LETTER

The teleost *agouti-related protein 2* gene is an ohnolog gone missing from the tetrapod genome

A central question in evolutionary genomics is how morphological differences between taxa arise from differences in gene repertoire. Answers require rigorous orthology assignments. Recent work (1) illuminated the mechanism of background adaptation in teleost fishes by investigating the function of *agouti-related protein 2* (*agrp2*), a gene so far found only in teleosts.

Agouti family proteins antagonize melanocortin receptors, function in energy homeostasis, and regulate pigmentation. The authors (1) proposed a neurocrine axis in which zebrafish Agrp2 is expressed exclusively in the pineal gland, where it binds to the melanocortin 1 receptor and thereby, controls expression of melanin-concentrating hormones; this causes melanosome contraction in melanophores and adaptation to light backgrounds (1). These findings intriguingly implied that the acquisition of *agrp2* in teleosts enabled the evolution of background adaptation by regulation through the pineal.

Tetrapods have two agouti family members [*Agouti signaling protein* (*Asip*) and *Agrp*], but teleosts have four (*asip1*, *asip2*, *agrp1*, and *agrp2*). Published studies (1, 2) vaguely suggest that *agrp2* is a paralog of *agrp1* generated during the teleost-specific genome duplication (TSGD). Phylogenetic analyses, however, remain ambiguous regarding vertebrate agouti gene relationships (2) (Fig. 1*A*).

Our investigation of this gene family using synteny data clearly reveals that the teleost *agrp2* chromosomal region shares syntenies neither with the teleost *agrp1* region nor with the tetrapod *Agrp* region (Fig. 1*B*), contradicting the current gene nomenclature (2). In contrast, teleost *agrp2* and *asip2* regions show conserved synteny to a region on human chromosome 8 (Hsa8) (Fig. 1*C*). Furthermore, we find at least three regions of paralogy in the human genome (Fig. 1*D*)—Hsa16 (*AGRP* region), Hsa8, and Hsa20 (*ASIP* region)—most likely derived from a single *Asip*/*Agrp* region on ancestral vertebrate protochromosome B (3). The *ASIP* region on Hsa20 shares more paralogies with the region on Hsa8 (Fig. 1*D*). According to our model (Fig. 1*E*), an ancestral *Asip*/*Agrp* precursor gene was duplicated two times during the two rounds of vertebrate genome duplication (R1 and R2). After R2, *Asip* and *Agrp* were retained in all bony vertebrates, but a third ohnolog (4) went missing in the tetrapod lineage. This gene was retained, however, in the rayfin fish lineage, giving rise to *agrp2* and *asip2* after the TSGD.

Our results support a revision of agouti family nomenclature (Fig. 1*E*). Importantly, our model suggests that the genetic basis of teleost background adaptation may not root in the agouti family expansion during the TSGD but to R2, far deeper in vertebrate evolution (Fig. 1*E*). It is unlikely that zebrafish *agrp2* (new name: *asip2b*) newly evolved its single pineal-specific function after the TSGD (as suggested by ref. 1), because *asip2* (*asip2a*) is absent in zebrafish but present in other teleosts. The interesting findings of Zhang et al. (1) should, thus, spur research on the function of *asip2a*/*b* in other teleosts and agouti genes in basal rayfin fishes, basal lobefins like lungfish and coelacanth, background-adapting tetrapods, and more basal vertebrates in the light of our rigorous phylogenetic framework (Fig. 1*E*).

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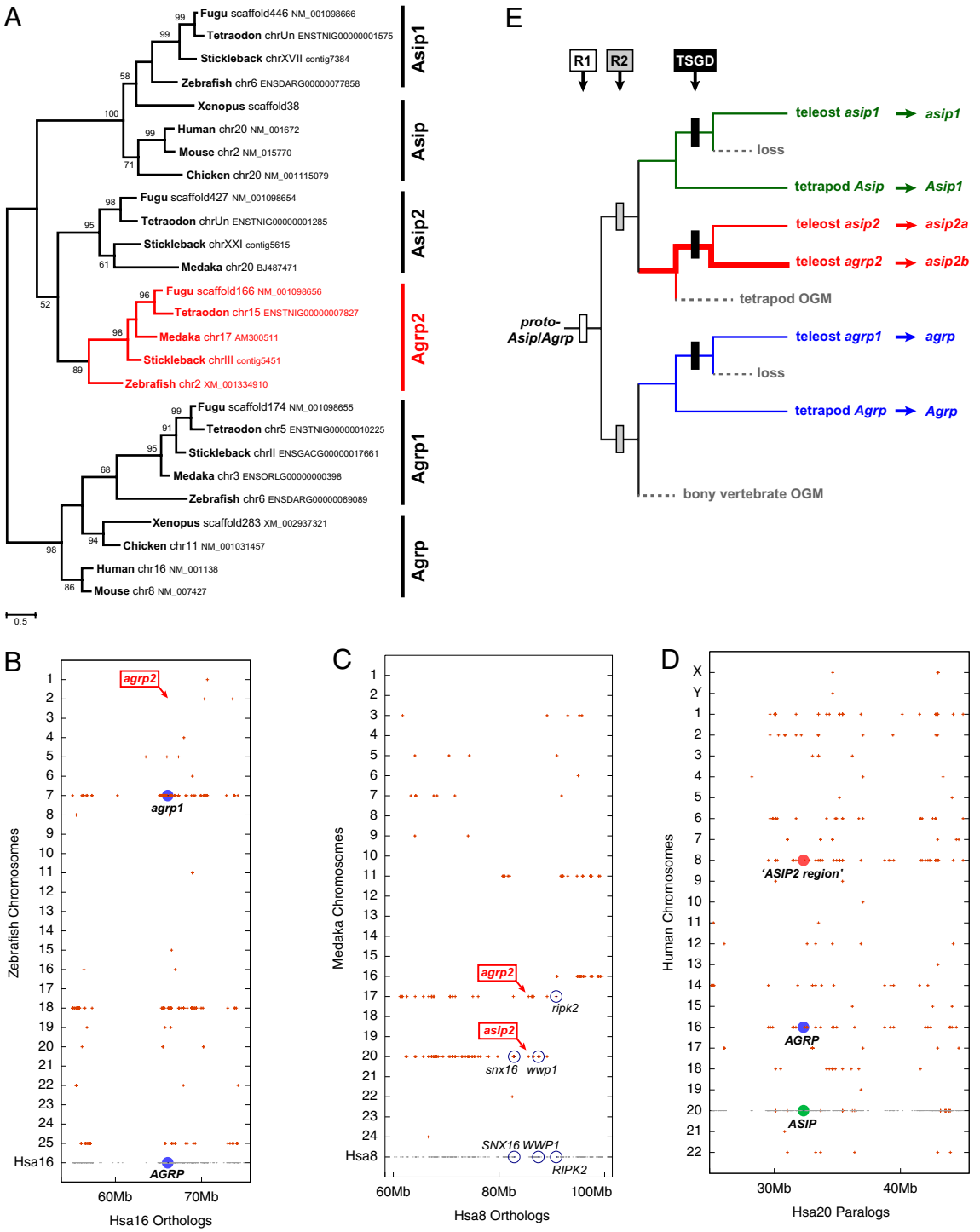

Fig. 1. Evolution of vertebrate agouti genes. (*A*) Maximum likelihood phylogeny of vertebrate agouti family proteins with current names (JTT+G+I model; 100 bootstrap replicates). (*B–D*) Conserved synteny dot plots derived from the Synteny Database (5). (*B*) The zebrafish *agrp2* region on Dre2 (red box) shares conserved syntenies neither with the zebrafish *agrp1* region (Dre7) nor with the human *AGRP* region (Hsa16). (*C*) The *agrp2* and *asip2* regions in medaka and other teleosts share conserved synteny with each other and with a region on human Hsa8, including several *agrp2*- and *asip2*-neighboring genes. (*D*) Analysis of the human genome shows that the *ASIP* region on Hsa20 shows more paralogous connections to the inferred *ASIP2* region on Hsa8 than to the *AGRP* region on Hsa16 (43 vs. 25 genes, respectively). (*E*) Model for the evolution of the vertebrate agouti family by three rounds of genome duplication, including a revised gene nomenclature. The pineal-specific function of teleost *agrp2* (new name: *asip2b*) could have evolved anywhere along the thick red line. OGM, ohnolog gone missing.