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The Aryl Hydrocarbon Receptor *sans* Xenobiotics: Endogenous Function in Genetic Model Systems

Brian J. McMillan and Christopher A. Bradfield

McArdle Laboratory for Cancer Research, University of Wisconsin School of Medicine and Public Health, 1400 University Avenue, Madison, WI 53706, USA; BJM, CAM

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Running title: Endogenous AHR signaling

Corresponding author:
Christopher A. Bradfield
1400 University Avenue, Room 213, Madison, Wisconsin 53706
Tel: (608) 262-2024; Fax (608) 262-2824
Email: Bradfield@oncology.wisc.edu

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Abbreviations: AHR, aryl hydrocarbon receptor; ARNT, Aryl hydrocarbon Receptor Nuclear Translocator; BNF, beta-napthoflavone; da, dendritic arborization; LOF, loss-of-function; PAS, Per-ARNT-Sim; PNS, peripheral nervous system; PR, photoreceptor; Ss, spineless; Tgo, Tango; TCDD, 2,3,7,8-tetrachlorodibenzo-*p*-dioxin; TGF-beta, transforming growth factor beta

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Abstract

For over thirty years, the aryl hydrocarbon receptor (AHR, Ah receptor) has been extensively scrutinized as the cellular receptor for numerous environmental contaminants, including dioxins, dibenzofurans and polychlorinated biphenyls (PCBs). Recent evidence argues that this description is incomplete and, perhaps, myopic. Ah receptor orthologs have been demonstrated to mediate diverse endogenous functions in our close vertebrate relatives as well as our distant invertebrate ancestors. Moreover, these endogenous functions suggest that xenobiotic toxicity may be best understood in the context of intrinsic AHR physiology. In this literature review, we survey the emerging picture of endogenous AHR biology from work in the vertebrate and invertebrate model systems *Mus musculus, Caenorhabditis elegans,* and *Drosophila melanogaster.*

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The aryl hydrocarbon receptor (AHR, Ah receptor) is a founding member of the bHLH-PAS superfamily of transcriptional regulators (for a review of PAS biology see Gu et al., 2000). This protein was originally identified and characterized due to its central role in the vertebrate response to many planar aromatic hydrocarbons (summarized in Swanson and Bradfield, 1993). In this capacity, the AHR binds exogenous ligand and transcriptionally activates a battery of enzymes that promote metabolic transformation and excretion of the xenobiotic from the organism (Nebert et al., 2004; Uno et al., 2004; Xu et al., 2005). This AHR-mediated pathway is commonly viewed as an "adaptive" response towards these xenobiotic agents. In addition to adaptive metabolism, the AHR also mediates a spectrum of toxic endpoints in response to high affinity agonists. The most potent known ligand for the AHR, 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD, dioxin), is highly resistant to metabolic degradation and elicits numerous AHR-dependent toxic events, including late stage terata, thymic atrophy, chloracne, tumor promotion, hepatomegaly, cachexia and death (reviewed in Birnbaum and Tuomisto, 2000; Schecter et al., 2006).

In order to elucidate the role of the AHR in normal physiology and development, the locus was deleted from the mouse genome (Fernandez-Salguero et al., 1995; Mimura et al., 1997; Schmidt et al., 1996). In addition to being refractory to most, if not all, aspects of TCDD toxicity, "*Ahr*-knockouts" exhibit multiple physiological abnormalities that appear independent of xenobiotic exposure (Fernandez-Salguero et al., 1995; Mimura et al., 1997; Schmidt et al., 1996). Studies of the mutant mouse models have reproducibly demonstrated that the AHR has an intrinsic role in diverse aspects of mammalian biology that extend beyond (or are entirely independent from) xenobiotic metabolism (Abbott et al., 1999; Andreola et al., 1997; Baba et al.,

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2005; Benedict et al., 2000; Benedict et al., 2003; Corchero et al., 2004; Fernandez-Salguero et al., 1997; Guo et al., 2004; Lahvis et al., 2000; Lahvis et al., 2005; Lund et al., 2003; Lund et al., 2005a; Thackaberry et al., 2002; Vasquez et al., 2003; Zaher et al., 1998). Moreover, these studies of null models have led to speculation that aspects of TCDD toxicity may be the result of sustained interference in these endogenous, physiological pathways mediated by the AHR.

Evolutionary studies have revealed that the *Ahr* gene is highly conserved across both vertebrate and invertebrate species. (for a review of AHR diversity and evolution see Hahn, 2002 and Hahn et al., 2006). Furthermore, this research suggests that aryl hydrocarbon metabolism was not involved in the origins of AHR function. Several groups have cloned ancestral AHR orthologs, which either do not bind any ligand or bind a spectrum of ligands that is qualitatively different than their vertebrate counterparts (Butler et al., 2001; Duncan et al., 1998; Hahn, 2002; Powell-Coffman et al., 1998; Wiesner L, 2001). If aryl hydrocarbon metabolism is a "recent" adaptation of the vertebrate lineage, what are the original functions of the AHR? How have these functions changed in response to evolutionary adaptation? Are these endogenous functions related to the toxicity of potent xenobiotic agonists like TCDD?

In an effort to answer these questions, we have summarized what is currently known regarding endogenous function of the vertebrate AHR and its invertebrate orthologs. Forward and reverse genetic studies in *Caenorhabditis elegans* (nematode), *Drosophila melanogaster* (fruit fly), and *Mus Musculus* (mouse) have identified essential roles for AHR signaling in multiple aspects of normal physiology, including vascular development, reproduction, neural function, limb patterning and vision. We propose that a comprehensive review of these phenotypes will foster a broader understanding of AHR signaling in relation to normal

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physiology as well as TCDD toxicity, which has eluded explanation despite decades of extensive research.

AHR structure and mechanism. Most insight into the mechanisms of AHR signaling has been derived from studies in mammalian cells following xenobiotic induction. These studies have indicated that AHR-mediated transcription occurs via two main steps: (i) ligand activation, which facilitates nuclear translocation of the AHR, and (ii) ARNT heterodimerization, which facilitates gene transcription (Figure 1). The latter stage appears to be highly conserved amongst all species expressing the AHR and its orthologs (Emmons et al., 1999; Prasch et al., 2005; Qin et al., 2006). The role of ligand-binding in ancestral orthologs of the AHR, however, remains uncertain. In mammalian cells, unliganded AHR is primarily found in a cytoplasmic complex, which includes an Hsp90 homodimer and the immunophilin-like Ara9, also known as XAP2 or AIP (the AHR:chaperone complex is reviewed in Petrulis and Perdew, 2002). Ligand binding increases nuclear shuttling of the receptor via a process presumed to involve structural transformation and/or nuclear localization sequence (NLS) presentation (Henry and Gasiewicz, 2003; Ikuta et al., 1998; Petrulis et al., 2003).

Once in the nucleus, the Per-ARNT-Sim (PAS) domain of AHR mediates dimerization with the corresponding PAS domain of a constitutively nuclear protein known as ARNT (Aryl hydrocarbon Receptor Nuclear Translocator) (Gu et al., 2000). The basic-Helix-Loop-Helix (bHLH) domains present in both AHR and ARNT then facilitate contact with gene regulatory elements found in DNA, commonly referred to as Dioxin or Xenobiotic Response Elements (DRE or XRE, respectively) (Bacsi and Hankinson, 1996; Gu et al., 2000; Murre et al., 1989;

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Swanson et al., 1995). Several genes that contain functional DRE sequences have been shown to encode enzymes, such as the *Cyp1* family of Cytochromes P450, that mediate metabolism of exogenous AHR ligands. The preponderance of AHR-regulated genes, however, has no known role in AHR biology, either xenobiotic or endogenous.

Caenorhabditis elegans

The *C. elegans* ortholog of the mammalian AHR, called AHR-1, shares 38% amino acid identity with the human AHR over the N-terminal region that mediates DNA binding, PAS dimerization, ligand binding, and interactions with cytoplasmic chaperones (Powell-Coffman et al., 1998). The nematode genome also contains an ortholog of the mammalian *Arnt*, which was cloned under the locus name *aha-1* (Powell-Coffman et al., 1998). Similar to ARNT, AHA-1 functions as a signaling partner for multiple PAS proteins, including the *C. elegans* homologs of AHR and HIF1α (Jiang et al., 2001; Powell-Coffman et al., 1998). In contrast to what is commonly thought for mammalian ARNT, nuclear localization of AHA-1 is dependent on PAS heterodimerization (Jiang et al., 2001).

In vitro studies have shown that AHR-1 and AHA-1 share several key biochemical features with their predicted mammalian orthologs. In rabbit reticulocyte lysates, AHR-1:AHA-1 as well as AHR-1:ARNT form a transcriptionally active complex that binds to canonical DRE sequences (Powell-Coffman et al., 1998). In addition to ARNT dimerization, AHR-1 is also capable of interacting with the mammalian chaperone Hsp90 (Powell-Coffman et al., 1998). The *C. elegans* homolog of Hsp90, however, appears to be dispensable in AHR-1 function (Huang et al., 2004; Qin et al., 2006). Like the vertebrate AHR, a series of AHR-1 deletion mutants has

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indicated that the PAS domain represses nuclear translocation and/or transcriptional activity when expressed in yeast (Powell-Coffman et al., 1998). In mammalian AHR signaling, PAS-mediated repression is ablated via ligand binding and subsequent structural transformation of the receptor; these results suggest that post-translational PAS modification could also be an integral part of AHR-1 signaling. However, AHR-1 does not bind traditional ligand modifiers of the vertebrate PAS domain such as TCDD analogs or β-napthoflavone (βNF) (Butler et al., 2001; Powell-Coffman et al., 1998). To date, no ligand agonist of AHR-1--or any invertebrate AHR homolog--has been identified.

Expression. To elucidate the expression pattern of AHR-1, transgenic worms have been generated with an AHR-1::GFP fusion protein driven by *ahr-1* regulatory elements (Qin and Powell-Coffman, 2004). Fusion protein expression is detectable in the early stages of embryonic development, approximately 260 minutes after the first cell cleavage (for a searchable review of *C. elegans* development and physiology see Donald L. Riddle, 1997, available free online). By the first larval stage, 35 cells express observable levels of GFP, including blast cells, phasmid socket cells, and numerous neurons belonging to multiple subtypes (Qin and Powell-Coffman, 2004). No function has yet been identified for AHR-1 in the blast or phasmid socket cells. Several neuronal phenotypes, however, have been observed in animals homozygous for loss of function (LOF) mutants of AHR-1 (Table 1) (Huang et al., 2004; Qin and Powell-Coffman, 2004; Qin et al., 2006).

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Neuron development. A role for AHR-1 in neuron development was first identified in the GABAergic ring motor (RME) neurons, which innervate the nematode head muscles (McIntire et al., 1993; White et al., 1986). The RME cluster is comprised of a set of four cells distributed around the neck ring in a symmetrical manner. Although sharing similar neurotransmitter specificity and the same synaptic targets, the RME neurons can be further divided into two subgroups (left/right and dorsal/ventral) based on cell lineage, axon extension, and gene expression--including that of *ahr-1*, which is expressed exclusively in the left/right subset (Dent et al., 1997; Hart et al., 1995; Huang et al., 2004). From a forward genetic screen, it was determined that loss of AHR-1 function results in all four RME cells adopting the dorsal/ventral phenotype, including axon morphology and gene expression (Huang et al., 2004). Conversely, ectopic expression of *ahr-1* in the dorsal/ventral cells was found to enforce a left/right morphology and gene expression pattern, suggesting that AHR-1 functions as a binary switch to determine RME subtype (Huang et al., 2004).

In addition to RME biology, other investigators have utilized reverse genetic techniques to determine the broader role of AHR-1 in nematode development (Qin and Powell-Coffman, 2004). In these studies, several neurons from multiple lineages were found to be mislocalized in the adult due to faulty migration during larval development (for details see Table 1) (Qin and Powell-Coffman, 2004). Moreover, AHR deficiency was found to dysregulate expression of several key genes found in the pseudocoelomic neurons that regulate aspects of foraging behavior (Qin and Powell-Coffman, 2004). These initial observations have since been extended to demonstrate an important role for the AHR in oxygen-sensitive aggregation (Qin and Powell-Coffman, 2004; Qin et al., 2006).

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Feeding behavior. Wild isolates of *C. elegans* exhibit either solitary or social (aggregated) feeding behavior on bacterial lawns, depending on natural allelic variation (de Bono and Bargmann, 1998; Rogers et al., 2003). This behavior is determined, in part, by the strain's relative threshold to ambient oxygen (Cheung et al., 2004; Gray et al., 2004). Some strains are more sensitive to oxygen and thus aggregate to reduce exposure, whereas others seek out a more oxygen-rich solitary environment. This oxygen-sensing pathway is expressed in four neurons (AQR, PQR, URXR and URXL), which directly contact the pseudocoelomic fluid (Coates and de Bono, 2002). Aggregated feeding is induced via neuron depolarization, which is positively regulated by the oxygen-binding heterodimer of soluble guanylyl cyclases 35 and 36 (GCY-35, GCY-36) and attenuated via the neuropeptide receptor, NPR-1 (Cheung et al., 2004; de Bono and Bargmann, 1998; Gray et al., 2004) (see Figure 2 for details).

Social feeding is highly suppressed in AHR-1 LOF mutants (Qin and Powell-Coffman, 2004; Qin et al., 2006). This behavior can be restored, however, via *ahr-1* expression in the pseudocoloemic sensors URXR and URXL (Qin et al., 2006). In contrast to the other neuronal phenotypes observed in AHR-1 mutants, defects in URX-mediated behavior are due to an ongoing role for AHR-1 in mature cell function--not neuron development or differentiation. Studies utilizing temporal control of *ahr-1* expression via heat shock have demonstrated that developmental expression is not required for wildtype feeding behavior (Qin et al., 2006). Current evidence indicates that AHR-1 controls feeding behavior via indirect transcriptional control of both NPR-1 and several members of the soluble guanylyl cyclase (sGC) family, including the oxygen-binding heterodimer GCY-35:GCY-36 (Qin and Powell-Coffman, 2004;

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Qin et al., 2006) (Figure 2). Overall, these results indicate that AHR-1 mutation leads to widespread defects in the pathways that regulate pseudocoelomic cell depolarization. The mechanism underlying AHR-1 regulation of these genes, however, is currently unclear. Although putative DREs are present in the regulatory regions of *gcy-35* and *-36*, mutation of these sequences does not alter dysregulation by AHR-1 LOF suggesting the involvement of a yet unidentified transcriptional intermediate (Qin et al., 2006).

Drosophila melanogaster

The *Drosophila* ortholog of the AHR, called Spineless (Ss), shares all splicing junctions and 41% amino acid identity with its murine counterpart (Duncan et al., 1998). A higher degree of identity is conserved in the bHLH (71%) and PAS (45%) regions (Duncan et al., 1998). Spineless heterodimerizes with an ARNT ortholog called Tango (Tgo) that, similar to AHA-1, requires concomitant expression of a PAS signaling partner for nuclear localization (Emmons et al., 1999; Ward et al., 1998). Like other invertebrate homologs of AHR, Spineless does not bind prototypical xenobiotic ligands of the vertebrate receptor such as TCDD or βNF (Butler et al., 2001). *In vitro* co-expression studies have shown that the Ss:Tgo complex induces DRE-mediated transcription in the absence of exogenous ligand in both *Drosophila* and yeast cell lines, indicating that either (i) post-translational modification of Spineless via ligand is not required, or (ii) endogenous ligand is ubiquitously produced (Emmons et al., 1999 and unpublished observation from our lab). Consistent with these *in vitro* observations, ectopic expression of Spineless in the fly has been shown to result in nuclear translocation of Tango independent of tissue/cell type (Emmons et al., 1999).

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Expression. *Spineless* is expressed in numerous tissues during embryonic and pupal development (for a searchable review of *Drosophila* development see Gilbert, 2000, available free online). *Spineless* mRNA first appears at the 8th stage of embryonic development just anterior to the cephalic furrow (Duncan et al., 1998). As developmental segmentation proceeds, *ss* is expressed in the maxillary, labial, and mandibullar segments in the head, the leg anlages of the thorax, and the peripheral nervous system in each thoracic and abdominal segment (Duncan et al., 1998). In the pupa, *ss* is expressed in multiple imaginal discs, which develop into the adult leg, antenna, palp, wing, haltere, eye and genital structures, as well as bristle cell precursors (Adachi-Yamada et al., 2005; Duncan et al., 1998; Wernet et al., 2006).

Homeotic function. Research into the developmental functions of Spineless has a long history. Calvin Bridges and T.H. Morgan first identified the *ss* locus in 1923 and it has since been recognized as an essential regulator of appendage identity (Bridges, 1923; Burgess and Duncan, 1990; McMillan and McGuire, 1992; Struhl, 1982). *Spineless* belongs to a framework of loci, known as homeotic genes, that direct development of each segment into specialized structures. In simplistic terms, homeotic genes function as regulatory switches that direct pluripotent cell clusters into specialized structures, such as the leg or antenna or wing. Mutations to homeotic genes can lead to deletion and/or transformation of appendages to a homologous structure, e.g. a leg developing where an antenna should be. *Spineless* is expressed in many of these developing regions and, in general, predisposes cells towards the antennal fate. Like many homeotic genes, however, the developmental program of Spineless is highly complex. Recent work has shown

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that *spineless* functions in a spatio-temporal fashion both upstream (*dan*/*fer*, *danr*/*hern*, *bric-a-brac*, *BarH1* and *BarH2*) and downstream (*distal-less*, *homothorax*, *cut*) of multiple homeotic genes, each of which has their own complex expression patterns and regulatory networks (Chu et al., 2002; Emerald et al., 2003; Emmons et al., 2006; Kozu et al., 2006; Suzanne et al., 2003). Experimental dysregulation of *spineless*, therefore, has varied consequences that are largely dependent on the strength of mutation or driver of expression. Ectopic expression of *spineless* via the *ptc*-GAL4 driver has been shown to cause transformation of the maxillary palp, rostral membrane, and distal leg to antennal identity (Duncan et al., 1998). Conversely, deletion of *ss* or *tgo* results in a variety of defects, including transformation of the distal antenna to a leg-like structure, truncation of the maxillary palps, deletion of the medial tarsal structures of the leg, and markedly reduced bristle size (Figure 3) (Duncan et al., 1998; Emmons et al., 1999; Lindsley and Zimm, 1992; Struhl, 1982). Overall, these results indicate that Spineless directs appendages from an epigenetic ground state into a distal antenna identity, but also suggest a distinct role in the elongation/elaboration of non-antennal structures, as well.

Dendrite diversification. Similar to AHR-1, Spineless appears to have an essential role in neuronal development. In this regard, Spineless has recently been identified as a central regulator of peripheral nervous system (PNS) morphology (Kim et al., 2006). The *Drosophila* PNS, replicated in each thoracic and abnominal segment, is comprised of ≈ 40 neuronal cells containing a range of dendrite complexity from single extensions to highly branched structures (Bodmer, 1987). The dendritic arborization (da) class of PNS neurons displays highly elaborate dendrite structures that innervate the epidermal surface and transmit signals relating to

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locomotion, thermosensation, and pain (Ainsley et al., 2003; Liu et al., 2003; Tracey et al., 2003). This family of neurons is commonly categorized into four classes based upon increasing dendrite complexity; class I contains the simplest da neurons, whereas class IV contains the most complex structures (Grueber et al., 2002). The process of dendrite branching is influenced via numerous developmental pathways (Brenman et al., 2001; Grueber et al., 2003; Li et al., 2004; Parrish et al., 2006; Sugimura et al., 2004). The regulatory genes that have been studied in detail have generally been classified as promoters or inhibitors of branching complexity, such as *cut* and *abrupt* (respectively), which are preferentially expressed in opposing neuron classes (Grueber et al., 2003; Li et al., 2004; Sugimura et al., 2004). To date, the effects of Spineless on dendrite morphogenesis are unique in that *ss* is equally expressed in all da classes and LOF results in opposing effects: increased branching in the "simple" Class I and II neurons, but reduced branching in "complex" Class III and IV cells (Kim et al., 2006). Moreover, loss of *ss* expression attenuates class-specific features such as Class III actin spikes contributing to the idea that *ss* LOF promotes a "generic" da form (Kim et al., 2006). Overall, these results are consistent with the idea that Spineless has an essential role in the diversification of neuron morphology and, potentially, function. The mechanism underlying this developmental pathway, including the role of Tango, is currently unclear. Cursory experiments indicate that Tango may be dispensable for normal dendrite morphogenesis (Kim et al., 2006). The possibility that Spineless controls PNS branching independent of Tango, however, has not been confirmed using more robust methodology.

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Photoreceptor development. A recent report has further implicated Spineless signaling in sensory cell diversification. In this study, *spineless* expression was demonstrated to be a master regulator of the *Drosophila* photoreceptor mosaic required for color vision (Figure 4) (Wernet et al., 2006). In the fly, vision is achieved via a compound eye composed of ≈ 800 optical units (ommatidia), each of which contains eight photoreceptor cells (PRs) (Gilbert, 2000). The six outer PRs (R1-R6) are functionally analogous to the vertebrate rod cells that sense motion in dim light (Miller et al., 1981; Wernet et al., 2006) whereas the two inner cells (R7 and R8) express color-sensitive rhodopsins (Rh) similar to the vertebrate cone cell (Chou et al., 1996; Fryxell and Meyerowitz, 1987; Huber et al., 1997; Papatsenko et al., 1997). The color-sensing R7/R8 photoreceptor pair comes in two "flavors", which are randomly distributed throughout the compound eye. In wildtype animals, the R7 cell in each ommatidium stochastically expresses either *Rhodopsin 3* (*Rh3*) or *Rh4*, which then instructs R8 cells to express *Rh5* or *Rh6*, respectively (Chou et al., 1996; Chou et al., 1999). The resulting mosaic of ommatidia produces a compound eye capable of recognizing a broad spectrum of light. The Rh4/Rh6 rhodopsin combination present in 70% of ommatidia recognizes longer or "yellow" light wavelengths, whereas the remaining ommatidia express Rh3/Rh5 receptors that sense shorter "pale" wavelengths (Feiler et al., 1992).

Although *spineless* null animals exhibit no gross morphological changes to the eye, their ommatidia were found to be comprised solely of Rh3/Rh5, i.e. "pale", photoreceptors (Figure 4) (Wernet et al., 2006). Conversely, forced expression of *spineless* was found to induce a 100% Rh4/Rh6, i.e. "yellow", eye indicating that *ss* is both necessary and sufficient for the "yellow" photoreceptor phenotype (Wernet et al., 2006). Temporal regulation of *ss* via heat shock further

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revealed that a single burst of *ss* during mid-pupation--similar to *ss* expression during normal development--was sufficient to program adult photoreceptor phenotype (Duncan et al., 1998; Wernet et al., 2006). During normal development, however, an eye-specific enhancer region present in the *ss* promoter confers stochastic *ss* expression and, thus, stochastic cell fate (Wernet et al., 2006). Overall, these findings demonstrate that Spineless expression in the *Drosophila* eye functions as a binary (but stochastic) switch, which produces the ommatidial mosaic necessary for color vision.

Mus Musculus

Expression. The aryl hydrocarbon receptor is expressed in numerous murine embryonic and adult tissues. *Ahr* expression can be observed as early as gestation day 10-12 in the nasal pit, branchial arches, heart, liver, maxillary prominence, neuroepithelium and several neurons, including the trigeminal ganglion, spinal ganglia, and posterior branches of spinal nerves (Abbott et al., 1995; Birnbaum and Perdew, 1995; Mimura et al., 1997). Given the role of *ahr-1* and *spineless* in neuron development, it is interesting to note that brain/neural expression of mouse *Ahr* largely dissipates as gestation proceeds (Abbott et al., 1995). From gestation day 13.5 to 15.5, the overall range of *Ahr* expression is greatly expanded and can be detected in the developing pituitary, choroid plexus, adrenal gland, palatal shelf, dorsal surface of the tongue, nasal cartilage, olfactory tissues, thymus, lung, liver, bone, muscle, gut, kidney, retina, bladder epithelium, urogenital sinus and tip of the genital tubercle (Abbott et al., 1995; Jain et al., 1998) (Figure 5A). In adult 8-11 week old mice, *Ahr* is most highly expressed in the oocyte, epidermis, bladder, lung, digits, vomeronasal organ, liver, trachea, olfactory epithelium, and retina (Su et

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al., 2002) (Figure 5B). Xenobiotic studies, however, have demonstrated the presence of functional AHR in countless other tissues and cell types.

Knockout. Three independent laboratories have generated *Ahr* null mice via excision of either the first or second exon of the gene (Fernandez-Salguero et al., 1995; Mimura et al., 1997 and Schmidt et al., 1996, respectively). Although all models share important characteristics, such as resistance to TCDD toxicity, reduced fecundity, portal fibrosis and smaller livers, they also appear to differ in some respects, such as lymphoid cell number and age-related lesions (reviewed in Lahvis and Bradfield, 1998). For the purpose of this review, we have focused on the *in vivo* phenotypes that are shared amongst all *Ahr* null constructs. Alterations in cultured cell phenotype have been reviewed elsewhere (Barouki et al., 2007).

Cardiovascular. Perhaps the most overt phenotype of the AHR knockout mouse is a markedly reduced liver size (25 – 50% smaller than controls) (Fernandez-Salguero et al., 1995; Schmidt et al., 1996). Our own studies have indicated that this liver phenotype is the result of defects in the resolution of fetal vascular structure (Lahvis et al., 2000; Lahvis et al., 2005). In the embryonic vasculature, the flow of blood partially bypasses the liver sinusoids via a shunt known as the ductus venosus (DV), which directly connects the inferior vena cava (IVC) and the portal vein (PV) (Edelstone et al., 1978; Kiserud et al., 2000). During normal development, the DV closes shortly after birth forcing oxygen- and nutrient-rich blood to migrate through the liver sinusoids. In the absence of AHR signaling, however, the DV remains patent (open) throughout adulthood and is presumed to result in reduced postnatal liver growth via nutrient deprivation (Figure 6A)

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(Lahvis et al., 2000; Lahvis et al., 2005). A series of follow-up experiments have demonstrated that this physiological role for the AHR is mechanistically analogous to xenobiotic activation. DV closure depends on co-expression of ARNT (Walisser et al., 2004b), the nuclear localization/DNA-binding domain of AHR (Bunger et al., 2003), as well as the degree of AHR activation (Walisser et al., 2004a). Most recently, experiments using a conditional allele of *Ahr* have demonstrated that the DV phenotype fully depends on endothelial and/or hematopoeitic, but not hepatocyte, expression of the receptor (Walisser et al., 2005).

In addition to the DV phenotype, AHR null mice exhibit several other less characterized alterations to embryonic, neonatal and adult cardiovascular biology. In embryos, a reduction in liver perfusion can be observed as early as gestation day 15.5, indicating that vascular abnormalities precede normal DV closure (Harstad et al., 2006). In neonates, *Ahr* nulls have abnormalities in the vascular architectures of the kidney (Figure 6B), liver sinusoid, and eye, including persistence of the embryonic hyaloid artery (Lahvis et al., 2000). In adults, the knockout genotype is linked to cardiac hypertrophy, hypertension, and elevated levels of the potent vasoconstrictors endothelin-1 (ET-1) and angiotensin II (Ang II) (Fernandez-Salguero et al., 1997; Lund et al., 2003; Lund et al., 2005a; Lund et al., 2005b; Thackaberry et al., 2002; Vasquez et al., 2003). Studies using pharmacological inhibitors of ET-1/Ang-II signaling have suggested that these vasoconstrictors directly contribute to the age-dependent increase in blood pressure and heart size (Lund et al., 2003; Lund et al., 2005a; Lund et al., 2005b). It is unclear, however, if these age-related changes are due to a continuing role for the AHR in vascular homeostasis or simply a downstream consequence of the altered liver and/or kidney vasculature.

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Several lines of evidence indicate that vascular AHR signaling is mechanistically linked to fluid shear stress. Shear forces are generated via the passage of fluid, i.e. blood, through a constricted space such as the vascular system. This process generates (i) a tangential, frictional force against the inner wall of the blood vessel, which is highly studied due to its direct effects on the vascular endothelium (Resnick et al., 2003), and (ii) a velocity gradient within the fluid itself, which has the capacity to alter serum protein structure and function (Alexander-Katz et al., 2006; Shankaran et al., 2003; Siedlecki et al., 1996). Independent reports have demonstrated that the AHR is highly activated by cellular exposure to fluid shear (McMillan and Bradfield, 2007; Mufti et al., 1995). Moreover, recent evidence indicates that fluid shear stress activates the AHR, at least in part, via a direct effect on serum low-density lipoprotein (LDL) function and/or structure (McMillan and Bradfield, 2007). Interestingly, the role of modified LDL in AHR biology does not appear to be limited to shear-induced modification. Conventional methods of LDL modification, such as hypochlorite oxidation, also produce an AHR-activating isoform of this pro-atherogenic macromolecule (McMillan and Bradfield, 2007). In light of this recent data as well as the cardiovascular phenotype of the *Ahr* null mouse, we speculate that vascular AHR signaling could function as a developmental mechanism to increase blood flow, and thus serum/LDL filtration, through both the liver and kidney.

Fibrosis. In addition to vascular defects, several other pathologies have been observed in the *Ahr* null liver including prolonged extramedullary hematopoeisis, fatty metamorphosis, and portal tract fibrosis (Fernandez-Salguero et al., 1995; Schmidt et al., 1996). Of these changes, only portal fibrosis has been characterized in a more detailed fashion. Data suggest that interplay

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between the AHR, retinoic acid, and TGF-β pathways contribute to this fibrotic phenotype. Studies in cultured cells as well as the liver have shown that AHR deficiency results in increased secretion of active TGF-β, a potent pro-fibrotic peptide (Border and Noble, 1994; Branton and Kopp, 1999), via a post-translational mechanism (Santiago-Josefat et al., 2004; Zaher et al., 1998). This process likely involves direct transcriptional regulation of the latent TGF-β binding protein-1 (LTBP-1) (Corchero et al., 2004; Gomez-Duran et al., 2005; Santiago-Josefat et al., 2004) as well as deficiency in retinoid metabolism (Andreola et al., 2004; Andreola et al., 1997), which has been shown to increase levels of TGF-β activating enzymes (Kojima and Rifkin, 1993; Okuno et al., 1997). Studies outside the liver have also indicated interaction between the TGF-β and AHR pathways, suggesting that additional endpoints of endogenous biology may also be affected by this relationship (Guo et al., 2004; Thomae et al., 2005).

Reproduction. AHR null females exhibit defects in multiple aspects of reproduction, including conception, litter number, and pup survival (Abbott et al., 1999; Baba et al., 2005). Accordingly, the AHR has been found to have a physiological role in several reproductive tissues, most notably the ovarian follicle. During normal estrous, hormonal signals induce maturation of the granulosa cell layer surrounding the oocyte and trigger release of the follicle into the fallopian tube (Gilbert, 2000). Although the number and morphology of immature, preovulatory follicles is unchanged between wildtype and *Ahr* knockout females, several reports have noted a marked reduction in the number of mature follicles (Baba et al., 2005; Benedict et al., 2000; Benedict et al., 2003). Evidence indicates that these defects are not due to upstream changes in the endocrine regulation of ovulatory-stimulating hormones (Baba et al., 2005; Trewin et al., 2007).

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Rather, data suggests that maturation of *Ahr* null follicles is disrupted due to insufficient synthesis of estrodiol within the follicle itself (Baba et al., 2005; Barnett et al., 2007). Recently, it has been demonstrated that the AHR acts synergistically with the orphan nuclear receptor Ad4BP (also known as SF-1) to transiently upregulate *Cyp19* (aromatase) levels during preovulation (Baba et al., 2005; Honda et al., 1993). The AHR-dependent proestrus peak in *Cyp19* expression is thought to catalyze estradiol production and downstream signaling in the developing follicle, which has been shown to be critical for both release and implantation (Dupont et al., 2000). Moreover, the role of estradiol in the ovarian defects of AHR null mice has been further strengthened by partial rescue of the null phenotype via exogenous estradiol administration (Baba et al., 2005). Interestingly, the *Cyp19* promoters of several vertebrate species, including multiple fish, humans, and mice contain both AHR and Ad4BP/SF-1 binding sites suggesting an evolutionarily-conserved role for this interaction in ovarian physiology (Baba et al., 2005; Kazeto et al., 2001; Nocillado et al., 2006; Tchoudakova et al., 2001).

Several lines of evidence indicate that the physiological role of the AHR in reproduction extends well beyond oocyte maturation. Fertilization of the oocyte triggers a transient, but robust increase in AHR activity as evidenced by a 100-fold increase in *Cyp1a1* (Dey and Nebert, 1998; Pocar et al., 2004). No physiological role for this phenomenon has yet been demonstrated, although a further relationship with estradiol metabolism would not be unlikely. Post-implantation, the AHR is strongly expressed in the uterine vasculature and developing tissues between the embryo and dam (Kitajima et al., 2004). The absence of AHR signaling in these tissues has been shown to result in an enlarged placental labyrinth with altered dam-to-pup filtration (Thomae et al., 2004 and TL Thomae, personal communication). After birth, pups

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raised by null dams have a significantly lower survival rate than those raised by heterozygotes (Abbott et al., 1999), an observation that may be related to impaired development of the mammary gland (Hushka et al., 1998). Overall, these results indicate that *Ahr* deficiency is deleterious to numerous aspects of reproduction ranging from oocyte maturation to placental development to pup rearing.

Role of endogenous ligand(s) in AHR function

The putative connection between endogenous ligand(s) and the various AHR null phenotypes is a defining question for future study. In mammals, several lines of evidence suggest that intrinsically-produced ligands are an integral part of endogenous AHR biology. First, mutant constructs of AHR and ARNT have demonstrated that endogenous signaling is mechanistically analogous to induction via xenobiotic ligand (Bunger et al., 2003; Walisser et al., 2004a; Walisser et al., 2004b). Most notably, experiments using AHR hypomorphs have shown that physiological deficiency, as measured by ductus venosus closure, can be rescued via high-affinity xenobiotic ligand (Walisser et al., 2004b). Second, cell culture experiments have demonstrated that AHR activity is markedly increased in the absence of CYP1 metabolism, suggesting that the AHR-regulated *Cyp1* battery actively clears an AHR ligand of either cell or sera origin (Chang and Puga, 1998; Levine-Fridman et al., 2004). Third, several classes of endogenous compounds have been demonstrated to directly modulate AHR activity, including (i) tryptophan metabolites (Bittinger et al., 2003; Diani-Moore et al., 2006; Heath-Pagliuso et al., 1998; Wei et al., 1998; Wei et al., 1999) and other indole-containing structures (Adachi et al., 2001; Guengerich et al., 2004; Henry et al., 2006; Miller, 1997; Song et al., 2002), (ii)

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tetrapyroles such as bilirubin and biliverdin (Phelan et al., 1998; Sinal and Bend, 1997), (iii) sterols such as 7-ketocholesterol and equilenin (Jinno et al., 2005; Savouret et al., 2001), (iv) fatty acid metabolites including several prostaglandins and lipoxin A4 (Schaldach et al., 1999; Seidel et al., 2001), and (v) the ubiquitous second messenger cAMP (Oesch-Bartlomowicz et al., 2005) (AHR ligand classes are reviewed in Denison and Nagy, 2003). The physiological relevance of these compounds to the abnormalities of the *Ahr* null mouse, however, is unclear.

Due to the relationship between AHR gene targets and adaptive metabolism of the receptor's agonists, a causative link between reduced ligand metabolism and the null phenotype cannot be overlooked. Potentially, accumulation of a parent compound and/or deficiency of a metabolite could result in the observed null phenotypes. In this context, it is interesting to note that the *Ahr* null mouse contains increased retinoid levels within the liver, which have been linked to the portal fibrosis phenotype, as well as an increased concentration of AHR-activating serum LDL (Andreola et al., 2004; Andreola et al., 1997; McMillan and Bradfield, 2007). However, it remains possible that endogenous agonists do not directly influence the null phenotype, but instead determine spatio-temporal expression of downstream AHR gene targets that are required for normal physiology but unrelated to ligand metabolism.

A connection between endogenous ligand and AHR function in invertebrate species is considerably more tenuous. Several studies have shown that invertebrate othologs of the AHR, including those in the nematode, fruit fly, zebra and blue mussels, and the soft-shell clam do not bind prototypical xenobiotic ligands; possible interaction with other ligand classes, however, has not been examined (Butler et al., 2001; Duncan et al., 1998; Hahn, 2002; Powell-Coffman et al., 1998; Wiesner L, 2001). Studies suggest that post-translational modification of the AHR-1 PAS

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domain, perhaps via ligand binding, could be required for transcriptional activity; modification of Spineless, however, does not appear to be required. Although it is conceivable that one subset of invertebrate AHR orthologs possesses ligand-binding capacity while another does not, this scenario would likely require an independent gain or loss of ligand function prior to vertebrate development. Given that the AHR is unique amongst all PAS superfamily members in its ligand inducibility, this possibility does not seem likely (Gu et al., 2000). Although no conclusion can yet be definitively drawn regarding the role of ligand in invertebrate AHR biology, it should be noted that the body of evidence that supports ligand-independent signaling in *Drosophila* is more substantial than the converse proposition in *C. elegans*. Biochemical studies using additional invertebrate homologs may be key in elucidating the molecular mechanisms underlying ancestral AHR signaling.

Comparative perspective and concluding remarks

What can we learn from the juxtaposition of AHR function in these diverse model systems? Thus far, the most compelling similarities are present in the *Ahr* deficient phenotypes of *C. elegans* and *Drosophila*. The cells/tissues affected by AHR loss of function in these organisms are, with very few exceptions, intimately related to environmental sensation. Spineless is required for normal development of the antenna and maxillary palp (the two olfactory organs in the fly), the mechosensory bristle cells, the eye mosaic required for color vision, and the PNS, which relays multiple sensory inputs. A similar role for AHR-1 in *C. elegans* sensation is emerging from studies into nematode feeding behavior and sensory neuron development. In both model systems, AHR appears to function as a diversification mechanism--often as a binary

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switch--whether directing development from an appendage ground state to the antenna, creating a photoreceptor mosaic, generating two RME neuron subtypes, or diversifying PNS morphogenesis.

The link between invertebrate and murine AHR biology is less clear. Although the liver and kidney, two prominent sites of AHR-mediated vascular diversification, can certainly be viewed as chemosensory organs, such a statement cannot be made regarding the oocyte or the various other reproductive tissues that require endogenous AHR function. Nonetheless, a more comprehensive picture of AHR physiology in the mouse may reveal more overt connections between vertebrate and invertebrate function. Two of the regions which most highly express *Ahr* in the mouse are the vomeronasal olfactory organ and the retina (Jain et al., 1998; Su et al., 2002), an observation which bears a marked similarity to *spineless* expression in the fly (Duncan et al., 1998). However, no studies have yet been conducted into AHR function in these tissues. Moreover, the only studies into AHR function in regards to vertebrate neuronal development, a principle theme in invertebrate biology, have been conducted in the framework of xenobiotic toxicity (Carvalho and Tillitt, 2004; Hays et al., 2002; Hill et al., 2003b; Hood et al., 2006; Petersen et al., 2006; Powers et al., 2005; Williamson et al., 2005).

In addition to loss of function phenotype, another prominent difference between AHR signaling in vertebrate and invertebrate biology appears to be the addition of direct sensory function to its original role in sensory organ development. The vertebrate AHR signaling pathway has clearly developed into an inducible system that mediates the adaptive clearance of a variety of endogenous and exogenous compounds. How could such adaptation occur? Insight may be provided by the recent observation that Spineless regulates basal expression of the insect

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P450 enzyme CYP6B1, which mediates metabolism of a variety of phytochemicals, via conserved DRE promoter elements (Brown et al., 2005; Hung et al., 1996; Li et al., 2002; McDonnell et al., 2004; Petersen et al., 2003). Substrates of the insect CYP6B family include several dietary agonists of the vertebrate AHR such as furocoumarins (Baumgart et al., 2005), flavones (Ciolino et al., 1999; Reiners Jr et al., 1999), and indole-3-carbinol (Bjeldanes et al., 1991). The addition of ligand inducibility (or expansion of the receptor's ligand repertoire) to AHR function, therefore, could have provided significant advantage over plant defenses. Although such an evolutionary mechanism for the alteration/expansion of AHR function during early vertebrate development bears a certain level of allure, this idea is currently speculative.

The evolutionary chasm between invertebrates and higher mammals may be the most difficult obstacle in comparing murine AHR signaling to that seen in the fly or nematode. Less complex vertebrates such as the zebrafish (*Danio rerio*), however, may better illustrate the similarities, as well as differences, between the pre and post-chordate AHR. Although zebrafish molecular techniques still lag behind the more established model organisms, reverse genetic studies are becoming increasingly more tractable in this organism (Lekven et al., 2000; Nasevicius and Ekker, 2000; Wienholds et al., 2002; Wienholds et al., 2003). Xenobiotic studies have already established that AHR biology has a dramatic impact on the zebrafish cardiovascular system, neurodevelopment, and oogenesis, although the role of AHR signaling in normal physiology is not yet known (reviewed in Carney et al., 2006b, also Andreasen et al., 2002; Antkiewicz et al., 2005; Antkiewicz et al., 2006; Bello et al., 2004; Carney et al., 2006a; Handley-Goldstone et al., 2005; Heiden et al., 2006; Henry et al., 1997; Hill et al., 2003a;

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Tanguay et al., 1999). The identification of *Ahr* deficient phenotypes in the zebrafish is undoubtedly on the horizon.

Highly diverse functions for the AHR have already been established in our most powerful model systems and more surprises are likely in store--in these organisms as well as others. Many of the observations detailed herein were published within the last year suggesting that we are only now beginning to elucidate the true physiological functions of the protein we know best as the aryl hydrocarbon receptor. With few exceptions, AHR signaling is still a story of disconnected parts: isolated phenotypes and unexplained toxicology. In the coming years, our understanding of AHR biology will inevitably improve. In doing so, these disparate elements of AHR signaling may yet form a cohesive whole.

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Molecular Pharmacology Fast Forward. Published on May 29, 2007 as DOI: 10.1124/mol.107.037259
This article has not been copyedited and formatted. The final version may differ from this version.

References:

Abbott BD, Birnbaum LS and Perdew GH (1995) Developmental expression of two members of a new class of transcription factors: I. Expression of aryl hydrocarbon receptor in the C57BL/6N mouse embryo. *Dev Dyn* **204**:133–143.

Abbott BD, Schmid JE, Pitt JA, Buckalew AR, Wood CR, Held GA and Diliberto JJ (1999) Adverse Reproductive Outcomes in the Transgenic Ah Receptor-Deficient Mouse. *Toxicology and Applied Pharmacology* **155**(1):62-70.

Adachi-Yamada T, Harumoto T, Sakurai K, Ueda R, Saigo K, O'Connor MB and Nakato H (2005) Wing-to-Leg homeosis by spineless causes apoptosis regulated by Fish-lips, a novel leucine-rich repeat transmembrane protein. *Mol Cell Biol* **25**(8):3140-3150.

Adachi J, Mori Y, Matsui S, Takigami H, Fujino J, Kitagawa H, Miller CA, Kato T, Saeki K and Matsuda T (2001) Indirubin and Indigo Are Potent Aryl Hydrocarbon Receptor Ligands Present in Human Urine. *J Biol Chem* **276**(34):31475-31478.

Ainsley JA, Pettus JM, Bosenko D, Gerstein CE, Zinkevich N, Anderson MG, Adams CM, Welsh MJ and Johnson WA (2003) Enhanced locomotion caused by loss of the Drosophila DEG/ENaC protein Pickpocket1. *Curr Biol* **13**(17):1557-1563.

Alexander-Katz A, Schneider MF, Schneider SW, Wixforth A and Netz RR (2006) Shear-flow-induced unfolding of polymeric globules. *Phys Rev Lett* **97**(13):138101.

Andreasen EA, Hahn ME, Heideman W, Peterson RE and Tanguay RL (2002) The zebrafish (Danio rerio) aryl hydrocarbon receptor type 1 is a novel vertebrate receptor. *Mol Pharmacol* **62**(2):234-249.

Andreola F, Calvisi DF, Elizondo G, Jakowlew SB, Mariano J, Gonzalez FJ and De Luca LM (2004) Reversal of liver fibrosis in aryl hydrocarbon receptornull mice by dietary vitamin A depletion. *Hepatology* **39**(1):157-166.

Andreola F, Fernandez-Salguero PM, Chiantore MV, Petkovich MP, Gonzalez FJ and De Luca LM (1997) Aryl hydrocarbon receptor knockout mice (AHR-/-) exhibit liver retinoid accumulation and reduced retinoic acid metabolism. *Cancer Res* **57**(14):2835-2838.

Antkiewicz DS, Burns CG, Carney SA, Peterson RE and Heideman W (2005) Heart malformation is an early response to TCDD in embryonic zebrafish. *Toxicol Sci* **84**(2):368-377.

Antkiewicz DS, Peterson RE and Heideman W (2006) Blocking expression of AHR2 and ARNT1 in zebrafish larvae protects against cardiac toxicity of 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Toxicol Sci* **94**(1):175-182.

Baba T, Mimura J, Nakamura N, Harada N, Yamamoto M, Morohashi K and Fujii-Kuriyama Y (2005) Intrinsic Function of the Aryl Hydrocarbon (Dioxin) Receptor as a Key Factor in Female Reproduction. *Molecular and Cellular Biology* **25**(22):10040-10051.

Bacsi SG and Hankinson O (1996) Functional Characterization of DNA-binding Domains of the Subunits of the Heterodimeric Aryl Hydrocarbon Receptor Complex Imputing Novel and Canonical Basic Helix-Loop-Helix Protein-DNA Interactions. *Journal of Biological Chemistry* **271**(15):8843.

Barnett KR, Tomic D, Gupta RK, Miller KP, Meachum S, Paulose T and Flaws JA (2007) The Aryl Hydrocarbon Receptor Affects Mouse Ovarian Follicle Growth Via Mechanisms Involving Estradiol Regulation and Responsiveness. *Biol Reprod*.

Downloaded from molpharm.aspetjournals.org at ASPET Journals on April 17, 2024

Molecular Pharmacology Fast Forward. Published on May 29, 2007 as DOI: 10.1124/mol.107.037259
This article has not been copyedited and formatted. The final version may differ from this version.

Barouki R, Coumoul X and Fernandez-Salguero PM (2007) The aryl hydrocarbon receptor, more than a xenobiotic-interacting protein. *FEBS Lett*.

Baumgart A, Schmidt M, Schmitz HJ and Schrenk D (2005) Natural furocoumarins as inducers and inhibitors of cytochrome P450 1A1 in rat hepatocytes. *Biochem Pharmacol* **69**(4):657-667.

Bello SM, Heideman W and Peterson RE (2004) 2,3,7,8-Tetrachlorodibenzo-p-dioxin inhibits regression of the common cardinal vein in developing zebrafish. *Toxicol Sci* **78**(2):258-266.

Benedict JC, Lin TM, Loeffler IK, Peterson RE and Flaws JA (2000) Physiological Role of the Aryl Hydrocarbon Receptor in Mouse Ovary Development. *Toxicological Sciences* **56**(2):382.

Benedict JC, Miller KP, Lin TM, Greenfeld C, Babus JK, Peterson RE and Flaws JA (2003) Aryl Hydrocarbon Receptor Regulates Growth, But Not Atresia, of Mouse Preantral and Antral Follicles. *Biology of Reproduction* **68**(5):1511-1517.

Birnbaum LS and Perdew GH (1995) Developmental expression of two members of a new class of transcription factors: I. Expression of aryl hydrocarbon receptor in the C57BL/6N mouse embryo. *Dev Dyn* **204**:133–143.

Birnbaum LS and Tuomisto J (2000) Non-carcinogenic effects of TCDD in animals. *Food Additives and Contaminants* **17**(4):275-288.

Bittinger MA, Nguyen LP and Bradfield CA (2003) Aspartate Aminotransferase Generates Proagonists of the Aryl Hydrocarbon Receptor. *Mol Pharmacol* **64**(3):550-556.

Bjeldanes LF, Kim J, Grose KR, Bartholomew JC and Bradfield CA (1991) Aromatic Hydrocarbon Responsiveness-Receptor Agonists Generated from Indole-3-Carbinol in vitro and in vivo: Comparisons with 2, 3, 7, 8-Tetrachlorodibenzo-p-Dioxin. *Proceedings of the National Academy of Sciences* **88**(21):9543-9547.

Bodmer RaJ, Yuh Nung (1987) Morphological differentiation of the embryonic peripheral neurons in Drosophila. *Development Genes and Evolution* **196**(2):69-77.

Border WA and Noble NA (1994) Transforming Growth Factor ß in Tissue Fibrosis. *New England Journal of Medicine* **331**(19):1286-1292.

Branton MH and Kopp JB (1999) TGF-beta and fibrosis. *Microbes Infect* **1**(15):1349-1365.

Brenman JE, Gao FB, Jan LY and Jan YN (2001) Sequoia, a tramtrack-related zinc finger protein, functions as a pan-neural regulator for dendrite and axon morphogenesis in Drosophila. *Dev Cell* **1**(5):667-677.

Bridges CaM, T.H. (1923) The third-chromosome group of mutant characters of *Drosophila melanogaster*. *Carnegie Institute of Washington, Washington DC Publ No 327*.

Brown RP, McDonnell CM, Berenbaum MR and Schuler MA (2005) Regulation of an insect cytochrome P450 monooxygenase gene (CYP6B1) by aryl hydrocarbon and xanthotoxin response cascades. *Gene* **358**:39-52.

Bunger MK, Moran SM, Glover E, Thomae TL, Lahvis GP, Lin BC and Bradfield CA (2003) Resistance to 2, 3, 7, 8-Tetrachlorodibenzo-p-dioxin Toxicity and Abnormal Liver Development in Mice Carrying a Mutation in the Nuclear Localization Sequence of the Aryl Hydrocarbon Receptor. *J Biol Chem* **278**(20):17767-17774.

Burgess EA and Duncan I (1990) Direct control of antennal identity by the spineless-aristapedia gene of Drosophila. *Mol Gen Genet* **221**(3):347-357.

Downloaded from molpharm.aspetjournals.org at ASPET Journals on April 17, 2024

Molecular Pharmacology Fast Forward. Published on May 29, 2007 as DOI: 10.1124/mol.107.037259
This article has not been copyedited and formatted. The final version may differ from this version.

Butler RA, Kelley ML, Powell WH, Hahn ME and Van Beneden RJ (2001) An aryl hydrocarbon receptor (AHR) homologue from the soft-shell clam, Mya arenaria: evidence that invertebrate AHR homologues lack 2,3,7,8-tetrachlorodibenzo-p-dioxin and beta-naphthoflavone binding. *Gene* **278**(1-2):223-234.

Carney SA, Chen J, Burns CG, Xiong KM, Peterson RE and Heideman W (2006a) Aryl hydrocarbon receptor activation produces heart-specific transcriptional and toxic responses in developing zebrafish. *Mol Pharmacol* **70**(2):549-561.

Carney SA, Prasch AL, Heideman W and Peterson RE (2006b) Understanding dioxin developmental toxicity using the zebrafish model. *Birth Defects Res A Clin Mol Teratol* **76**(1):7-18.

Carvalho PS and Tillitt DE (2004) 2,3,7,8-TCDD effects on visual structure and function in swim-up rainbow trout. *Environ Sci Technol* **38**(23):6300-6306.

Chang CY and Puga A (1998) Constitutive activation of the aromatic hydrocarbon receptor. *Mol Cell Biol* **18**(1):525-535.

Cheung BH, Arellano-Carbajal F, Rybicki I and de Bono M (2004) Soluble guanylate cyclases act in neurons exposed to the body fluid to promote C. elegans aggregation behavior. *Curr Biol* **14**(12):1105-1111.

Chou WH, Hall KJ, Wilson DB, Wideman CL, Townson SM, Chadwell LV and Britt SG (1996) Identification of a novel Drosophila opsin reveals specific patterning of the R7 and R8 photoreceptor cells. *Neuron* **17**(6):1101-1115.

Chou WH, Huber A, Bentrop J, Schulz S, Schwab K, Chadwell LV, Paulsen R and Britt SG (1999) Patterning of the R7 and R8 photoreceptor cells of Drosophila: evidence for induced and default cell-fate specification. *Development* **126**(4):607-616.

Chu J, Dong PDS and Panganiban G (2002) Limb type-specific regulation of bric a brac contributes to morphological diversity. *Development* **129**(3):695-704.

Ciolino HP, Daschner PJ and Yeh GC (1999) Dietary flavonols quercetin and kaempferol are ligands of the aryl hydrocarbon receptor that affect CYP1A1 transcription differentially. *Biochem J* **340**(Pt 3):715-722.

Coates JC and de Bono M (2002) Antagonistic pathways in neurons exposed to body fluid regulate social feeding in Caenorhabditis elegans. *Nature* **419**(6910):925-929.

Corchero J, Martin-Partido G, Dallas SL and Fernandez-Salguero PM (2004) Liver portal fibrosis in dioxin receptor-null mice that overexpress the latent transforming growth factor-beta-binding protein-1. *International Journal of Experimental Pathology* **85**(5):295-302.

de Bono M and Bargmann CI (1998) Natural variation in a neuropeptide Y receptor homolog modifies social behavior and food response in C. elegans. *Cell* **94**(5):679-689.

Denison MS and Nagy SR (2003) Activation of the aryl hydrocarbon receptor by structurally diverse exogenous and endogenous chemicals. *Annu Rev Pharmacol Toxicol* **43**:309-334.

Dent JA, Davis MW and Avery L (1997) avr-15 encodes a chloride channel subunit that mediates inhibitory glutamatergic neurotransmission and ivermectin sensitivity in Caenorhabditis elegans. *The EMBO Journal* **16**:5867-5879.

Dey A and Nebert DW (1998) Markedly increased constitutive CYP1A1 mRNA levels in the fertilized ovum of the mouse. *Biochem Biophys Res Commun* **251**(2):657-661.

Downloaded from molpharm.aspetjournals.org at ASPET Journals on April 17, 2024

Molecular Pharmacology Fast Forward. Published on May 29, 2007 as DOI: 10.1124/mol.107.037259
This article has not been copyedited and formatted. The final version may differ from this version.

Diani-Moore S, Labitzke E, Brown R, Garvin A, Wong L and Rifkind AB (2006) Sunlight Generates Multiple Tryptophan Photoproducts Eliciting High Efficacy CYP1A Induction in Chick Hepatocytes and In Vivo. *Toxicological Sciences* **90**(1):96.

Donald L. Riddle TB, Barbara J. Meyer, and James R. Priess. (1997) *C. Elegans II*. Cold Spring Harbor.

Duncan DM, Burgess EA and Duncan I (1998) Control of distal antennal identity and tarsal development in Drosophila by spineless-aristapedia, a homolog of the mammalian dioxin receptor. *Genes Dev* **12**(9):1290-1303.

Dupont S, Krust A, Gansmuller A, Dierich A, Chambon P and Mark M (2000) Effect of single and compound knockouts of estrogen receptors alpha (ERalpha) and beta (ERbeta) on mouse reproductive phenotypes. **127**(19):4277-4291.

Edelstone DI, Rudolph AM and Heymann MA (1978) Liver and ductus venosus blood flows in fetal lambs in utero. *Circulation Research* **42**(3):426-433.

Emerald BS, Curtiss J, Mlodzik M and Cohen SM (2003) distal antenna and distal antenna related encode nuclear proteins containing pipsqueak motifs involved in antenna development in Drosophila. *Development* **130**(6):1171-1180.

Emmons RB, Duncan D and Duncan I (2006) Regulation of the Drosophila distal antennal determinant spineless. *Dev Biol* **302**(2):412-423.

Emmons RB, Duncan D, Estes PA, Kiefel P, Mosher JT, Sonnenfeld M, Ward MP, Duncan I and Crews ST (1999) The spineless-aristapedia and tango bHLH-PAS proteins interact to control antennal and tarsal development in Drosophila. *Development* **126**(17):3937-3945.

Feiler R, Bjornson R, Kirschfeld K, Mismer D, Rubin GM, Smith DP, Socolich M and Zuker CS (1992) Ectopic expression of ultraviolet-rhodopsins in the blue photoreceptor cells of Drosophila: visual physiology and photochemistry of transgenic animals. *J Neurosci* **12**(10):3862-3868.

Fernandez-Salguero P, Pineau T, Hilbert DM, McPhail T, Lee SS, Kimura S, Nebert DW, Rudikoff S, Ward JM and Gonzalez FJ (1995) Immune system impairment and hepatic fibrosis in mice lacking the dioxin-binding Ah receptor. *Science* **268**(5211):722.

Fernandez-Salguero PM, Ward JM, Sundberg JP and Gonzalez FJ (1997) Lesions of aryl-hydrocarbon receptor-deficient mice. *Veterinary Pathology Online* **34**(6):605-614.

Fryxell KJ and Meyerowitz EM (1987) An opsin gene that is expressed only in the R7 photoreceptor cell of Drosophila. *Embo J* **6**(2):443-451.

Gilbert SF (2000) *Developmental biology*. Sinauer Associates, Sunderland, Mass.

Gomez-Duran A, Mulero-Navarro S, Chang X and Fernandez-Salguero PM (2005) LTBP-1 blockade in dioxin receptor-null mouse embryo fibroblasts decreases TGF-beta activity: Role of extracellular proteases plasmin and elastase. *J Cell Biochem* **97**(2):380-392.

Gray JM, Karow DS, Lu H, Chang AJ, Chang JS, Ellis RE, Marletta MA and Bargmann CI (2004) Oxygen sensation and social feeding mediated by a C. elegans guanylate cyclase homologue. *Nature* **430**(6997):317-322.

Grueber WB, Jan LY and Jan YN (2002) Tiling of the Drosophila epidermis by multidendritic sensory neurons. *Development* **129**(12):2867-2878.

Grueber WB, Jan LY and Jan YN (2003) Different levels of the homeodomain protein cut regulate distinct dendrite branching patterns of Drosophila multidendritic neurons. *Cell* **112**(6):805-818.

Downloaded from molpharm.aspetjournals.org at ASPET Journals on April 17, 2024

Molecular Pharmacology Fast Forward. Published on May 29, 2007 as DOI: 10.1124/mol.107.037259
This article has not been copyedited and formatted. The final version may differ from this version.

Gu YZ, Hogenesch JB and Bradfield CA (2000) The PAS Superfamily: Sensors of Environmental and Developmental Signals. *Annual Review of Pharmacology and Toxicology* **40**(1):519-561.

Guengerich FP, Martin MV, McCormick WA, Nguyen LP, Glover E and Bradfield CA (2004) Aryl hydrocarbon receptor response to indigoids in vitro and in vivo. *Archives of biochemistry and biophysics(Print)* **423**(2):309-316.

Guo J, Sartor M, Karyala S, Medvedovic M, Kann S, Puga A, Ryan P and Tomlinson CR (2004) Expression of genes in the TGF-beta signaling pathway is significantly deregulated in smooth muscle cells from aorta of aryl hydrocarbon receptor knockout mice. *Toxicol Appl Pharmacol* **194**(1):79-89.

Hahn ME (2002) Aryl hydrocarbon receptors: diversity and evolution. *Chem Biol Interact* **141**(1-2):131-160.

Hahn ME, Karchner SI, Evans BR, Franks DG, Merson RR and Lapseritis JM (2006) Unexpected diversity of aryl hydrocarbon receptors in non-mammalian vertebrates: insights from comparative genomics. *J Exp Zoolog A Comp Exp Biol* **305**(9):693-706.

Handley-Goldstone HM, Grow MW and Stegeman JJ (2005) Cardiovascular gene expression profiles of dioxin exposure in zebrafish embryos. *Toxicol Sci* **85**(1):683-693.

Harstad EB, Guite CA, Thomae TL and Bradfield CA (2006) Liver deformation in Ahr-null mice: evidence for aberrant hepatic perfusion in early development. *Mol Pharmacol* **69**(5):1534-1541.

Hart AC, Sims S and Kaplan JM (1995) Synaptic code for sensory modalities revealed by C. elegans GLR-1 glutamate receptor. *Nature* **378**(6552):82-85.

Hays LE, Carpenter CD and Petersen SL (2002) Evidence That GABAergic Neurons in the Preoptic Area of the Rat Brain Are Targets of 2, 3, 7, 8-Tetrachlorodibenzo-p-Dioxin during Development. *Environmental Health Perspectives* **110**:369-376.

Heath-Pagliuso S, Rogers WJ, Tullis K, Seidel SD, Cenijn PH, Brouwer A and Denison MS (1998) Activation of the Ah receptor by tryptophan and tryptophan metabolites. *Biochemistry* **37**(11):508–511.

Heiden TK, Carvan MJ, 3rd and Hutz RJ (2006) Inhibition of follicular development, vitellogenesis, and serum 17beta-estradiol concentrations in zebrafish following chronic, sublethal dietary exposure to 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Toxicol Sci* **90**(2):490-499.

Henry EC, Bemis JC, Henry O, Kende AS and Gasiewicz TA (2006) A potential endogenous ligand for the aryl hydrocarbon receptor has potent agonist activity in vitro and in vivo. *Arch Biochem Biophys* **450**(1):67-77.

Henry EC and Gasiewicz TA (2003) Agonist but Not Antagonist Ligands Induce Conformational Change in the Mouse Aryl Hydrocarbon Receptor as Detected by Partial Proteolysis. *Molecular Pharmacology* **63**(2):392-400.

Henry TR, Spitsbergen JM, Hornung MW, Abnet CC and Peterson RE (1997) Early life stage toxicity of 2,3,7,8-tetrachlorodibenzo-p-dioxin in zebrafish (Danio rerio). *Toxicol Appl Pharmacol* **142**(1):56-68.

Hill A, Howard CV, Strahle U and Cossins A (2003a) Neurodevelopmental defects in zebrafish (Danio rerio) at environmentally relevant dioxin (TCDD) concentrations. *Toxicol Sci* **76**(2):392-399.

Downloaded from molpharm.aspetjournals.org at ASPET Journals on April 17, 2024

Molecular Pharmacology Fast Forward. Published on May 29, 2007 as DOI: 10.1124/mol.107.037259
This article has not been copyedited and formatted. The final version may differ from this version.

Hill A, Howard CV, Strahle U and Cossins A (2003b) Neurodevelopmental Defects in Zebrafish (Danio rerio) at Environmentally Relevant Dioxin (TCDD) Concentrations. *Toxicological Sciences* **76**(2):392.

Honda S, Morohashi K, Nomura M, Takeya H, Kitajima M and Omura T (1993) Ad4BP regulating steroidogenic P-450 gene is a member of steroid hormone receptor superfamily. *Journal of Biological Chemistry* **268**(10):7494-7502.

Hood DB, Woods L, Brown L, Johnson S and Ebner FF (2006) Gestational 2,3,7,8-tetrachlorodibenzo-p-dioxin exposure effects on sensory cortex function. *Neurotoxicology* **27**(6):1032-1042.

Huang X, Powell-Coffman JA and Jin Y (2004) The AHR-1 aryl hydrocarbon receptor and its co-factor the AHA-1 aryl hydrocarbon receptor nuclear translocator specify GABAergic neuron cell fate in C. elegans. *Development* **131**(4):819-828.

Huber A, Schulz S, Bentrop J, Groell C, Wolfrum U and Paulsen R (1997) Molecular cloning of Drosophila Rh6 rhodopsin: the visual pigment of a subset of R8 photoreceptor cells. *FEBS Lett* **406**(1-2):6-10.

Hung CF, Holzmacher R, Connolly E, Berenbaum MR and Schuler MA (1996) Conserved promoter elements in the CYP6B gene family suggest common ancestry for cytochrome P450 monooxygenases mediating furanocoumarin detoxification. *Proc Natl Acad Sci U S A* **93**(22):12200-12205.

Hushka LJ, Williams JS and Greenlee WF (1998) Characterization of 2, 3, 7, 8-Tetrachlorodibenzofuran-Dependent Suppression and AH Receptor Pathway Gene Expression in the Developing Mouse Mammary Gland. *Toxicology and Applied Pharmacology* **152**(1):200-210.

Ikuta T, Eguchi H, Tachibana T, Yoneda Y and Kawajiri K (1998) Nuclear Localization and Export Signals of the Human Aryl Hydrocarbon Receptor. *Journal of Biological Chemistry* **273**(5):2895-2904.

Jain S, Maltepe E, Lu MM, Simon C and Bradfield CA (1998) Expression of ARNT, ARNT2, HIF1, HIF2 and Ah receptor mRNAs in the developing mouse. *Mech Dev* **73**:117-123.

Jiang H, Guo R and Powell-Coffman JA (2001) The Caenorhabditis elegans hif-1 gene encodes a bHLH-PAS protein that is required for adaptation to hypoxia. *Proc Natl Acad Sci U S A* **98**(14):7916-7921.

Jinno A, Maruyama Y, Ishizuka M, Kazusaka A, Nakamura A and Fujita S (2005) Induction of cytochrome P450-1A by the equine estrogen equilenin, a new endogenous aryl hydrocarbon receptor ligand. *J Steroid Biochem Mol Biol* **98**(1):48-55.

Kazeto Y, Ijiri S, Place AR, Zohar Y and Trant JM (2001) The 5'-flanking regions of CYP19A1 and CYP19A2 in zebrafish. *Biochem Biophys Res Commun* **288**(3):503-508.

Kim MD, Jan LY and Jan YN (2006) The bHLH-PAS protein Spineless is necessary for the diversification of dendrite morphology of Drosophila dendritic arborization neurons. *Genes Dev* **20**(20):2806-2819.

Kiserud T, Rasmussen S and Skulstad S (2000) Blood flow and the degree of shunting through the ductus venosus in the human fetus. *Am J Obstet Gynecol* **182**:1 Pt 1-147-153.

Kitajima M, Khan KN, Fujishita A, Masuzaki H, Koji T and Ishimaru T (2004) Expression of the arylhydrocarbon receptor in the peri-implantation period of the mouse uterus and the impact of dioxin on mouse implantation. *Arch Histol Cytol* **67**(5):465-474.

Downloaded from molpharm.aspetjournals.org at ASPET Journals on April 17, 2024

Molecular Pharmacology Fast Forward. Published on May 29, 2007 as DOI: 10.1124/mol.107.037259
This article has not been copyedited and formatted. The final version may differ from this version.

Kojima S and Rifkin DB (1993) Mechanism of retinoid-induced activation of latent transforming growth factor-beta in bovine endothelial cells. *J Cell Physiol* **155**(2):323-332.

Kozu S, Tajiri R, Tsuji T, Michiue T, Saigo K and Kojima T (2006) Temporal regulation of late expression of Bar homeobox genes during Drosophila leg development by Spineless, a homolog of the mammalian dioxin receptor. *Dev Biol* **294**(2):497-508.

Lahvis GP and Bradfield CA (1998) Ahr null alleles: distinctive or different? *Biochem Pharmacol* **56**(7):781-787.

Lahvis GP, Lindell SL, Thomas RS, McCuskey RS, Murphy C, Glover E, Bentz M, Southard J and Bradfield CA (2000) Portosystemic shunting and persistent fetal vascular structures in aryl hydrocarbon receptor-deficient mice. *Proc Natl Acad Sci U S A* **97**(19):10442-10447.

Lahvis GP, Pyzalski RW, Glover E, Pitot HC, McElwee MK and Bradfield CA (2005) The aryl hydrocarbon receptor is required for developmental closure of the ductus venosus in the neonatal mouse. *Mol Pharmacol* **67**(3):714-720.

Lekven AC, Helde KA, Thorpe CJ, Rooke R and Moon RT (2000) Reverse genetics in zebrafish. *Physiol Genomics* **2**(2):37-48.

Levine-Fridman A, Chen L and Elferink CJ (2004) Cytochrome P4501A1 Promotes G1 Phase Cell Cycle Progression by Controlling Aryl Hydrocarbon Receptor Activity. *Molecular Pharmacology* **65**(2):461-469.

Li W, Petersen RA, Schuler MA and Berenbaum MR (2002) CYP6B cytochrome p450 monooxygenases from Papilio canadensis and Papilio glaucus: potential contributions of sequence divergence to host plant associations. *Insect Mol Biol* **11**(6):543-551.

Li W, Wang F, Menut L and Gao FB (2004) BTB/POZ-zinc finger protein abrupt suppresses dendritic branching in a neuronal subtype-specific and dosage-dependent manner. *Neuron* **43**(6):823-834.

Lindsley DL and Zimm GG (1992) *The genome of Drosophila melanogaster*. Academic Press San Diego, CA.

Liu L, Yermolaieva O, Johnson WA, Abboud FM and Welsh MJ (2003) Identification and function of thermosensory neurons in Drosophila larvae. *Nat Neurosci* **6**(3):267-273.

Lund AK, Goens MB, Kanagy NL and Walker MK (2003) Cardiac hypertrophy in aryl hydrocarbon receptor null mice is correlated with elevated angiotensin II, endothelin-1, and mean arterial blood pressure. *Toxicol Appl Pharmacol* **193**(2):177-187.

Lund AK, Goens MB, Nunez BA and Walker MK (2005a) Characterizing the role of endothelin-1 in the progression of cardiac hypertrophy in aryl hydrocarbon receptor (AhR) null mice. *Toxicol Appl Pharmacol* **212**(2):127-135.

Lund AK, Peterson SL, Timmins GS and Walker MK (2005b) Endothelin-1-Mediated Increase in Reactive Oxygen Species and NADPH Oxidase Activity in Hearts of Aryl Hydrocarbon Receptor (AhR) Null Mice. *Toxicological Sciences* **88**(1):265.

McDonnell CM, Brown RP, Berenbaum MR and Schuler MA (2004) Conserved regulatory elements in the promoters of two allelochemical-inducible cytochrome P450 genes differentially regulate transcription. *Insect Biochem Mol Biol* **34**(10):1129-1139.

McIntire SL, Jorgensen E, Kaplan J and Horvitz HR (1993) The GABAergic nervous system of Caenorhabditis elegans. *Nature* **364**(6435):337-341.

Downloaded from molpharm.aspetjournals.org at ASPET Journals on April 17, 2024

Molecular Pharmacology Fast Forward. Published on May 29, 2007 as DOI: 10.1124/mol.107.037259
This article has not been copyedited and formatted. The final version may differ from this version.

McMillan BJ and Bradfield CA (2007) The aryl hydrocarbon receptor is activated by modified low-density lipoprotein. *Proc Natl Acad Sci U S A* **104**(4):1412-1417.

McMillan PA and McGuire TR (1992) The homeotic gene spineless-aristapedia affects geotaxis in Drosophila melanogaster. *Behav Genet* **22**(5):557-573.

Miller CA (1997) Expression of the Human Aryl Hydrocarbon Receptor Complex in Yeast: Activation of Transcription by Indole Compounds. *Journal of Biological Chemistry* **272**(52):32824-32829.

Miller G, Hansen K and Stark W (1981) Phototaxis in *Drosophila*: R1-R6 input and interaction among ocellar and compound eye receptors. *J Insect Physiol* **27**:813-819.

Mimura J, Yamashita K, Nakamura K, Morita M, Takagi TN, Nakao K, Ema M, Sogawa K, Yasuda M and Katsuki M (1997) Loss of teratogenic response to 2, 3, 7, 8-tetrachlorodibenzo-p-dioxin (TCDD) in mice lacking the Ah (dioxin) receptor. *Genes to Cells* **2**(10):645-654.

Mufti NA, Bleckwenn NA, Babish JG and Shuler ML (1995) Possible involvement of the Ah receptor in the induction of cytochrome P-450IA1 under conditions of hydrodynamic shear in microcarrier-attached hepatoma cell lines. *Biochem Biophys Res Commun* **208**(1):144-152.

Murre C, McCaw PS, Vaessin H, Caudy M, Jan LY, Jan YN, Cabrera CV, Buskin JN, Hauschka SD and Lassar AB (1989) Interactions between heterologous helix-loop-helix proteins generate complexes that bind specifically to a common DNA sequence. *Cell* **58**(3):537-544.

Nasevicius A and Ekker SC (2000) Effective targeted gene 'knockdown' in zebrafish. *Nat Genet* **26**(2):216-220.

Nebert DW, Dalton TP, Okey AB and Gonzalez FJ (2004) Role of Aryl Hydrocarbon Receptor-mediated Induction of the CYP1 Enzymes in Environmental Toxicity and Cancer. *Journal of Biological Chemistry* **279**(23):23847-23850.

Nocillado JN, Elizur A, Avitan A, Carrick F and Levavi-Sivan B (2006) Cytochrome P450 aromatase in grey mullet: cDNA and promoter isolation; brain, pituitary and ovarian expression during puberty. *Mol Cell Endocrinol* **263**(1):65-78.

Oesch-Bartlomowicz B, Huelster A, Wiss O, Antoniou-Lipfert P, Dietrich C, Arand M, Weiss C, Bockamp E and Oesch F (2005) Aryl hydrocarbon receptor activation by cAMP vs. dioxin: Divergent signaling pathways. *Proceedings of the National Academy of Sciences* **102**(26):9218-9223.

Okuno M, Moriwaki H, Imai S, Muto Y, Kawada N, Suzuki Y and Kojima S (1997) Retinoids exacerbate rat liver fibrosis by inducing the activation of latent TGF-beta in liver stellate cells. *Hepatology* **26**(4):913-921.

Papatsenko D, Sheng G and Desplan C (1997) A new rhodopsin in R8 photoreceptors of Drosophila: evidence for coordinate expression with Rh3 in R7 cells. *Development* **124**(9):1665-1673.

Parrish JZ, Kim MD, Jan LY and Jan YN (2006) Genome-wide analyses identify transcription factors required for proper morphogenesis of Drosophila sensory neuron dendrites. *Genes Dev* **20**(7):820-835.

Downloaded from molpharm.aspetjournals.org at ASPET Journals on April 17, 2024

Molecular Pharmacology Fast Forward. Published on May 29, 2007 as DOI: 10.1124/mol.107.037259
This article has not been copyedited and formatted. The final version may differ from this version.

Petersen RA, Niamsup H, Berenbaum MR and Schuler MA (2003) Transcriptional response elements in the promoter of CYP6B1, an insect P450 gene regulated by plant chemicals. *Biochim Biophys Acta* **1619**(3):269-282.

Petersen SL, Krishnan S and Hudgens ED (2006) The aryl hydrocarbon receptor pathway and sexual differentiation of neuroendocrine functions. *Endocrinology* **147**(6 Suppl):S33-42.

Petrulis JR, Kusnadi A, Ramadoss P, Hollingshead B and Perdew GH (2003) The hsp90 Co-chaperone XAP2 Alters Importin ß Recognition of the Bipartite Nuclear Localization Signal of the Ah Receptor and Represses Transcriptional Activity. *Journal of Biological Chemistry* **278**(4):2677-2685.

Petrulis JR and Perdew GH (2002) The role of chaperone proteins in the aryl hydrocarbon receptor core complex. *Chem Biol Interact* **141**(1-2):25-40.

Phelan D, Winter GM, Rogers WJ, Lam JC and Denison MS (1998) Activation of the Ah receptor signal transduction pathway by bilirubin and biliverdin. *Arch Biochem Biophys* **357**(1):155-163.

Pocar P, Augustin R and Fischer B (2004) Constitutive Expression of CYP1A1 in Bovine Cumulus Oocyte-Complexes in Vitro: Mechanisms and Biological Implications. *Endocrinology* **145**(4):1594-1601.

Powell-Coffman JA, Bradfield CA and Wood WB (1998) Caenorhabditis elegans orthologs of the aryl hydrocarbon receptor and its heterodimerization partner the aryl hydrocarbon receptor nuclear translocator. *Proc Natl Acad Sci U S A* **95**(6):2844-2849.

Powers BE, Lin TM, Vanka A, Peterson RE, Juraska JM and Schantz SL (2005) Tetrachlorodibenzo-p-dioxin exposure alters radial arm maze performance and hippocampal morphology in female AhR mice. *Genes Brain Behav* **4**(1):51-59.

Prasch AL, Tanguay RL, Mehta V, Heideman W and Peterson RE (2005) Identification of zebrafish ARNT1 homologs: TCDD toxicity in the developing zebrafish requires ARNT1. *Molecular Pharmacology*.

Qin H and Powell-Coffman JA (2004) The Caenorhabditis elegans aryl hydrocarbon receptor, AHR-1, regulates neuronal development. *Dev Biol* **270**(1):64-75.

Qin H, Zhai Z and Powell-Coffman JA (2006) The Caenorhabditis elegans AHR-1 transcription complex controls expression of soluble guanylate cyclase genes in the URX neurons and regulates aggregation behavior. *Dev Biol* **298**(2):606-615.

Reiners Jr JJ, Clift R and Mathieu P (1999) Suppression of cell cycle progression by flavonoids: dependence on the aryl hydrocarbon receptor. *Carcinogenesis* **20**(8):1561-1566.

Resnick N, Yahav H, Shay-Salit A, Shushy M, Schubert S, Zilberman LC and Wofovitz E (2003) Fluid shear stress and the vascular endothelium: for better and for worse. *Prog Biophys Mol Biol* **81**(3):177-199.

Rogers C, Reale V, Kim K, Chatwin H, Li C, Evans P and de Bono M (2003) Inhibition of Caenorhabditis elegans social feeding by FMRFamide-related peptide activation of NPR-1. *Nat Neurosci* **6**(11):1178-1185.

Santiago-Josefat B, Mulero-Navarro S, Dallas SL and Fernandez-Salguero PM (2004) Overexpression of latent transforming growth factor-ß binding protein 1 (LTBP-1) in dioxin receptor-null mouse embryo fibroblasts. *Journal of Cell Science* **117**(6):849-859.

Downloaded from molpharm.aspetjournals.org at ASPET Journals on April 17, 2024

Molecular Pharmacology Fast Forward. Published on May 29, 2007 as DOI: 10.1124/mol.107.037259
This article has not been copyedited and formatted. The final version may differ from this version.

Savouret JF, Antenos M, Quesne M, Xu J, Milgrom E and Casper RF (2001) 7-Ketocholesterol Is an Endogenous Modulator for the Arylhydrocarbon Receptor. *Journal of Biological Chemistry* **276**(5):3054-3059.

Schaldach CM, Riby J and Bjeldanes LF (1999) Lipoxin A4: a new class of ligand for the Ah receptor. *Biochemistry* **38**(23):7594-7600.

Schecter A, Birnbaum L, Ryan JJ and Constable JD (2006) Dioxins: An overview. *Environ Res* **101**(3):419-428.

Schmidt JV, Su GHT, Reddy JK, Simon MC and Bradfield CA (1996) Characterization of a murine Ahr null allele: Involvement of the Ah receptor in hepatic growth and development. *Proc Natl Acad Sci U S A* **93**(13):6731-6736.

Seidel SD, Winters GM, Rogers WJ, Ziccardi MH, Li V, Keser B and Denison MS (2001) Activation of the Ah Receptor Signaling Pathway by Prostaglandins. *Journal of Biochemical and Molecular Toxicology* **15**(4):187-196.

Shankaran H, Alexandridis P and Neelamegham S (2003) Aspects of hydrodynamic shear regulating shear-induced platelet activation and self-association of von Willebrand factor in suspension. *Blood* **101**(7):2637-2645.

Siedlecki CA, Lestini BJ, Kottke-Marchant KK, Eppell SJ, Wilson DL and Marchant RE (1996) Shear-dependent changes in the three-dimensional structure of human von Willebrand factor. *Blood* **88**(8):2939-2950.

Sinal CJ and Bend JR (1997) Aryl Hydrocarbon Receptor-Dependent Induction of Cyp1a1 by Bilirubin in Mouse Hepatoma Hepa 1c1c7 Cells. *Molecular Pharmacology* **52**(4):590-599.

Song J, Clagett-Dame M, Peterson RE, Hahn ME, Westler WM, Sicinski RR and DeLuca HF (2002) A ligand for the aryl hydrocarbon receptor isolated from lung. *Proceedings of the National Academy of Sciences* **99**(23):14694-14699.

Struhl G (1982) Spineless-aristapedia: a homeotic gene that does not control the development of specific compartments in Drosophila. *Genetics* **102**(4):737-749.

Su AI, Cooke MP, Ching KA, Hakak Y, Walker JR, Wiltshire T, Orth AP, Vega RG, Sapinoso LM and Moqrich A (2002) Large-scale analysis of the human and mouse transcriptomes. *Proc Natl Acad Sci U S A* **99**(7):4465-4470.

Sugimura K, Satoh D, Estes P, Crews S and Uemura T (2004) Development of morphological diversity of dendrites in Drosophila by the BTB-zinc finger protein abrupt. *Neuron* **43**(6):809-822.

Suzanne M, Estella C, Calleja M and Sanchez-Herrero E (2003) The hernandez and fernandez genes of Drosophila specify eye and antenna. *Dev Biol* **260**(2):465-483.

Swanson HI and Bradfield CA (1993) The AH-receptor: genetics, structure and function. *Pharmacogenetics* **3**(5):213-230.

Swanson HI, Chan WK and Bradfield CA (1995) DNA Binding Specificities and Pairing Rules of the Ah Receptor, ARNT, and SIM Proteins. *J Biol Chem* **270**(44):26292-26302.

Tanguay RL, Abnet CC, Heideman W and Peterson RE (1999) Cloning and characterization of the zebrafish (Danio rerio) aryl hydrocarbon receptor. *Biochim Biophys Acta* **1444**(1):35-48.

Downloaded from molpharm.aspetjournals.org at ASPET Journals on April 17, 2024

Molecular Pharmacology Fast Forward. Published on May 29, 2007 as DOI: 10.1124/mol.107.037259
This article has not been copyedited and formatted. The final version may differ from this version.

Tchoudakova A, Kishida M, Wood E and Callard GV (2001) Promoter characteristics of two cyp19 genes differentially expressed in the brain and ovary of teleost fish. *J Steroid Biochem Mol Biol* **78**(5):427-439.

Thackaberry EA, Gabaldon DM, Walker MK and Smith SM (2002) Aryl Hydrocarbon Receptor Null Mice Develop Cardiac Hypertrophy and Increased Hypoxia-Inducible Factor-1 alpha in the Absence of Cardiac Hypoxia. *Cardiovascular Toxicology* **2**(4):263-273.

Thomae TL, Glover E and Bradfield CA (2004) A Maternal Ahr Null Genotype Sensitizes Embryos to Chemical Teratogenesis. *Journal of Biological Chemistry* **279**(29):30189-30194.

Thomae TL, Stevens EA and Bradfield CA (2005) Transforming Growth Factor-{beta} 3 Restores Fusion in Palatal Shelves Exposed to 2, 3, 7, 8-Tetrachlorodibenzo-p-dioxin. *J Biol Chem* **280**(13):12742-12746.

Tracey WD, Jr., Wilson RI, Laurent G and Benzer S (2003) painless, a Drosophila gene essential for nociception. *Cell* **113**(2):261-273.

Trewin AL, Woller MJ, Wimpee BA, Conley LK, Baldridge MG and Hutz RJ (2007) Short-Term Hormone Release from Adult Female Rat Hypothalamic and Pituitary Explants is not Altered by 2,3,7,8-Tetrachlorodibenzo-p-dioxin. *J Reprod Dev*.

Uno S, Dalton TP, Derkenne S, Curran CP, Miller ML, Shertzer HG and Nebert DW (2004) Oral Exposure to Benzo [a] pyrene in the Mouse: Detoxication by Inducible Cytochrome P450 Is More Important Than Metabolic Activation. *Molecular Pharmacology* **65**(5):1225-1237.

Vasquez A, Atallah-Yunes N, Smith FC, You X, Chase SE, Silverstone AE and Vikstrom KL (2003) A Role for the Aryl Hydrocarbon Receptor in Cardiac Physiology and Function as Demonstrated by AhR Knockout Mice. *Cardiovascular Toxicology* **3**(2):153-163.

Walisser JA, Bunger MK, Glover E and Bradfield CA (2004a) Gestational exposure of Ahr and Arnt hypomorphs to dioxin rescues vascular development. *Proc Natl Acad Sci U S A* **101**(47):16677-16682.

Walisser JA, Bunger MK, Glover E, Harstad EB and Bradfield CA (2004b) Patent Ductus Venosus and Dioxin Resistance in Mice Harboring a Hypomorphic Arnt Allele. *J Biol Chem* **279**(16):16326-16331.

Walisser JA, Glover E, Pande K, Liss AL and Bradfield CA (2005) Aryl hydrocarbon receptor-dependent liver development and hepatotoxicity are mediated by different cell types. *Proc Natl Acad Sci U S A* **102**(49):17858-17863.

Ward MP, Mosher JT and Crews ST (1998) Regulation of bHLH-PAS protein subcellular localization during Drosophila embryogenesis. *Development* **125**(9):1599-1608.

Wei YD, Helleberg H, Rannug U and Rannug A (1998) Rapid and transient induction of CYP1A1 gene expression in human cells by the tryptophan photoproduct 6-formylindolo [3, 2-b] carbazole. *Chem Biol Interact* **110**(1-2):39-55.

Wei YD, Rannug U and Rannug A (1999) UV-induced CYP1A1 gene expression in human cells is mediated by tryptophan. *Chem Biol Interact* **118**(2):127-140.

Wernet MF, Mazzoni EO, Celik A, Duncan DM, Duncan I and Desplan C (2006) Stochastic spineless expression creates the retinal mosaic for colour vision. *Nature* **440**(7081):174-180.

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White JG, Southgate E, Thomson JN and Brenner S (1986) The Structure of the Nervous System of the Nematode Caenorhabditis elegans. *Philosophical Transactions of the Royal Society of London Series B, Biological Sciences* **314**(1165):1-340.

Wienholds E, Schulte-Merker S, Walderich B and Plasterk RH (2002) Target-selected inactivation of the zebrafish rag1 gene. *Science* **297**(5578):99-102.

Wienholds E, van Eeden F, Kosters M, Mudde J, Plasterk RH and Cuppen E (2003) Efficient target-selected mutagenesis in zebrafish. *Genome Res* **13**(12):2700-2707.

Wiesner L PW, Karchner SI, Franks DG, Cooper EL, Kauschke E, Hahn ME (2001) cDNA cloning of aryl hydrocarbon receptor (Ahr) homologues in zebra mussel (Dreissena polymorpha) and blue mussel (Mytilus edulis). *Pollutant Responses in Marine Organisms* **11**.

Williamson MA, Gasiewicz TA and Opanashuk LA (2005) Aryl hydrocarbon receptor expression and activity in cerebellar granule neuroblasts: implications for development and dioxin neurotoxicity. *Toxicol Sci* **83**(2):340-348.

Xu C, Li CY and Kong AN (2005) Induction of phase I, II and III drug metabolism/transport by xenobiotics. *Arch Pharm Res* **28**(3):249-268.

Zaher H, Fernandez-Salguero PM, Letterio J, Sheikh MS, Fornace AJ, Roberts AB and Gonzalez FJ (1998) The Involvement of Aryl Hydrocarbon Receptor in the Activation of Transforming Growth Factor-ß and Apoptosis. *Molecular Pharmacology* **54**(2):313-321.

Footnotes:

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Figure Legends:

Figure 1. AHR structure and molecular mechanism. (A), Domain architecture of the mouse AHR protein. Text indicates key domain regions: NLS, nuclear localization sequence; NES, nuclear export sequence; bHLH, basic Helix-loop-Helix domain, PAS, Per-ARNT-Sim domain (A and B repeat regions); TAD, transactivation domain. (B), Mechanism of AHR-mediated gene regulation (see text for details).

Figure 2. Molecular mechanism of AHR-1 function in *C. elegans* social feeding behavior. Social feeding is controlled via pseudocoelomic neuron depolarization, which is coordinately regulated by the oxygen-activated GCY-35/36 heterodimer and NPR-1. GCY-35 directly binds environmental oxygen and facilitates production of cGMP, which subsequently activates Tax-2/Tax-4 ion channels. Neuron depolarization is attenuated by the neuropeptide receptor NPR-1. Loss of AHR-1 function results in a marked decrease in expression of the GCY-35/36 heterodimer as well as NPR-1. This phenomenon is hypothesized to occur via dysregulation of an unknown intermediate factor (see text for details).

Figure 3. Homeotic functions of Spineless. (A), *Upper*, Distal region of a wildtype fruit fly leg. Key: T1 - T5, 1^{st} - 5^{th} tarsal segments; Cl, claw. *Spineless* is transiently expressed in the T2 – T4 segments and the distal portion of T1 during the late 2^{nd} to late 3^{rd} instar. *Lower*, Spineless null animals exhibit truncated legs due to a distal T1 through T4 deletion. (B), Wildtype fly antenna (left). Key: A2 and A3, 2^{nd} and 3^{rd} antennal segments, respectively; Ar, arista. *Spineless* is expressed in A3 and arista beginning in the late 2^{nd} instar. A weak *ss* LOF allele causes

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transformation of A3 and arista to a distal leg phenotype, including a well-formed claw (right). Figure adapted from Duncan et al., 1998, permission granted from Cold Spring Harbor Laboratory Press.

Figure 4. Spineless is a dominant regulator of photoreceptor phenotype. (A), In wildtype *Drosophila*, the compound eye is composed of a mosaic of ommatidia, which express photoreceptors for either short ("pale") or long ("yellow") wavelengths of light (left). Eyes from *ss* nulls contain only "pale" ommatidia (center), whereas ectopic *ss* induces an all "yellow" phenotype (right). (B), During normal development, a stochastic burst of *ss* expression occurs mid-pupation in approximately 70% of all ommatidia. In the presence of Spineless, R7 cells adopt a Rhodopsin 4 (Rh4) phenotype and instruct R8 cells to express Rh6 resulting in a receptor pair that recognizes "yellow" wavelengths. In the absence of Spineless, R7 and R8 cells express the default Rh3 and Rh5 receptors, respectively, that recognize "pale" light.

Figure 5. Fetal and adult expression of murine *Ahr*. (A), Hemotoxylin-eosin stain (left) and corresponding *in situ* hybridization for *Ahr* mRNA (right) at gestation day 13.5. Text indicates key anatomical features: ne, neuroepithelium; lv, lateral ventricle; mb, midbrain; mv, mesencephalic vescicle; cp, choroid plexus; ap, anterior pituitary; P, palate; ds, dorsal tongue surface; T, tongue; t, thymus; H, heart; Lu, lung; Li, liver; g, gut; us, urogenital sinus; gt, genital tubercle. Figure adapted from Jain et al., 1998, permission granted from Elsevier Press. (B), *Ahr* expression in select tissues from 8-11 week old C57BL/6 mice, as measured by microarray hybridization.

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Figure 6. Vascular phenotype of *Ahr* null mice. (A), Time-lapse radiographs of contrast agent entering the liver of an *Ahr* wildtype (top) or null (bottom) mouse. Contrast agent is perfused throughout the branching vessels of the liver in wildtype mice, but shunted into the inferior vena cava in null mice via a persistent fetal vascular structure known as the ductus venosus (see text for details). Arrows indicate key features: PV, portal vein; IVC, inferior vena cava; DV, ductus venosus. (B), Latex corrosion cast of the wildtype (left) and null (right) renal vasculature. The mechanism underlying decreased vascular density in the null kidney is currently unknown.

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Table 1:

***ahr-1* loss of function phenotype**	**Cells**	**Cell type**
Failure to express genes required for social feeding behavior	URXR, URXL, AQR?, PQR?	Pseudocoelomic sensory neuron
Defects in cell pathfinding and axon extension	AVM, SQDR, PLML, PLMR	Mechanosensory neuron
Cell fate/identity	RMEL, RMER	Ring motor neuron
None reported	ALNR/L, PVM, BDUR/L, PLNR/L, PHCR/L, PVWR/L, SDQL, MI, I3, ASKP/R, RIPR/L	Neuron
	T.pa, T.ppa, T.ppp, G2, W	Blast cell
	PHso1, PHso2	Phasmid socket cell

Table 1. *C. elegans*: AHR-1 expression and loss of function phenotype. An *ahr-1::gfp* construct driven by the *ahr-1* promoter was utilized to determine localization of the protein. AHR-1 was detected in 28 neurons, 5 blast cells, and two phasmid socket cells. Animals homozygous for AHR-1 LOF mutations exhibit several defects in the development and/or function of these cells (see text for details).

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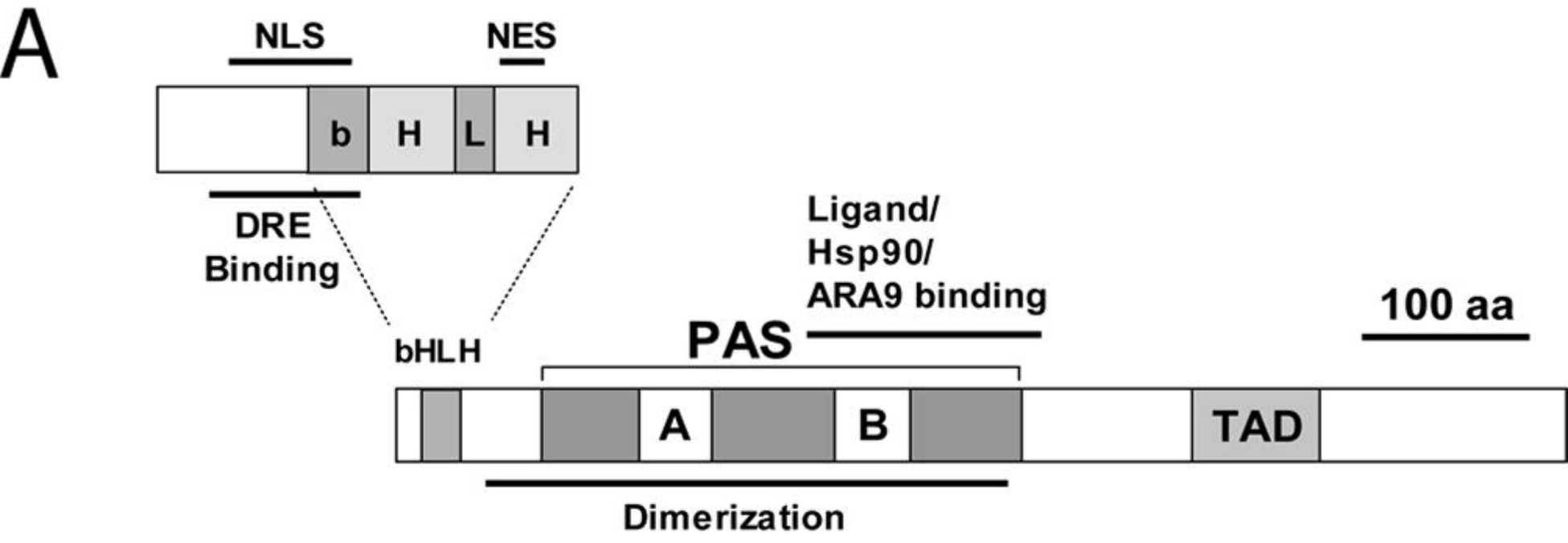
Figure 1


A
NLS
NES
b
H
L
H
DRE
Binding
Ligand/
Hsp90/
ARA9 binding
100 aa
bHLH
PAS
A
B
TAD
Dimerization

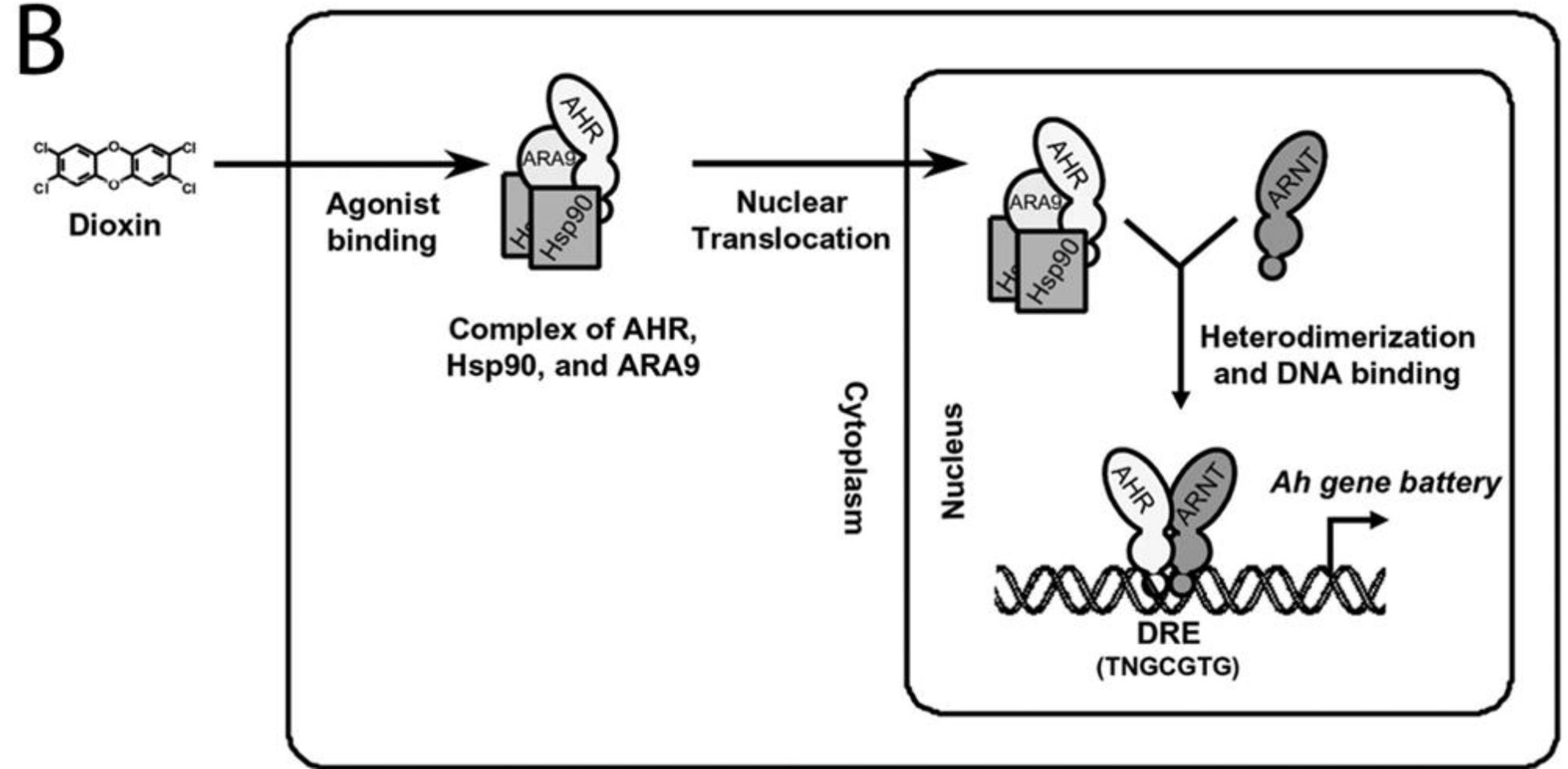

B
Dioxin
Agonist
binding
AHR
ARA9
Hsp90
Complex of AHR,
Hsp90, and ARA9
Nuclear
Translocation
ARNT
Heterodimerization
and DNA binding
Cytoplasm
Nucleus
Ah gene battery
DRE
(TNGCGTG)

Figure 2

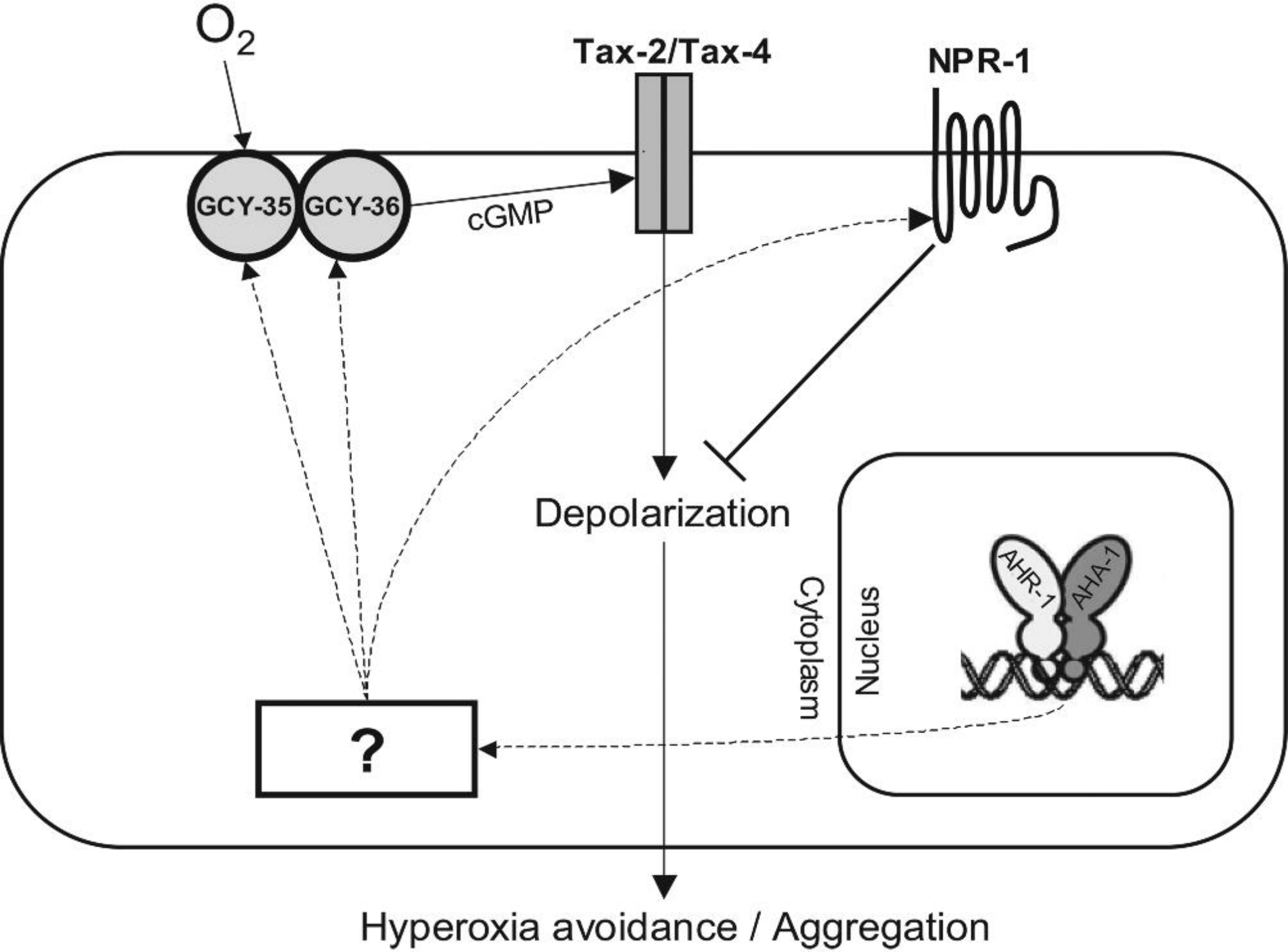

O_2
Tax-2/Tax-4
NPR-1
GCY-35
GCY-36
cGMP
Depolarization
Cytoplasm
Nucleus
AHR-1
AHA-1
?
Hyperoxia avoidance / Aggregation

Figure 3

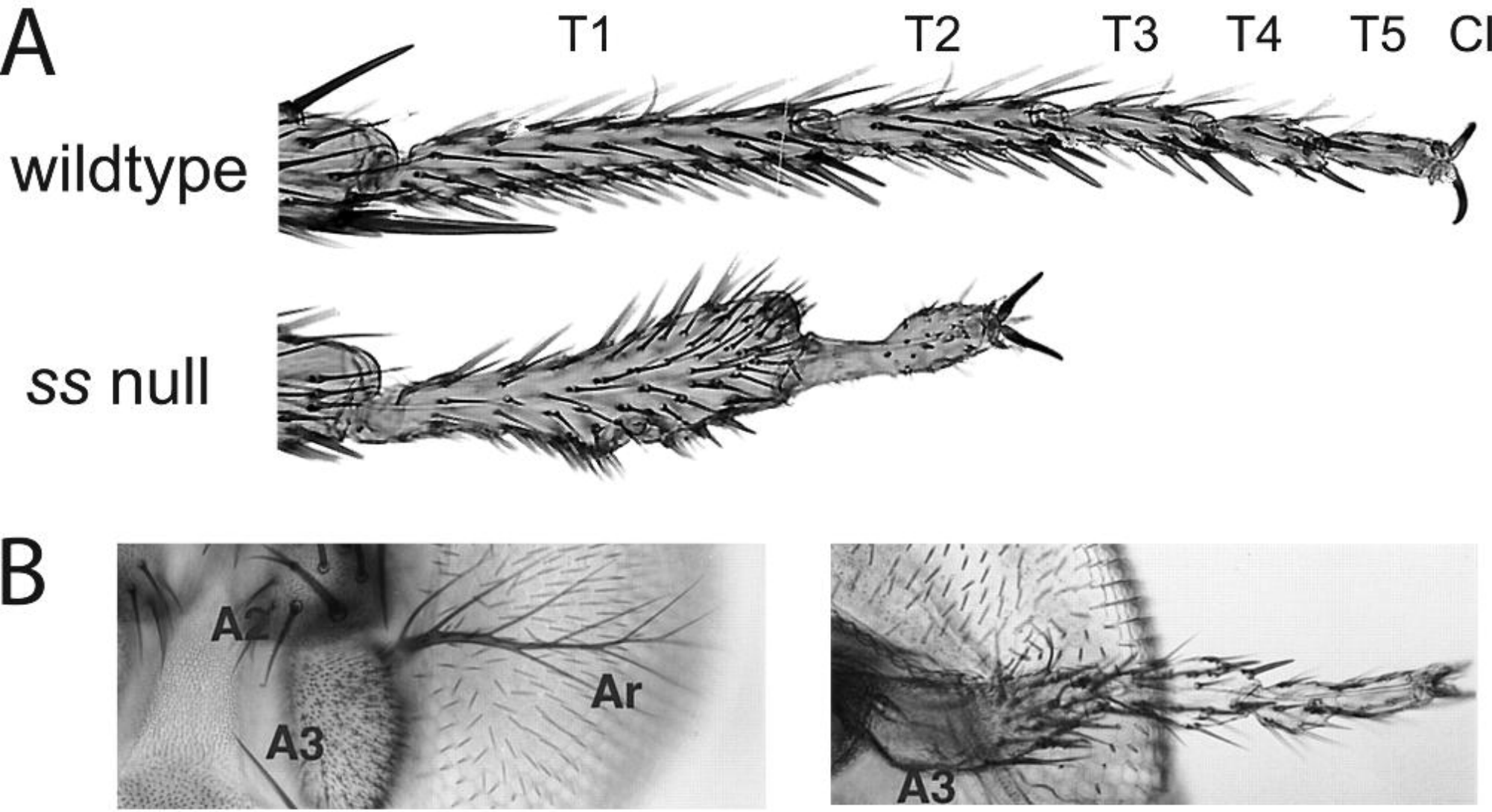

A
T1
T2
T3
T4
T5
wildtype
ss null
B
A2
Ar
A3
wildtype
A3
ss mutant

Figure 4

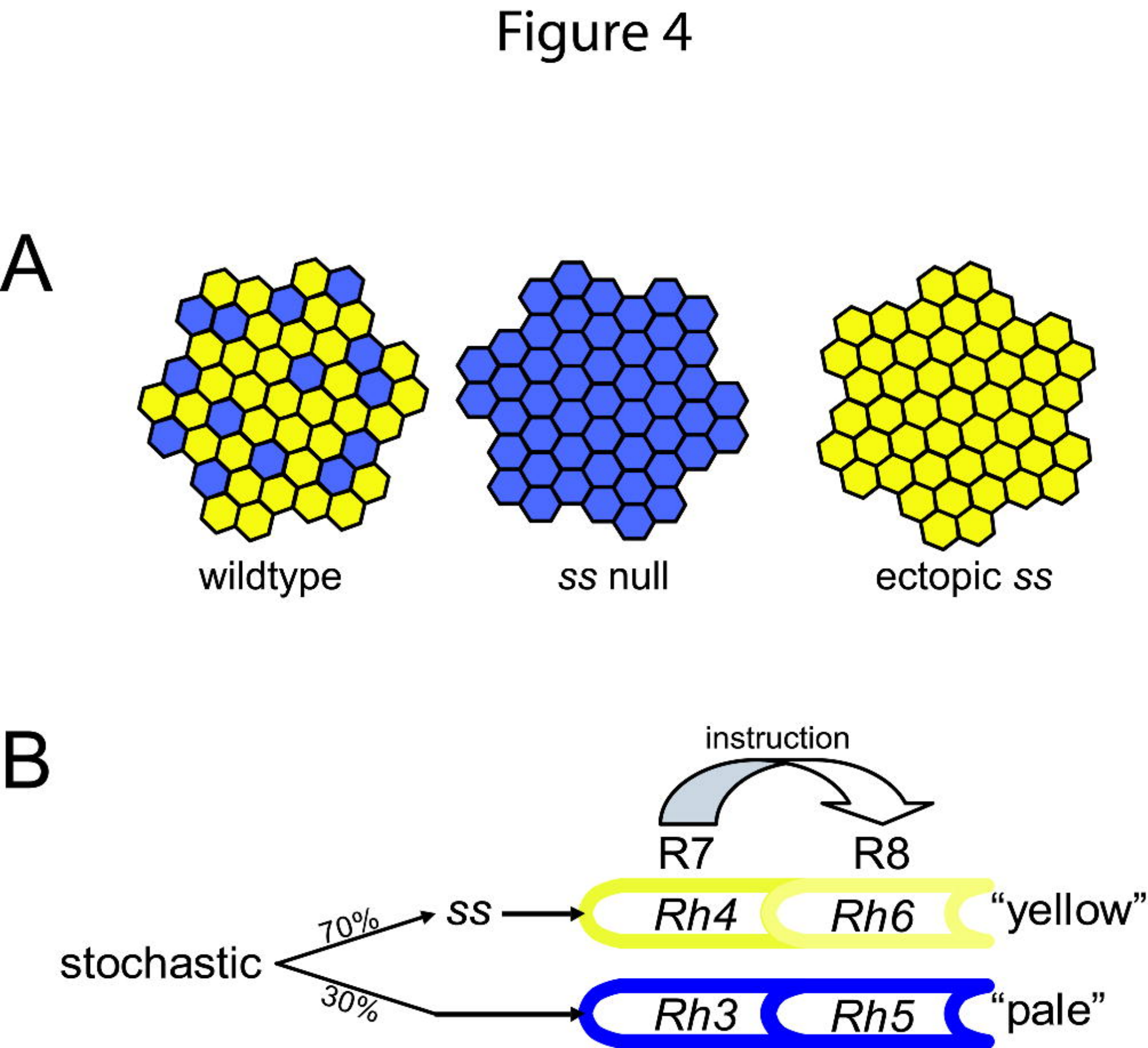
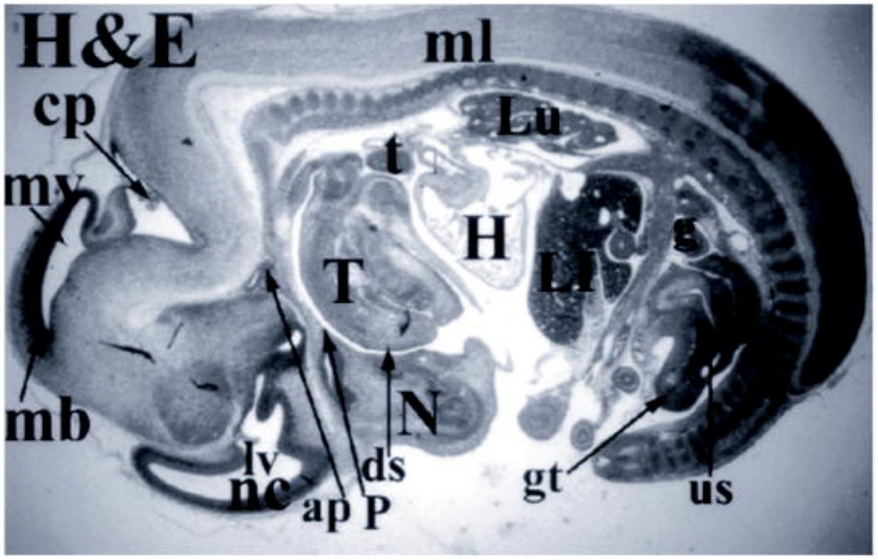
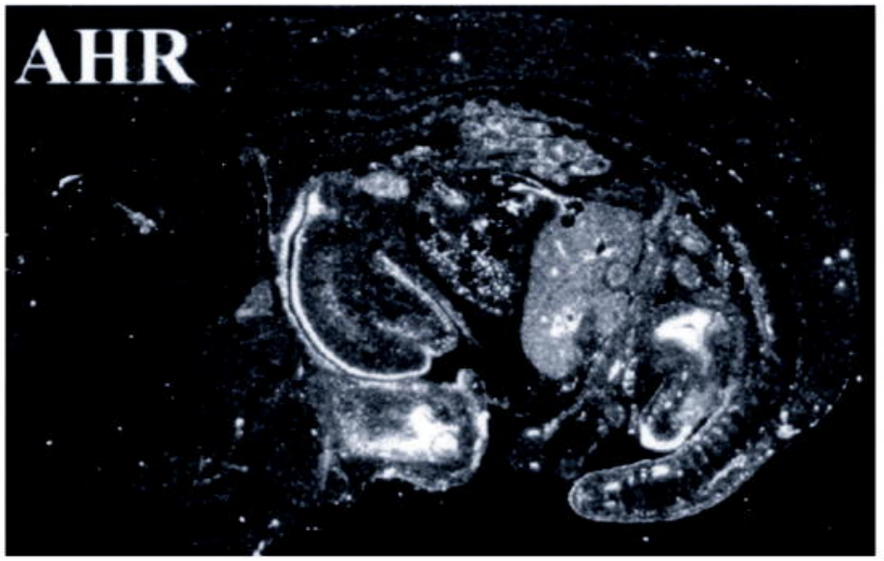

A
wildtype
ss null
ectopic ss
B
instruction
R7
R8
ss
Rh4
Rh6
"yellow"
70%
stochastic
30%
Rh3
Rh5
"pale"

Figure 5

A


H&E
ml
cp
Lu
t
my
H
T
Li
s
mb
N
lv
ds
nc
ap
P
gt
us


AHR

B

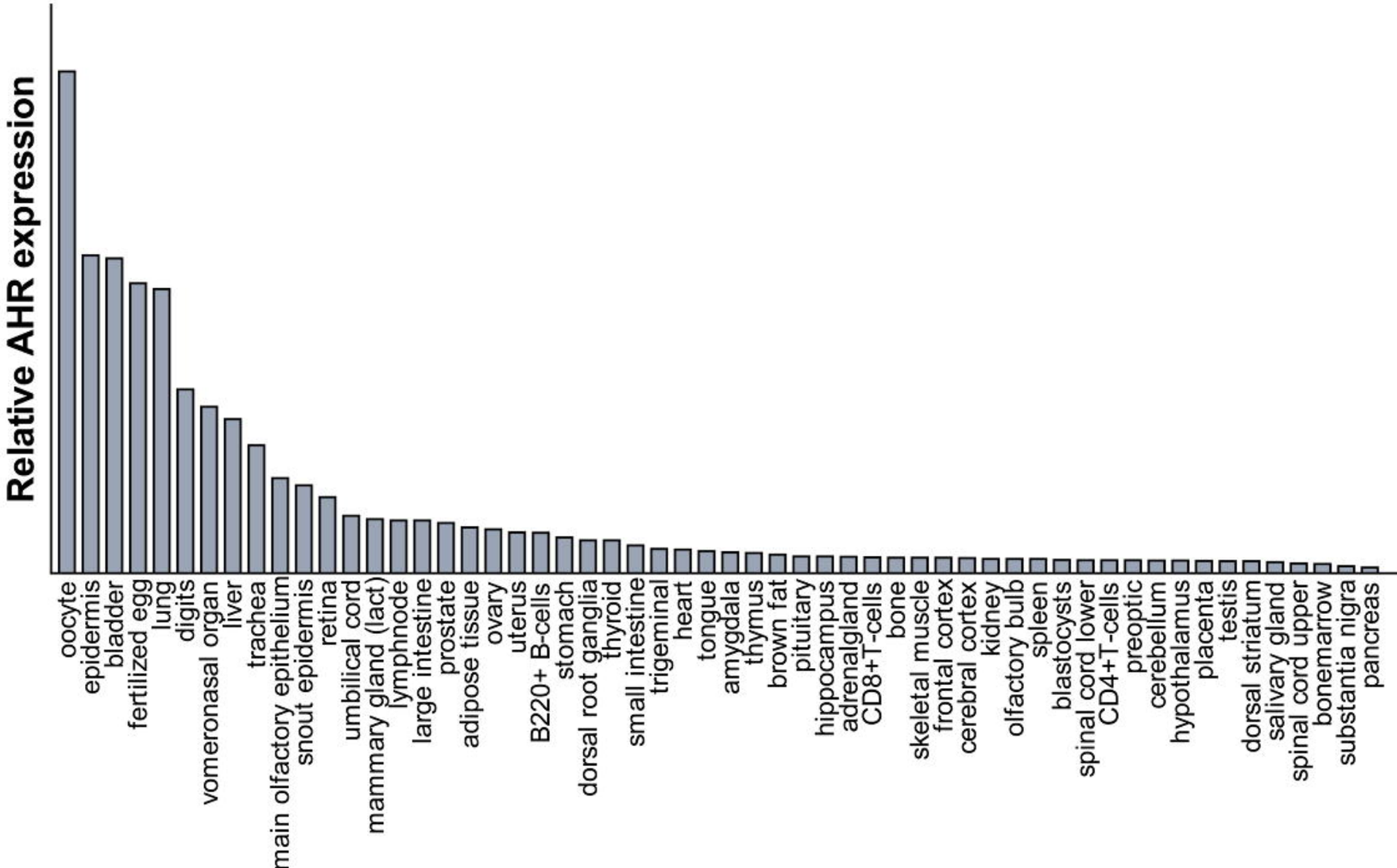

Relative AHR expression
oocyte
epidermis
bladder
fertilized egg
lung
digits
vomeronasal organ
liver
trachea
main olfactory epithelium
snout epidermis
retina
umbilical cord
mammary gland (lact)
lymphnode
large intestine
prostate
adipose tissue
ovary
uterus
B220+ B-cells
stomach
dorsal root ganglia
thyroid
small intestine
trigeminal
heart
tongue
amygdala
thymus
brown fat
pituitary
hippocampus
adrenalgland
CD8+T-cells
bone
skeletal muscle
frontal cortex
cerebral cortex
kidney
olfactory bulb
spleen
blastocysts
spinal cord lower
CD4+T-cells
preoptic
cerebellum
hypothalamus
placenta
testis
dorsal striatum
salivary gland
spinal cord upper
bonemarrow
substantia nigra
pancreas

Figure 6