC EFFECTS

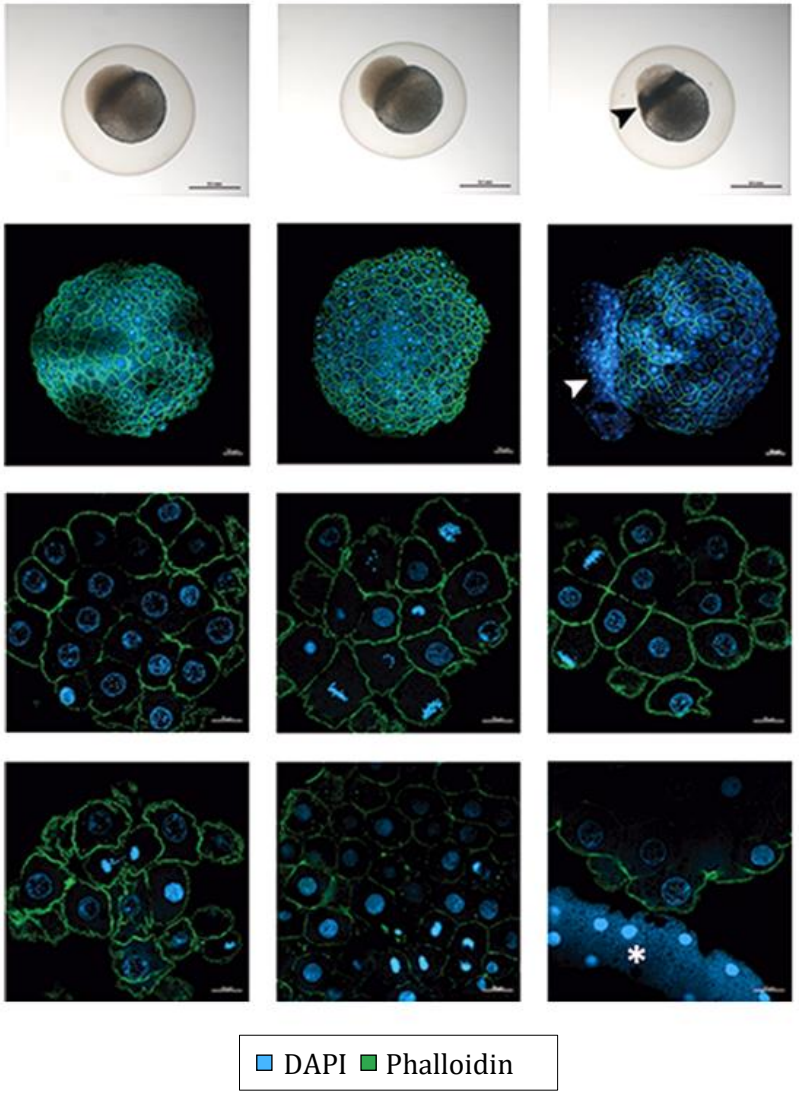
## Apoptosis and formation of YSL




Rock1 binds to Rho GTPases, allowing the entrance of DDR factors to the nucleus by modifying actin cytoskeleton.

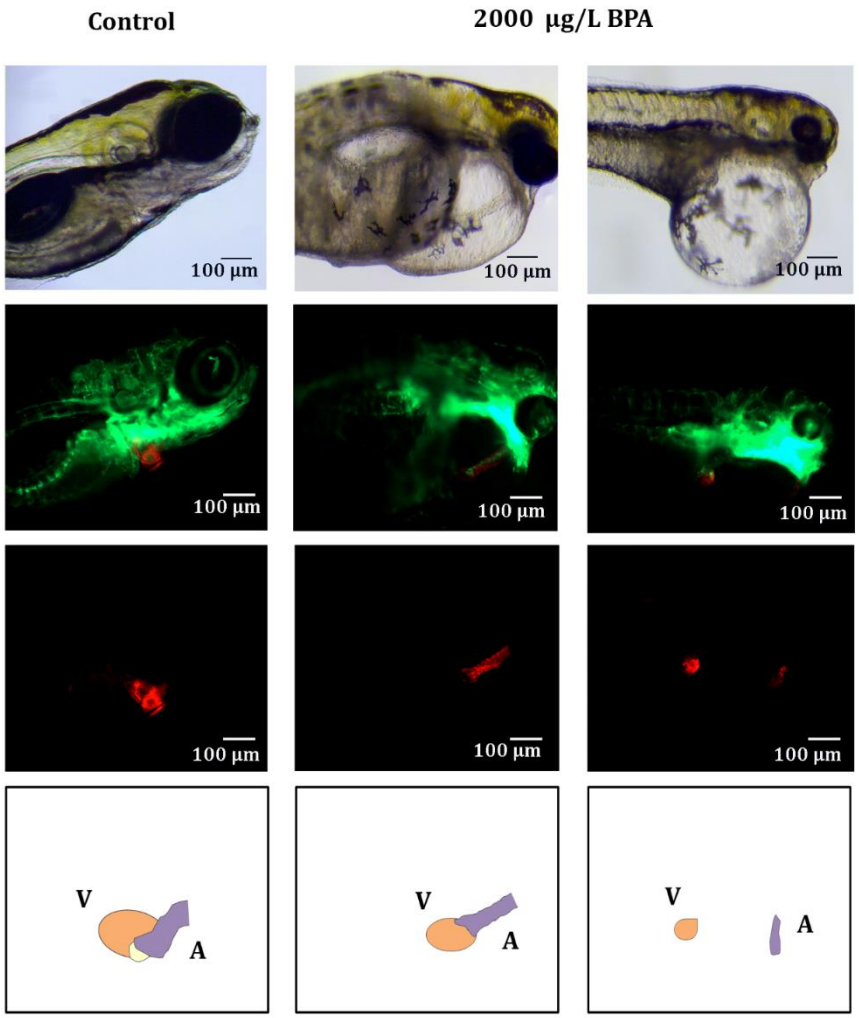
Fritz and Henninger, 2015



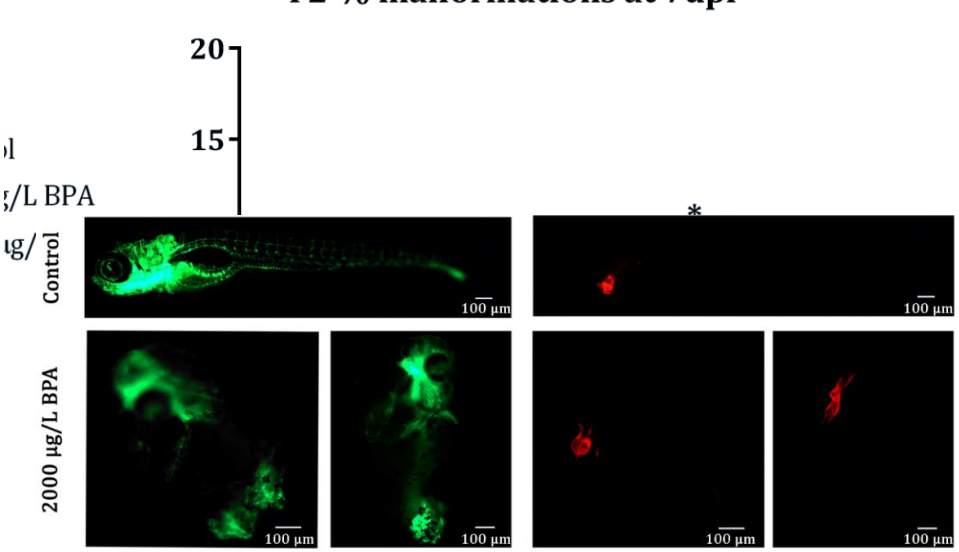
# F1 EMBRYO DEVELOPMENT

## Deve Heart malformations eny

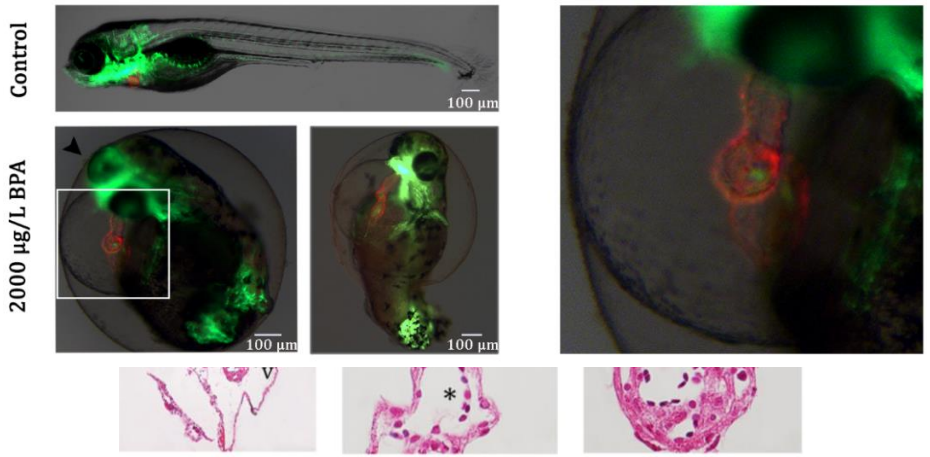
*Tg(fli1:EGFP;mlc2a: mCherry)*



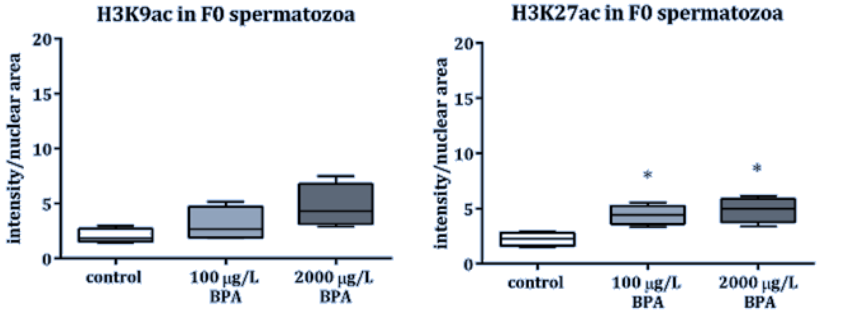

## Development of the F2 progeny

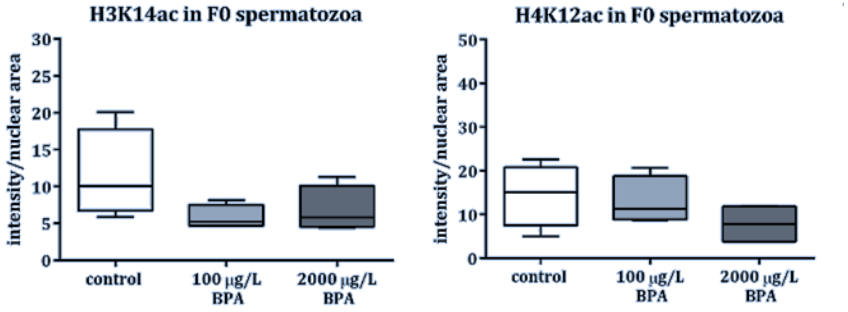


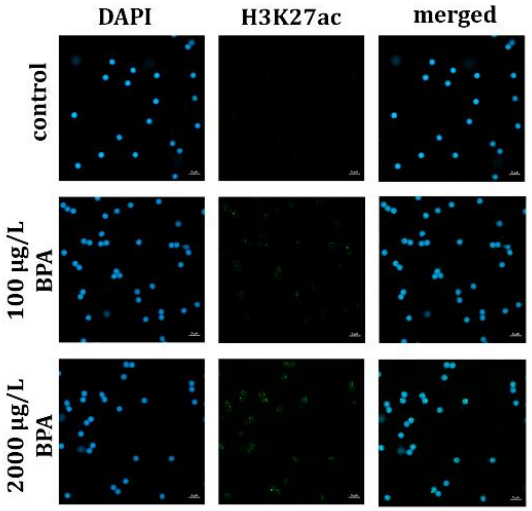

**F1 from males exposed to 2000 µg/L BPA**

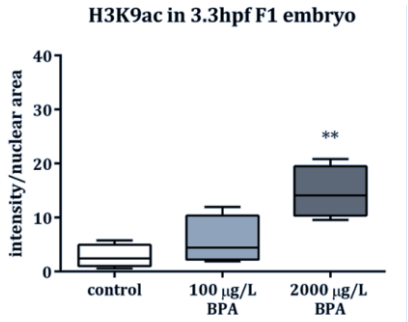


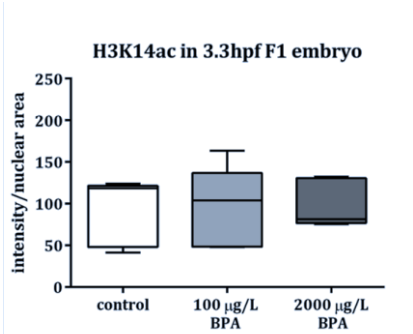
# EPIGENOTOXIC EFFECTS



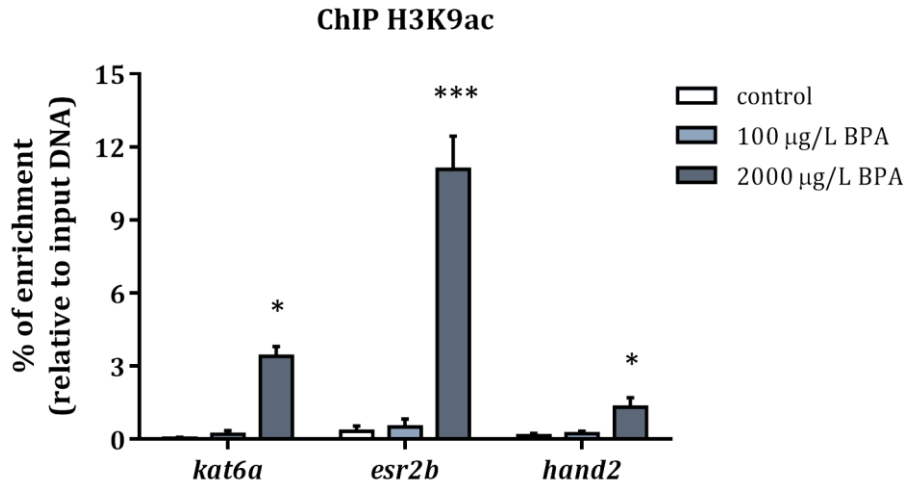



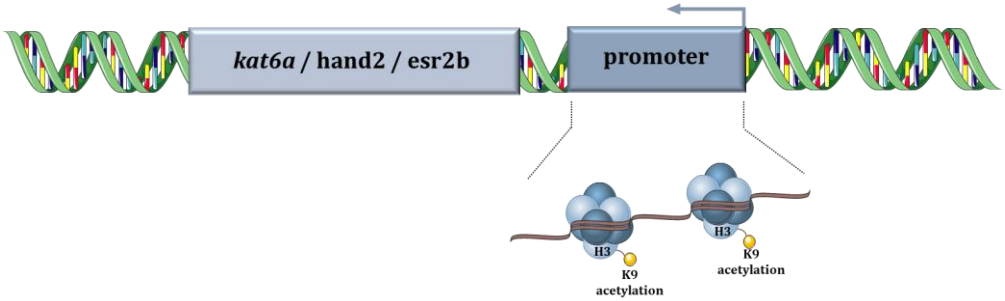



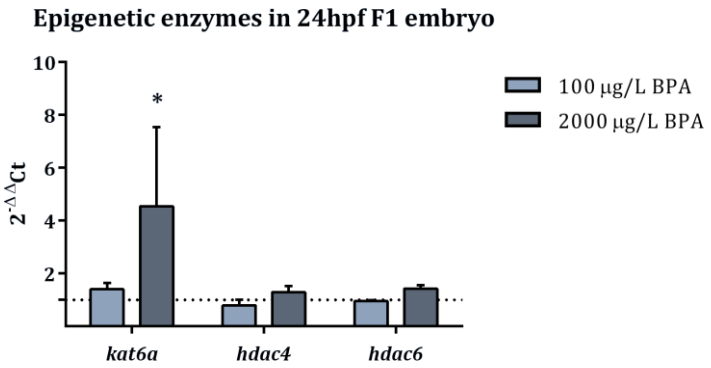



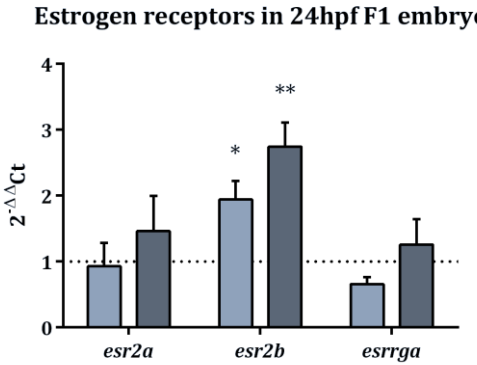
# GENE EXPRESSION

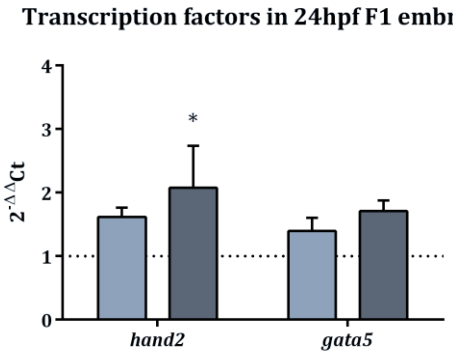
## Gene expression



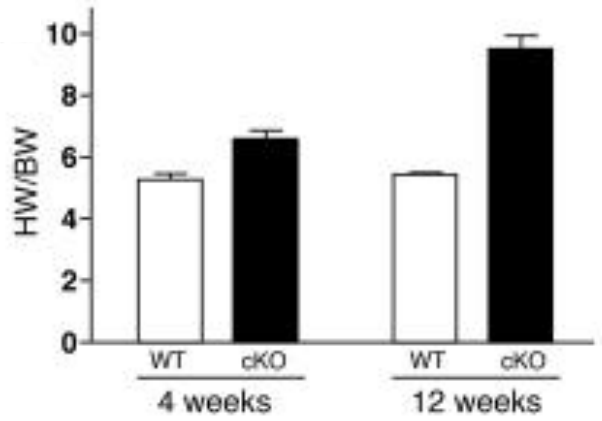



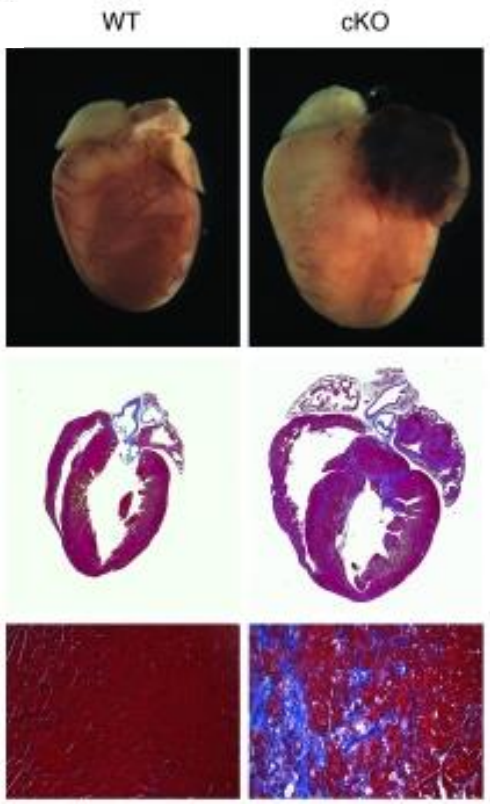
# HYPOTHESIS

The epigenetic toxicity of BPA might well lie behind the cardiac malformations we have observed since epigenetic mechanisms regulate gene expression during cardiogenesis




Deletion of *Hdac3* results in cardiac hypertrophy, left atrial thrombus, and cardiac fibrosis.

Montogomery *et al.*, 2008

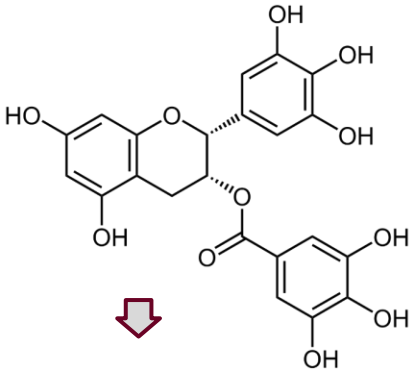


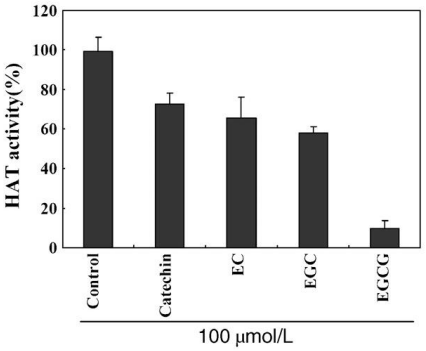
# EXPERIMENTAL DESIGN

## EGCG F1 treatment

*Camellia sinensis*

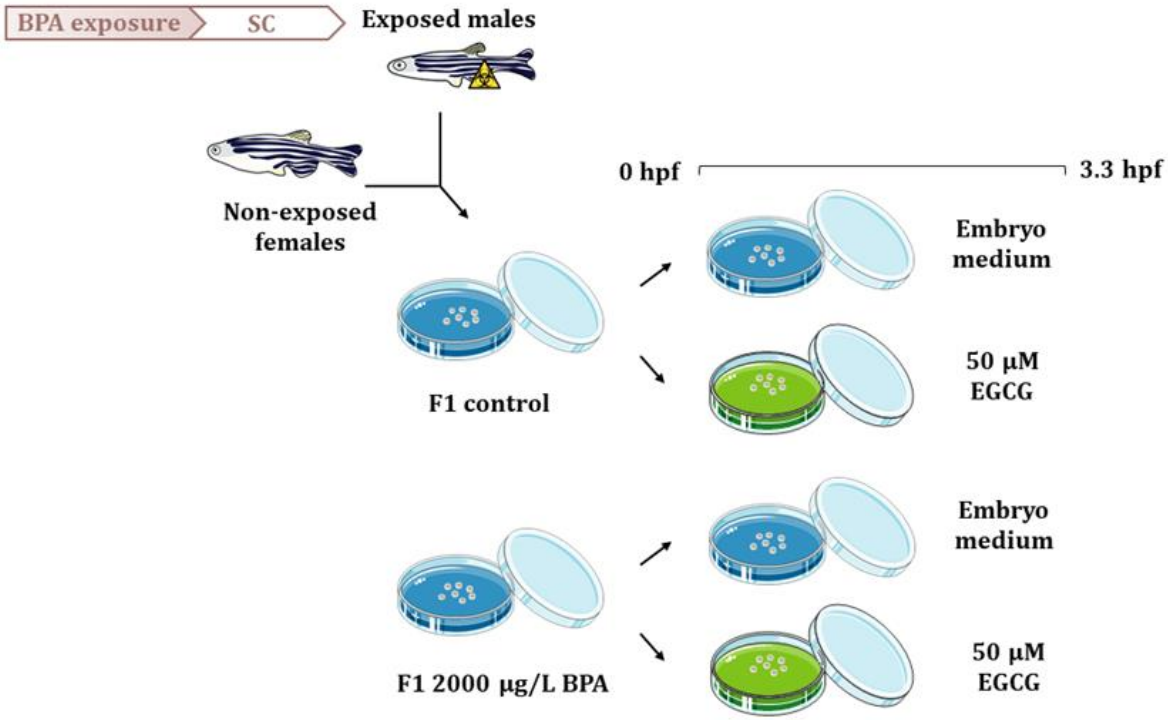
**Epigallocathechin-3-gallate**



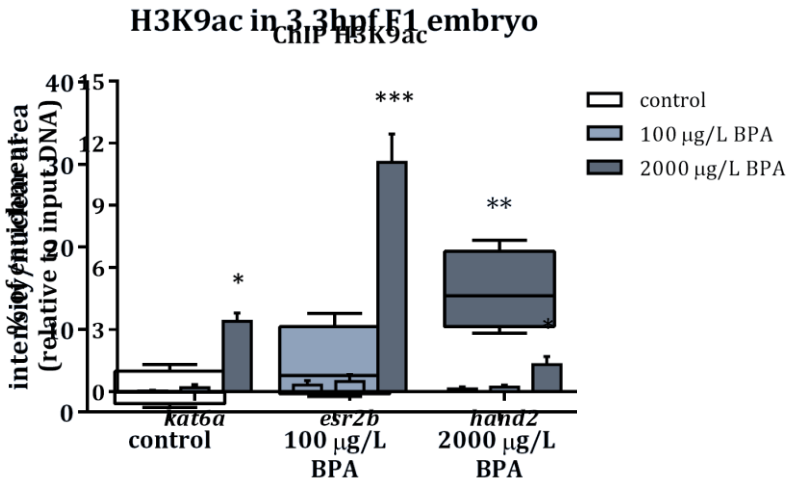

EGCG is a strong **HAT inhibitor**

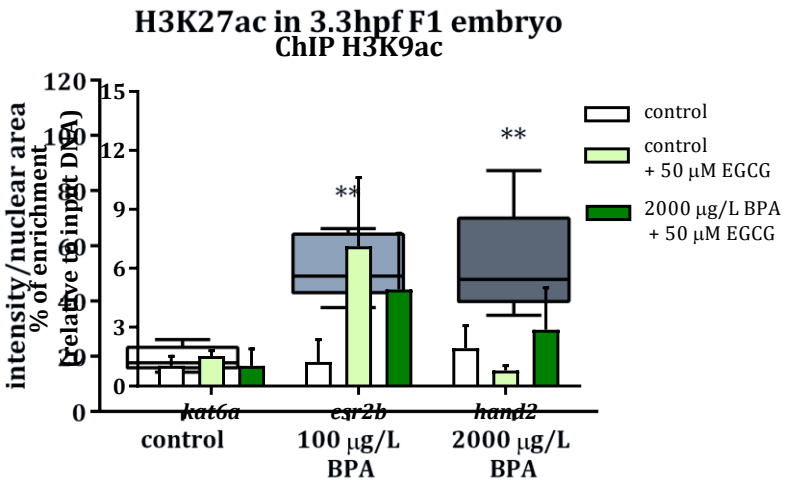
Choi *et al.*, 2009

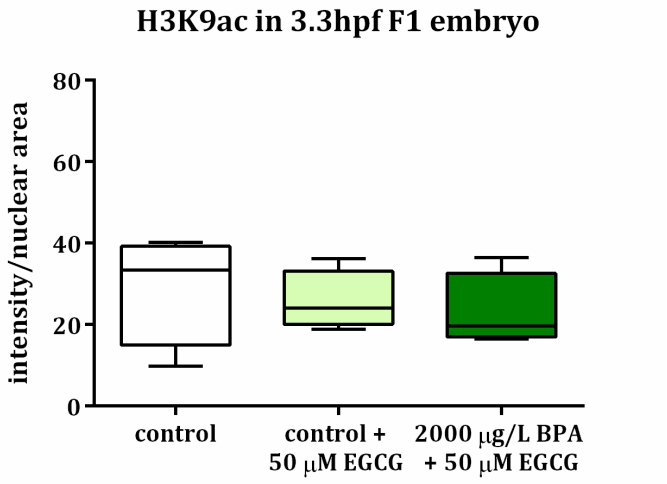


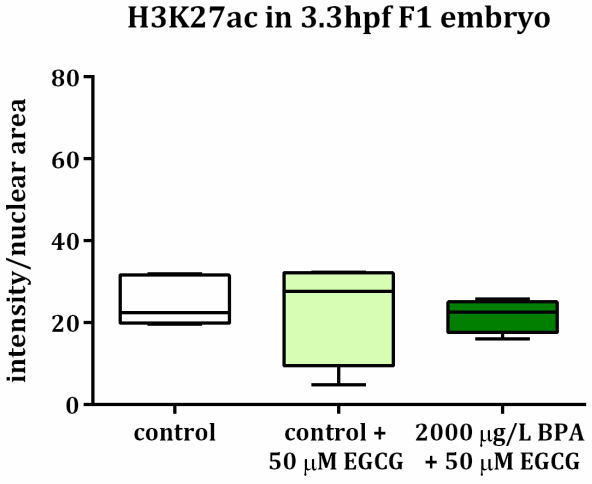
# EPIGENETIC MODIFICATIONS

## Histone acetylation



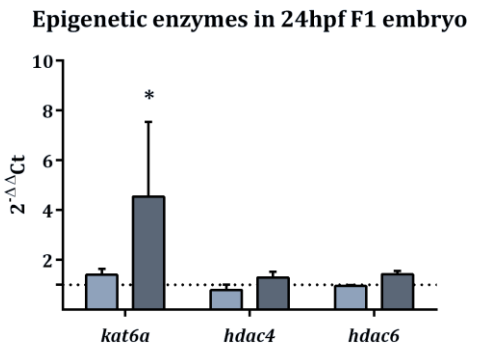



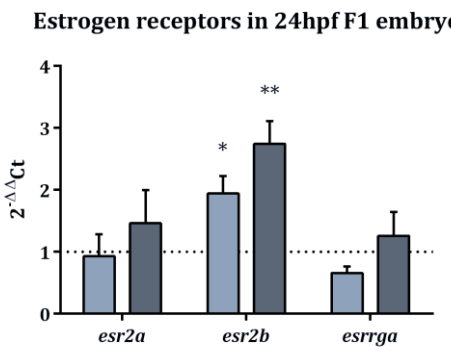
# GENE EXPRESSION

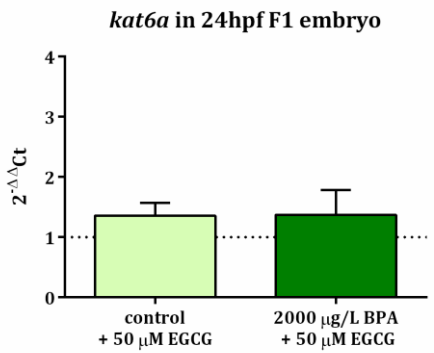
## Gene expression

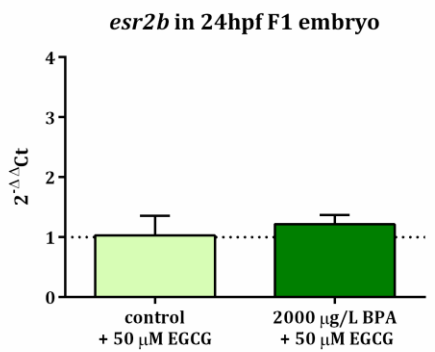
### Paternal BPA exposure

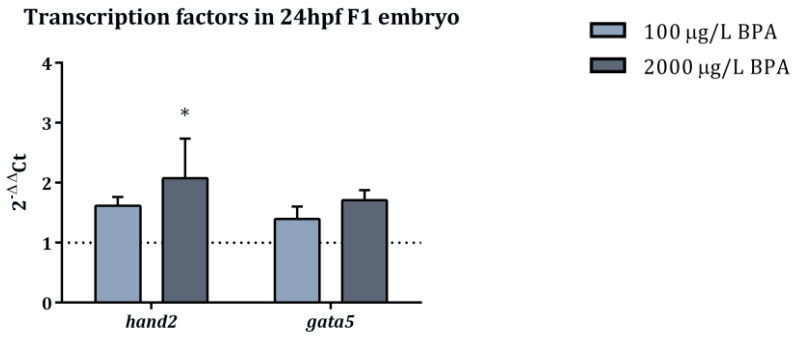
### F1 EGCG treatment

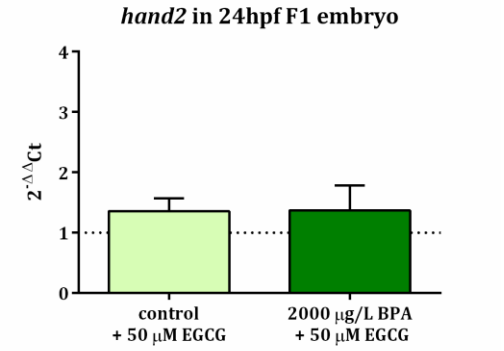


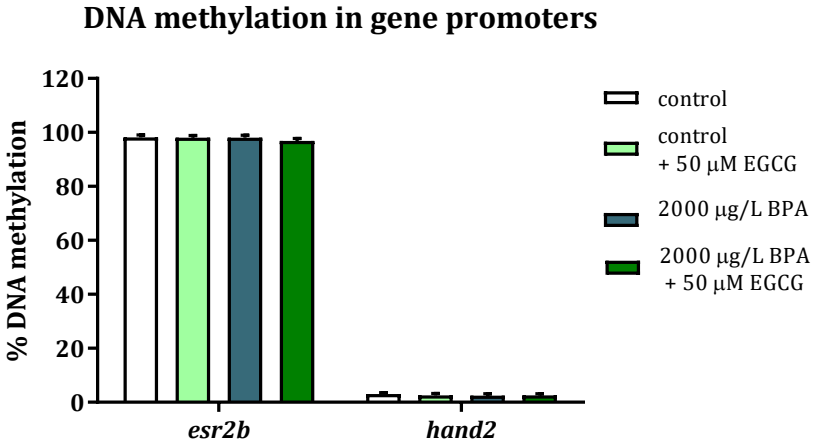



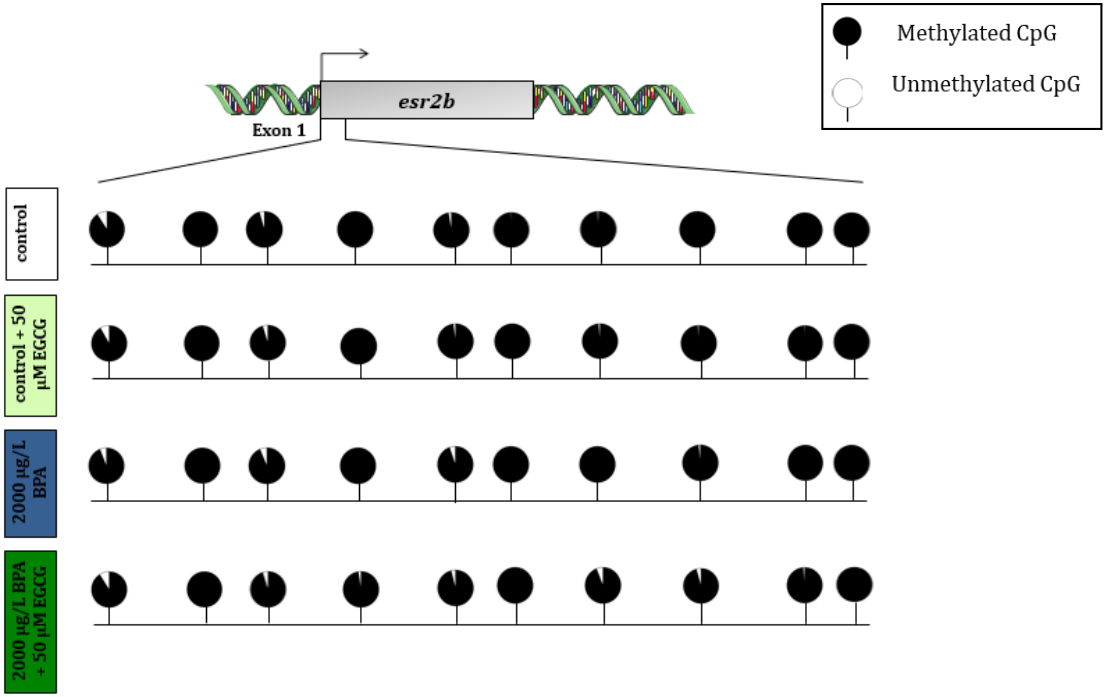

Treatment with 120 μM EGCG during cardiac-specific differentiation suffered a decrease in H3 acetylation leading to reduced levels of heart transcription factors.

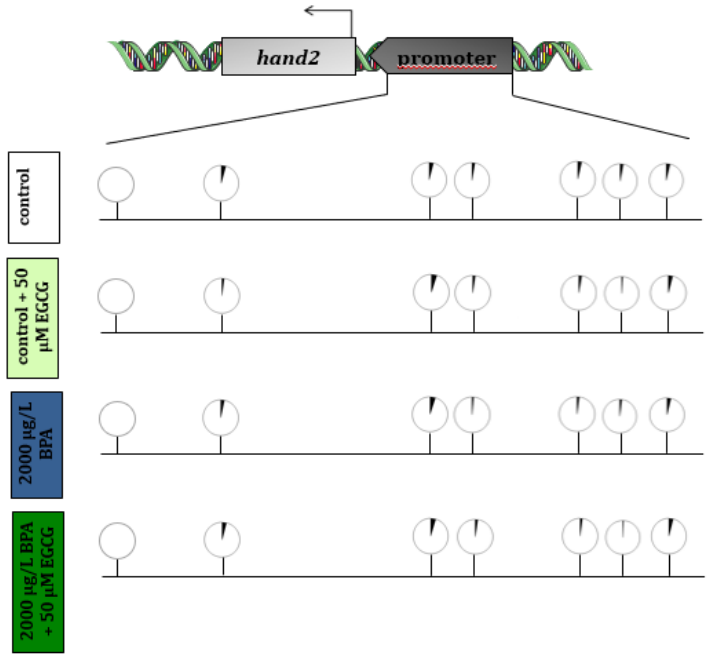
Yin *et al.*, 2014

# EPIGENETIC MODIFICATIONS

## DNA methylation

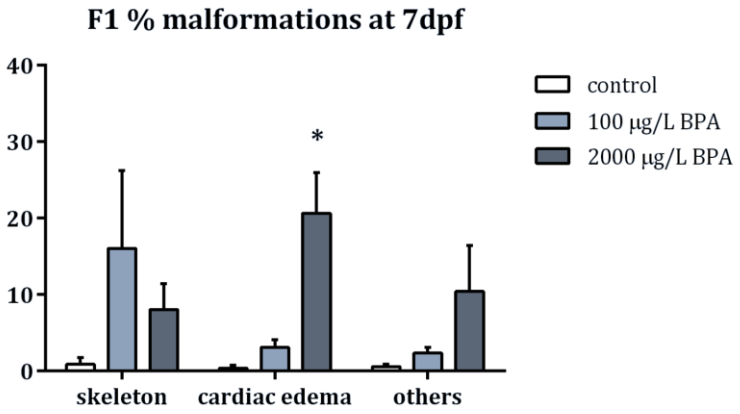


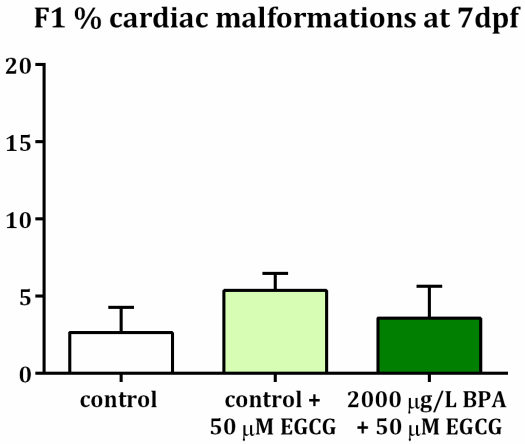



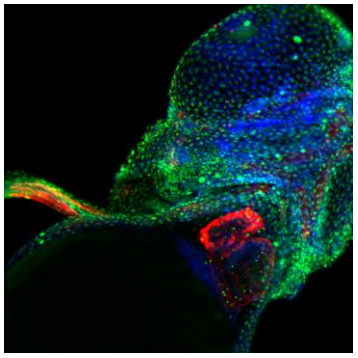
# F1 EMBRYO DEVELOPMENT

## Heart malformations

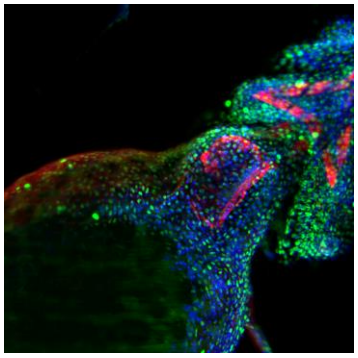


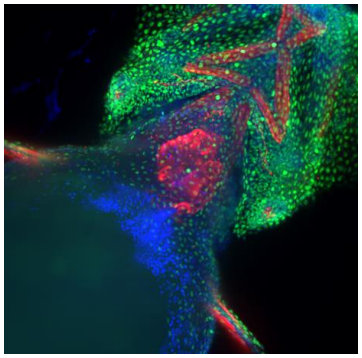

**control**

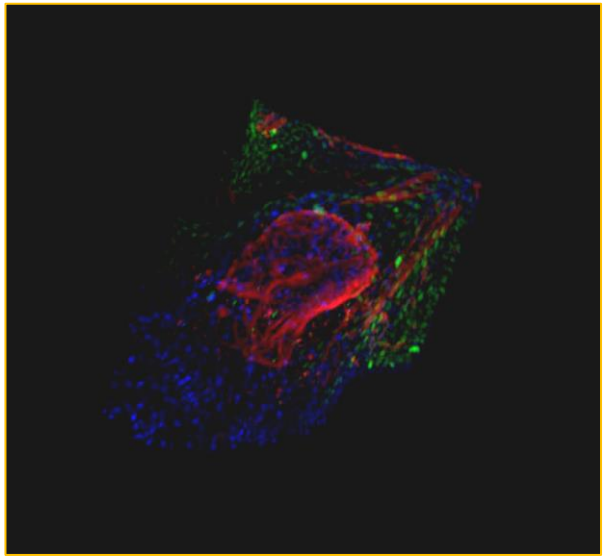


**F1 BPA**

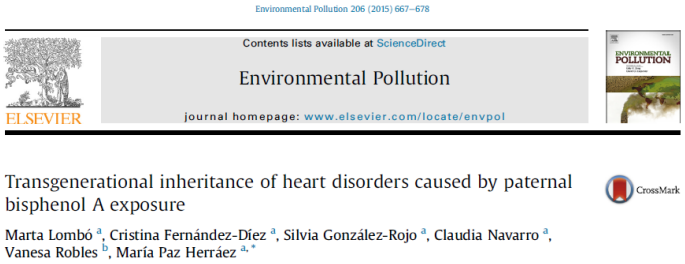


**F1 BPA + EGCG**





# CONCLUSIONS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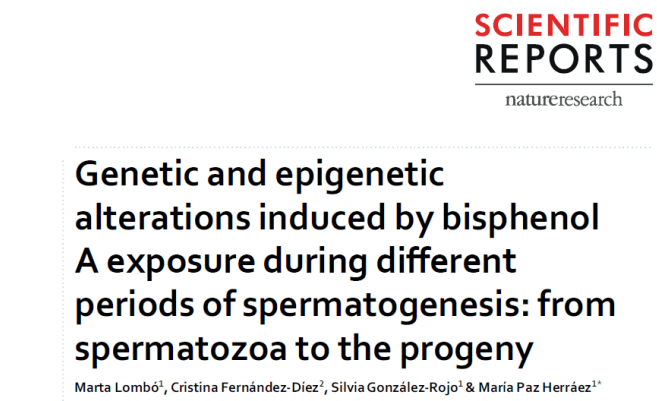
journal homepage: www.elsevier.com/locate/envpol

Transgenerational inheritance of heart disorders caused by paternal bisphenol A exposure

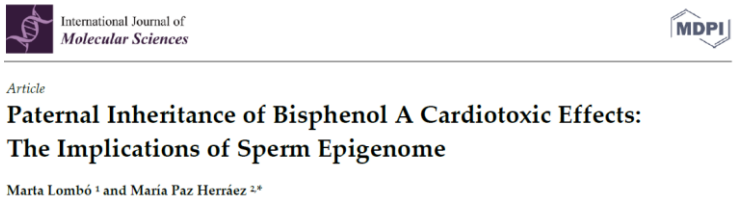
Marta Lombó [a], Cristina Fernández-Díez [a], Silvia González-Rojo [a], Claudia Navarro [a], Vanesa Robles [b], María Paz Herráez [a, *]


SCIENTIFIC REPORTS

nature research

Genetic and epigenetic alterations induced by bisphenol A exposure during different periods of spermatogenesis: from spermatozoa to the progeny

Marta Lombó[1], Cristina Fernández-Díez[2], Silvia González-Rojo[1] & María Paz Herráez[1,*]


International Journal of Molecular Sciences

MDPI

Article

Paternal Inheritance of Bisphenol A Cardiotoxic Effects: The Implications of Sperm Epigenome

Marta Lombó [1] and María Paz Herráez [2,*]

**1.** Paternal exposure to BPA impairs zebrafish embryo development up to F2, a generation never exposed to the toxicant.

**2.** Male exposure to BPA promotes sperm DNA fragmentation that, when overcoming the embryonic repairing capacity, triggers embryonic apoptosis.

**3.** Histone acetylation in F1 embryos coming from BPA-exposed males, specifically affects the expression of genes involved in heart development

**4.** EGCG embryonic treatment allows the mitigation of cardiotoxic effects caused by paternal BPA exposure

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