BEHAVIOR

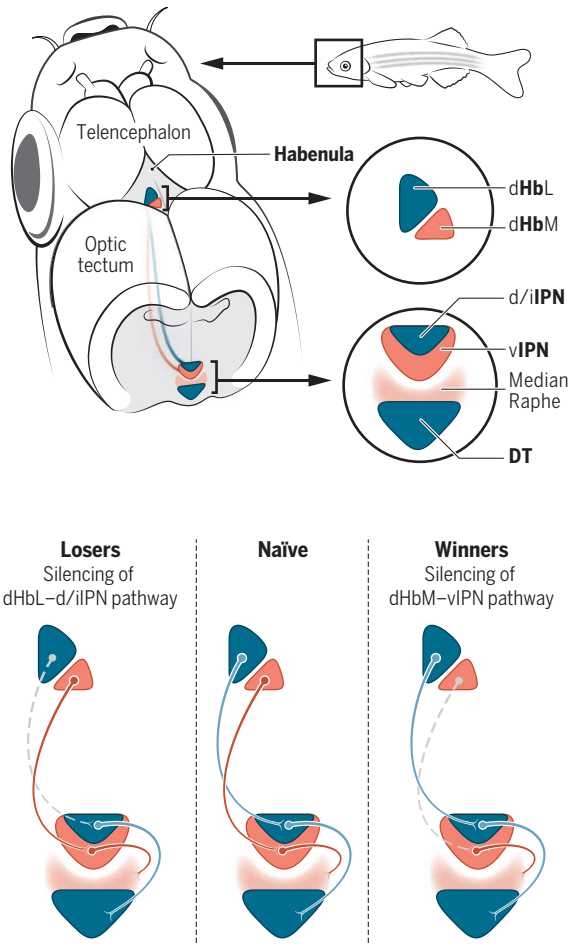
A brain conditioned for social defeat

A brain circuit in vertebrates determines who wins or loses a fight

By **Laura Desban** *and* **Claire Wyart**

Aggression is common in the animal kingdom, even though agonistic behaviors can lead to chronic stress or pain. So how does aggression remain conserved evolutionarily? In 1859, Darwin argued in his book *On the Origin of Species* that the conservation of any behavioral trait is ultimately explained by its necessity for survival and reproduction. To survive with limited resources, individuals express aggressive behaviors against competitors to pass on their genes. For social animals, dominance hierarchies establish rapidly (*1*), avoiding the cost of recurrent fighting within the group. Hierarchy formation and maintenance rely on the effect of prior experience (*2*). However, the underlying mechanisms and neural circuitry remain elusive. On page 87 of issue, Chou *et al.* (*3*) identify a key role for the dorsal habenula (dHb) region of the brain in zebrafish to determine who wins and who loses in a fight. This region is highly conserved across vertebrates, raising the possibility of manipulating neuronal circuits that govern innate social behaviors.

The social status of an individual determines the quality and the quantity of its opportunities. Individuals usually keep their social rank over time by means of positive or negative reinforcement induced by prior experience, referred to as the winner effect or loser effect, respectively. During dyadic fighting, zebrafish males show agonistic behavior with a stereotypic sequence (*4*): first display behaviors by erecting its fins along with body flaring, then circle and bite until dominance is established. The fight ends with the surrender of the losing fish, who exhibits fleeing behavior. By imaging calcium (as a readout for neuron activity) in acute brain slices obtained from fish immediately after fighting, Chou *et al.* observed differences between winners and losers at the level of the dorsal habenula–interpeduncular nucleus (dHb–IPN) pathway (see the figure). In losers, neuronal activity originating from dHb onto the dorsal and intermediate IPN (d/iIPN) was reduced. The authors confirmed by in vivo electrophysiology (that is, by inserting electrodes to record local field potentials in the IPN) that dominance status is associated with activity levels in the dHb-IPN circuitry.



Habenular circuitry implicated in the outcome of fighting. (**Top**) Schematic showing a dorsal view of habenular connections onto the IPN in the zebrafish brain. The dHbL–d/iIPN–DT pathway (blue) and the dHbM–vIPN–median raphe pathway (orange) are involved in fear behavior. (**Bottom**) Selective inactivation of dHb–IPN pathways affects behavior. Silencing of the projection from dHbL onto d/iIPN triggers the fish to lose (left); silencing of the projection from dHbM onto vIPN triggers the fish to win (right). Naïve fish are those who did not previously fight (middle).

In mammals, the habenula is a multitask brain region that integrates aversive inputs processed by the limbic system and the basal ganglia and adapts motor responses accordingly (*5*). To find homologous structures in zebrafish, Chou *et al.* drew upon knowledge of connections in the mammalian brain. Recent studies using anterograde tracers in zebrafish had provided evidence for the conservation of habenular pathways (*6*) and associated functions (*7*). The dHb–IPN pathway of zebrafish corresponds to the medial Hb–IPN pathway of mammals and is similarly involved in fear responses (*7*, *8*). Furthermore, the dIPN projects onto the dorsal tegmental (DT) region corresponding to the mammalian periaqueductal gray (PAG) (*7*). The PAG regulates fleeing behavior in mammals (*9*), providing further support for the idea that the zebrafish circuit contributes to loser-like behavior.

Chou *et al.* used transgenic zebrafish lines to target neurons of either the lateral or medial subnucleus of the dHb (dHbL and dHbM) and block their synaptic transmission to distinct regions of the IPN. Reduction of transmission in one pathway (dHbL–d/iIPN) increased the tendency to lose fights, whereas in the other pathway, dHbM–ventral IPN (vIPN), the tendency to lose was decreased. Selective manipulations of habenular transmission to the IPN were therefore sufficient to determine the winner (and loser) without prior experience—an effect that persisted over time and was reinforced during subsequent fights. These results suggest that the dHb–IPN pathway acts as a neural switch that conditions the outcome of social fighting, depending on differential transmission onto dorsal or ventral IPN subnuclei. This study, however, does not rule out the potential role of many other brain regions acting in concert with the habenula (*10*).

Because the manipulation of synaptic transmission within the dHb–IPN pathway led to antagonistic outcomes during social

[1]Institut du Cerveau et de la Moelle épinière, GH Pitié-Salpêtrière, 75013 Paris, France. E-mail: claire.wyart@icm-institute.org

ILLUSTRATION: K. SUTLIFF/*SCIENCE*

fighting, the habenula seemingly can induce differential motor responses. This is concordant with recent studies describing the habenula as a hub integrating multiple sensory cues (*11*) to produce an adapted motor response. How this signal is relayed at the molecular and neuronal level is not yet known. Multiple modulatory neurotransmitters and peptides are associated with aggressive behavior (*10*, *12*), but their diversity in the habenula and IPN is most likely underappreciated. The dissection of neuromodulatory systems using innovative genome-editing technologies may reveal the full circuitry governing social defeat in vertebrates.

Zebrafish has emerged as a powerful system to study social interactions (*13*), and there are many social behaviors in this animal to unravel (14). For example, the recent observation of social preference in juvenile zebrafish (*15*), at stages when whole-brain imaging is feasible, opens new paths of investigation into the sensory cues and the genes and circuits mediating these early social interactions. It should be possible to find out why some individuals are reluctant to go toward others when the majority are attracted by conspecifics.

Although zebrafish have a short generation time and are easy to maintain in captivity, the bottleneck often lies in the fine description and quantitative analysis of complex behaviors. The study of Chou *et al.* illustrates how the genetic manipulation of pathways governing innate behaviors in zebrafish can inspire investigations in other model organisms by providing new working hypotheses and brain regions to target. The next step will be to combine whole-brain imaging, optogenetic manipulations, and CRISPR-mediated genome editing to unravel the complete circuits underlying social behaviors in vertebrates. ■

REFERENCES

1. I. D. Chase, K. Seitz, *Adv. Genet.* **75**, 51 (2011).
2. C. Rutte, M. Taborsky, M. W. Brinkhof, *Trends Ecol. Evol.* **21**, 16 (2006).
3. M.-Y. Chou *et al.*, *Science* **352**, 87 (2016).
4. R. F. Oliveira, J. F. Silva, J. M. Simoes, *Zebrafish* **8**, 73 (2011).
5. O. Hikosaka, *Nat. Rev. Neurosci.* **11**, 503 (2010).
6. R. Amo *et al.*, *J. Neurosci.* **30**, 1566 (2010).
7. M. Agetsuma *et al.*, *Nat. Neurosci.* **13**, 1354 (2010).
8. T. Yamaguchi, T. Danjo, I. Pastan, T. Hikida, S. Nakanishi, *Neuron* **78**, 537 (2013).
9. R. Misslin, *Neurophysiol. Clin.* **33**, 55 (2003).
10. R. F. Oliveira *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **113**, E654 (2016).
11. E. Dreosti, N. Vendrell Llopis, M. Carl, E. Yaksi, S. W. Wilson, *Curr. Biol.* **24**, 440 (2014).
12. T. N. deCarvalho *et al.*, *Genesis* **52**, 636 (2014).
13. R. F. Oliveira, *Front. Neural Circuits* **7**, 131 (2013).
14. R. D. Fernald, *Cold Spring Harb. Symp. Quant. Biol.* **79**, 229 (2014).
15. E. Dreosti, G. Lopes, A. R. Kampff, S. W. Wilson, *Front. Neural Circuits* **9**, 39 (2015).

10.1126/science.aaf6016

ILLUSTRATION: P. HUEY/*SCIENCE*

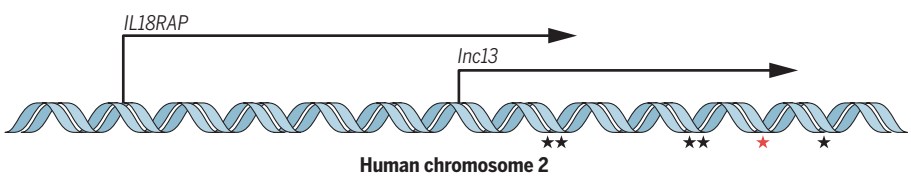
RNA

A lncRNA links genomic variation with celiac disease

A long noncoding RNA is associated with an intestinal autoimmune disorder

By **Maite Huarte**

The majority of human single-nucleotide polymorphisms (SNPs) associated with increased disease risk map to noncoding regions of the genome. The nucleotide variations therefore cannot be directly related to changes in the function of proteins. Indeed, SNPs frequently localize to DNA regulatory elements such as enhancers or promoters, or within intergenic regions that are transcribed to produce long noncoding RNAs (lncRNAs). LncRNAs are RNA molecules longer than 200 nucleotides that do not encode proteins; in many instances, they regulate gene expression through diverse mechanisms. On page 91 of this issue, Castellanos-Rubio *et al.* (*1*) report that a relationship between the function of a lncRNA and the SNPs within its locus underlies celiac disease, an autoimmune disorder that causes intolerance to gluten.



Location overlap. The genomic loci of *IL18RAP* and *lnc13* overlap in human chromosome 2. SNPs associated with celiac disease are indicated with blue stars. A red star shows the specific SNP, rs917997.

Genome-wide association studies revealed that six SNPs that are linked to increased risk of suffering from gluten intolerance form a haploblock (closely associated SNPs) located in chromosome 2 (*2*). One of the SNPs (rs917997) is positioned 1.5 kb downstream of interleukin-18 receptor accessory protein (IL18RAP), a gene that has been associated with susceptibility to celiac disease and other autoimmune diseases (*2*–*4*). In the mouse genome, a previously annotated lncRNA, *lnc13*, is transcribed in this position. Castellanos-Rubio *et al.* identified a lncRNA in the equivalent position of the human genome and showed that both human and murine lnc13 regulate the expression of genes of the inflammatory response. Despite limited DNA sequence conservation, the function and mechanism of lnc13 is conserved between mouse and human.

The fine regulation of lnc13 expression is probably crucial for its proper function. It is transcribed from the same strand of DNA as IL18RAP, and its 5′ end overlaps with the 3′ end of IL18RAP (see the first figure). However, despite this overlap, both genes have independent promoters and are independently regulated. IL18RAP expression is induced in the intestinal epithelium of celiac patients and in lipopolysaccharide (LPS)–stimulated macrophages (a treatment that mimics the inflammatory response). However, under these conditions, the expression of lnc13 is diminished. Reduction in the amount of lnc13 is a posttranscriptional event dependent on the signaling pathway controlled by the transcription factor nuclear factor kappa B (NF-κB). Indeed, despite not being translated, lnc13 is highly stable in nonstimulated macrophages. By contrast, in LPS-activated macrophages, NF-κB induces the expression of Dcp2, a negative regulator of the stability of capped RNAs. Dcp2 thereby keeps lnc13 amounts low (see the second figure). Thus, the co-inherited IL18RAP and lnc13 are transcribed from the same locus and take part in the same biological process. However, their regulation is independent, in part because of the posttranscriptional control of lnc13.

The expression levels of lnc13 correlate negatively with the expression of several genes of the inflammatory response, some of which are up-regulated in celiac disease patients' biopsies. In fact, lnc13 inhibits the expression of these genes in cells where the inflammatory response is inactive. Similar to several other characterized lncRNAs, lnc13 is preferentially present in the nucleus

A brain conditioned for social defeat

Laura Desban and Claire Wyart

Science **352**, 42 (2016);
DOI: 10.1126/science.aaf6016