

•••• ATAC-seq and RNA-seq analysis reveal new elements of planarian posterior organizer

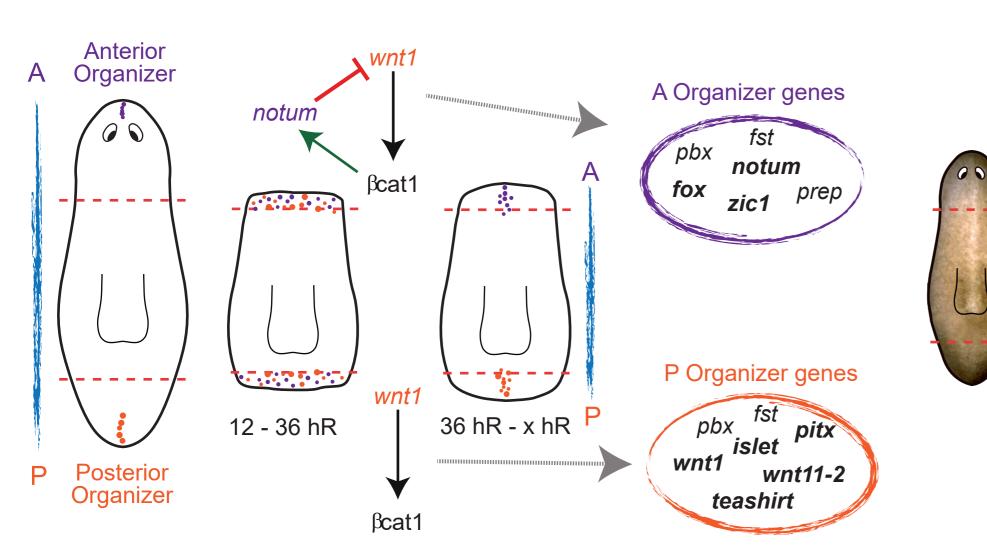


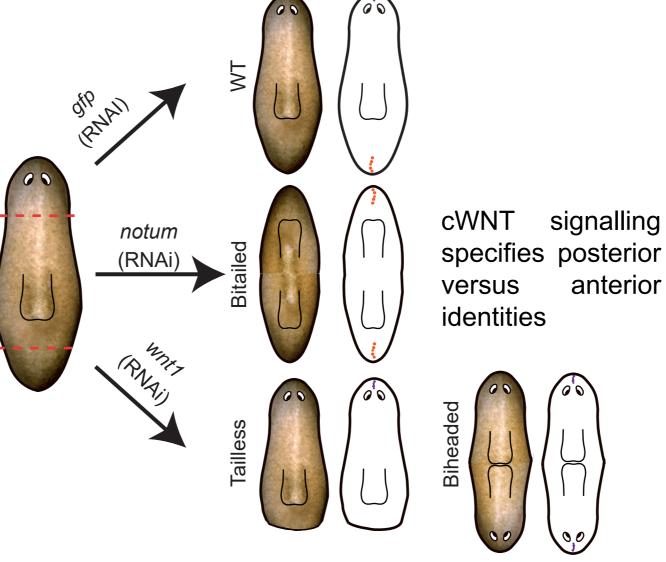
E. Pascual-Carreras¹, M. Marín¹, S. Castillo-Lara¹, P. Coronel-Córdoba¹, M.S. Magri², J.F. Abril¹, J.L. Gomez-Skarmeta², E. Saló¹ and T. Adell¹

1. Department of Genetics, Microbiology and Statistics, Universitat de Barcelona (UB) & Institute of Biomedicine of Universitat de Barcelona (IBUB), Barcelona, Spain. 2. Centro Andaluz de Biología del Desarollo (CABD), Universidad Pablo de Olavide, Sevilla.

Organizers or signaling centers are a group of cells with the ability to specify the fate of adjacent cells, allowing a patterned growth. Although organizers are mainly studied during embryogenesis, their function is also required in adults, for instance during regeneration. To better understand the formation and function of adult organizers, we study planarians, flatworms that are able to regenerate any missing body part. In planarians the anterior and posterior tips of the body behave as organizers, being defined by the expression of notum (a secreted Wnt inhibitor) and wnt1, respectively. The inhibition of any of those elements leads to a shift in polarity. Interestingly, during the first hours of regeneration both notum and wnt1 are expressed in both poles, and it's around 36 hours that their expression becomes restricted to their respective tip. To decipher the molecular interactions that restrict the expression of wnt1 to the posterior tip and confer the organizing activity we used genome wide approaches. ATAC-seq and RNA-seq analysis of regenerating wild-type and wnt1 (RNAi) planarians allowed the identification of specific Cis-Regulatory Elements (CREs) of posterior regeneration. We found that already at 12 hours of regeneration the accessible CREs in posterior and anterior blastemas have essentially changed, indicating that specific posterior chromatin changes induced by amputation occur much earlier than the formation of the organizers. Furthermore, we have identified specific transcription factors of the Otx and Fox families, which are enriched in posterior CREs and are essential for the specification of the posterior wnt1+ cells.

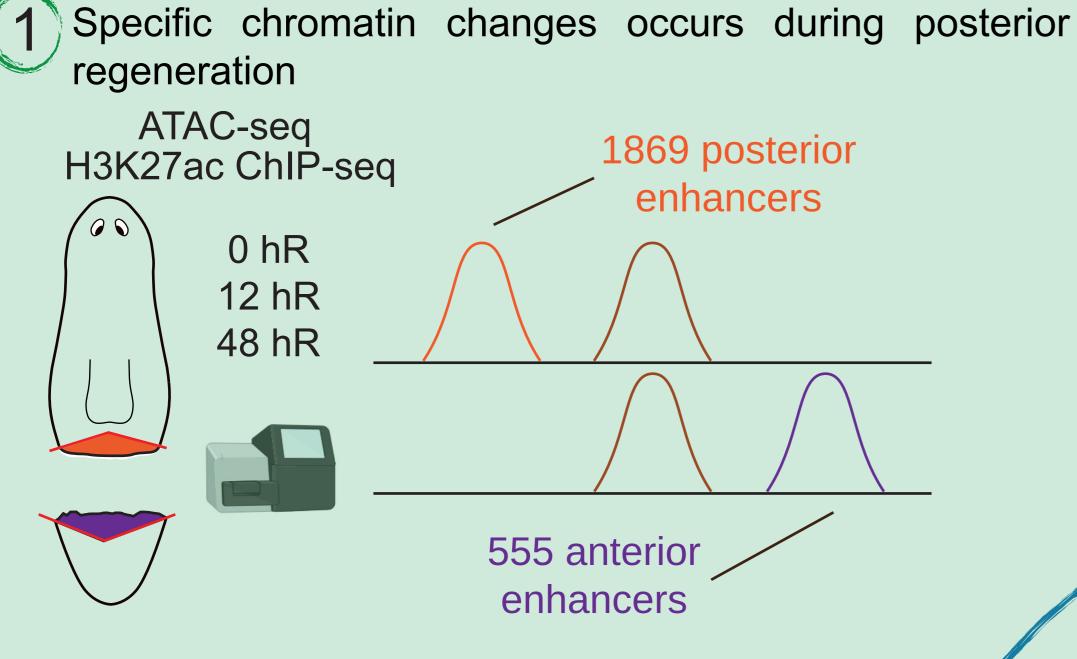
Anteriopoosterior axis establishment and WNT signalling pathways in planarians





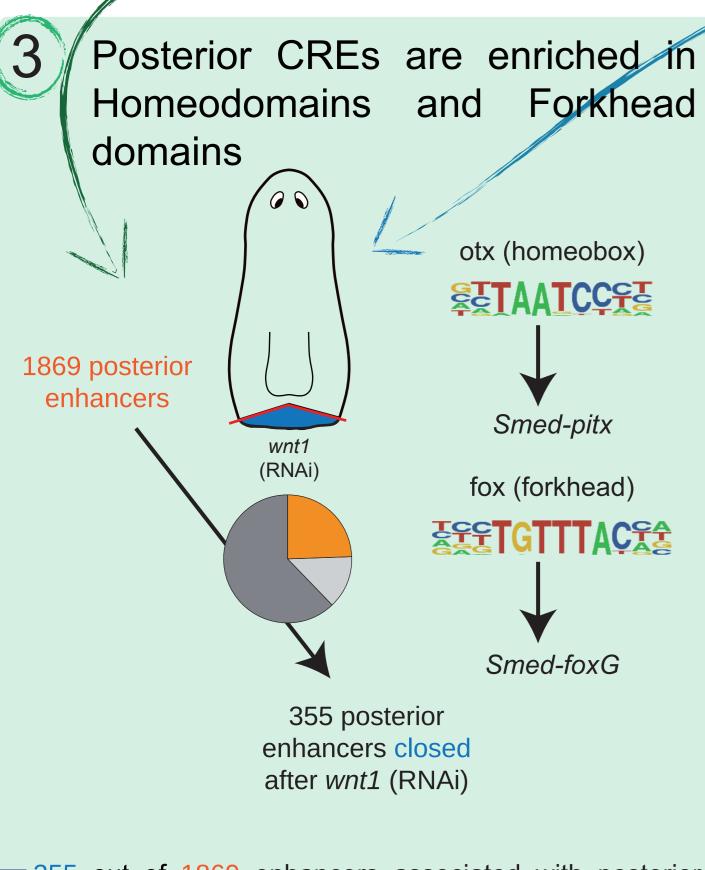
cWNT pathway the planarian affecting epigenome the formation posterior organizer?

We have performed ATAC-seq, ChIP-seq and RNA-seq during posterior regenerating.



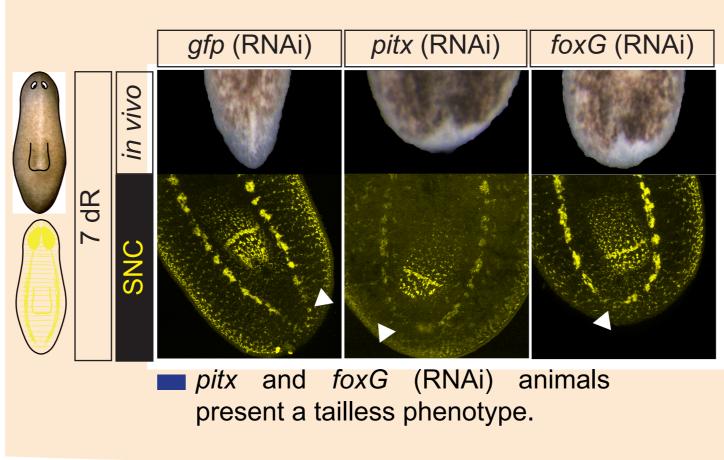
ATAC-seq and ChIP-seq comparison between anterior vs posterior regenerating planarians, reveals anterior and posterior specific CREs

notum and wnt1 inhibition change chromatin accessiblity in 12 hours regenerating blastemas wnt1 notum (RNAi) (RNAi) Non Accessible **Core Posterior Enhancers** Less Accessible More Accessible **Core Anterior** Enhancers Accessible (RNAi) anterior blastemas ■ In Wnt1(RNAi) **notum** posterior blastemas, present posterior CREs accessible, and **CREs** anterior being are more anterior CREs partially non accessible. accessible, and posterior CREs are being less accessible.



355 out of 1869 enhancers associated with posterior regeneration were none or less accessible in wnt1 (RNAi) animals. Homeodomains and forkheads motifs were highly represented, suggesting their putative role in normal posterior regeneration.

We identified *pitx* (Homeobox) and *foxG* (forkhead) as new elements required for specification of Posterior organizer



The ATAC-seq/ChIP-seq strategy allowed the identification of pitx and foxG as transcription factors essential for posterior identity specification during planarian regeneration



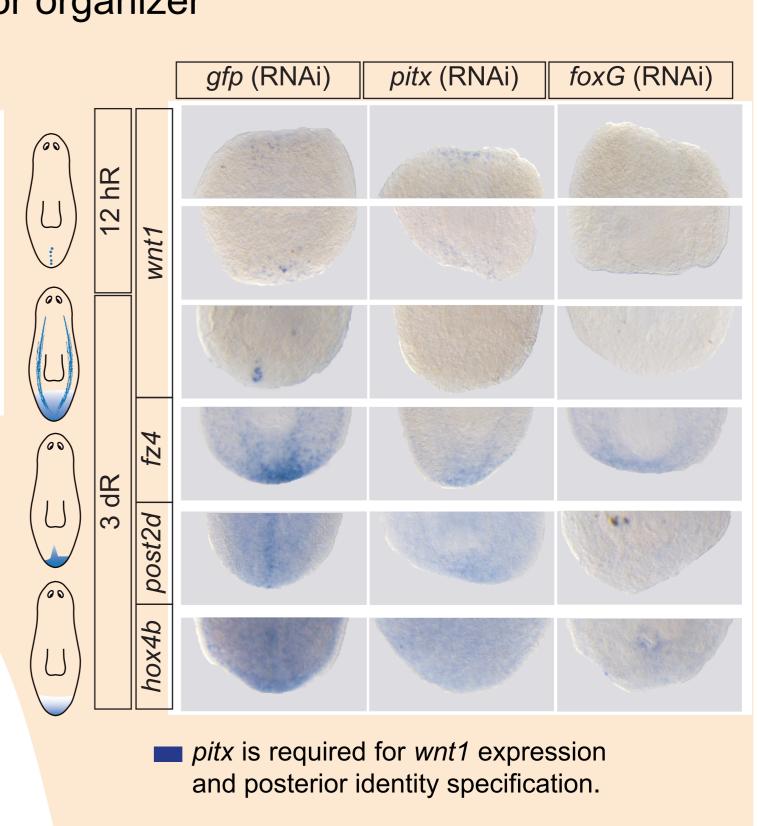
Agència

Universitaris

i de Recerca



This work was funded by grants BFU2014-560558 de Gestió d'Ajuts (MEI) and BFU2017-; and 2014SGR and 2016SGR (AGAUR) to E.S. // eudald.pascual@ub.edu // https://planarian.bio.ub.edu



foxG is required for early and late wnt1 expression, and posterior identity specification.