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Erratum

Minigenes Impart Odorant Receptor-Specific Axon Guidance in the Olfactory Bulb

Due to a printing error, Figures 1, 2, and 4 had poor color quality and image detail. These figures are being reprinted on the following pages as an erratum. They originally appeared in the article by Vassalli et al. (Neuron *35*, 681–696 [August 15, 2002]).

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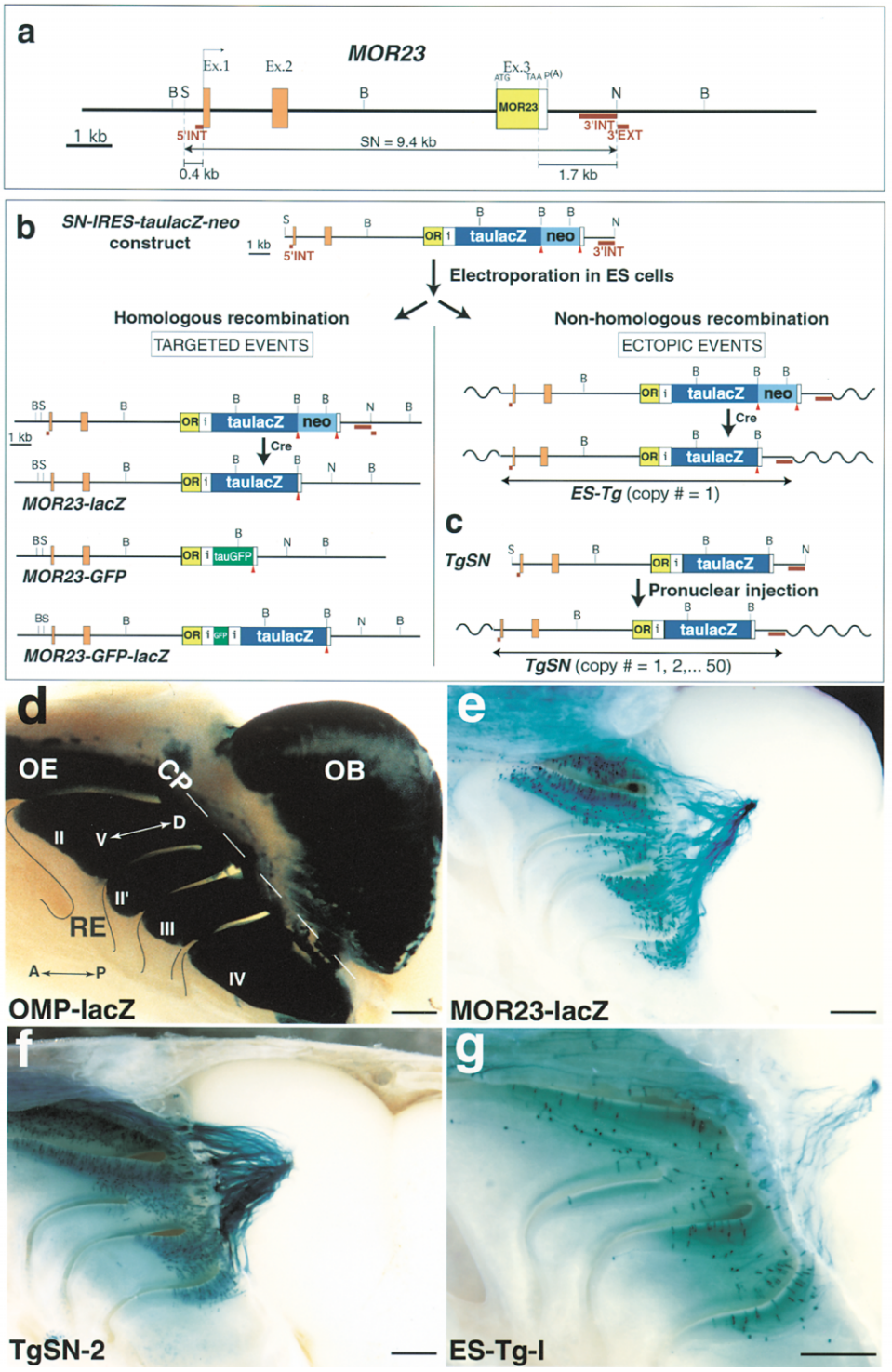

Figure 1. Structure, Targeted Mutagenesis, and Transgenesis of the *MOR23* Gene

(a) The *MOR23* gene, with its three-exon structure defined by 5′RACE analysis of C57BL/6J mouse RNA. The putative TSS (hooked arrow) is located at position 1618 in GenBank X92969. Two exons (orange boxes), of 154 bp and 349 bp, respectively, are present in the 5′ noncoding region. The introns span 1.4 and 4.5 kb, respectively. The coding region (930 bp, yellow box) is encoded within a single exon containing 16 bp of 5′ noncoding sequence. The 3′ noncoding region (white box) is shown to extend to a unique polyadenylation signal found within a 1.7 kb region downstream of the stop codon. This 3′ noncoding region contains putative donor and acceptor splice sites, defining a hypothetical ~100 bp intron. The 9.4 kb SacI-NheI (SN) fragment is the substrate for targeting vector and transgene construction. Red bars represent the probes used in genomic Southern blots: 5′INT, 3′INT, used as 5′ and 3′ internal probes, respectively, of the SN fragment; 3′EXT, probe external to the SN fragment, used to identify targeted ES clones. B, BamHI; N, NheI; S, SacI.

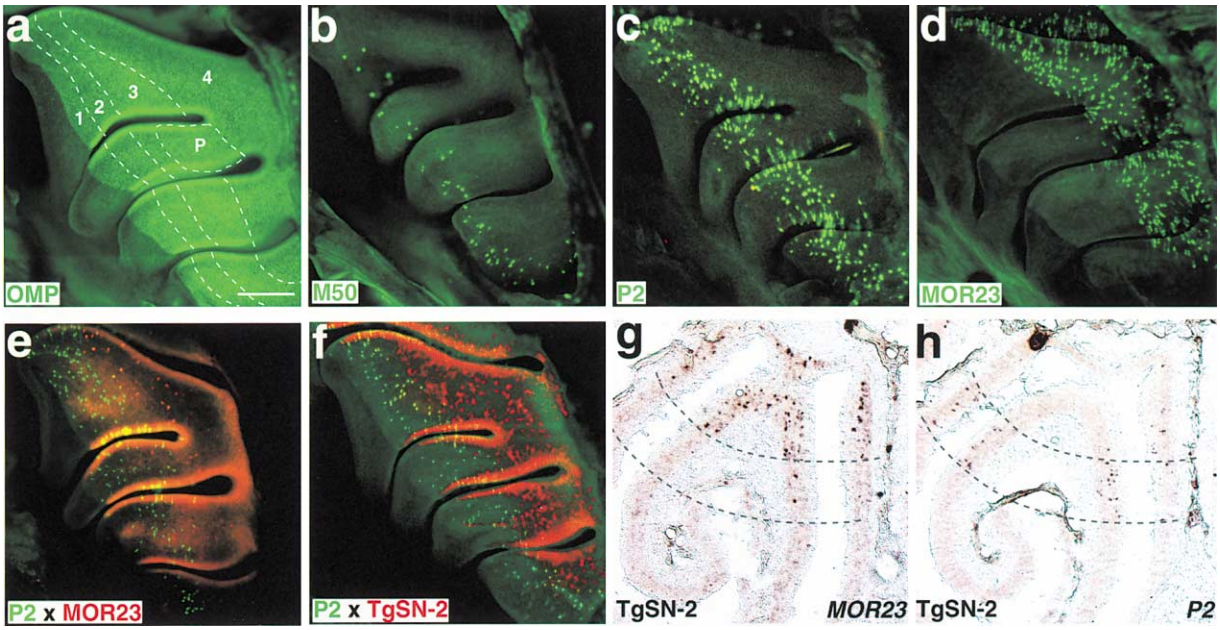

Figure 2. *MOR23* Targeted and Transgenic Expression in Zone 4 of the Olfactory Epithelium

Neuronal distribution of expression in the olfactory epithelium was assayed in whole-mount specimens. GFP was imaged by its intrinsic green fluorescence. β-galactosidase activity was revealed by exposure to Xgal and Fast Red Violet LB and imaging of the red fluorescence. (a–d) GFP-tagged targeted alleles of OMP or various OR genes illustrate the zonal boundaries. (a) OMP-GFP expression; four expression zones (1, 2, 3, 4) and a patch (P) (Strotmann et al., 2000; Pyrski et al., 2001) are shown diagrammatically. (b–d) Zone-specific expression of M50-GFP (zone 1) (P.F. et al., unpublished data); P2-GFP (zone 3); MOR23-GFP (zone 4). (e and f) β-galactosidase activity (red) reveals neurons expressing the targeted MOR23-lacZ (e) or the transgenic TgSN-2 (f) alleles. These cells define a zone dorsal to the zone of P2-GFP-expressing neurons and thus corresponding to zone 4. Note that a fine degree of intermingling between MOR23 and P2-expressing cells exists at the zone 3/4 boundary. TgSN2 is expressed in ~4-fold as many cells as the reference MOR23 targeted allele. Scale bar (a–f), 500 μm. (g and h) In situ RNA hybridization of serial coronal sections of a TgSN-2 heterozygous mouse using probes for *MOR23* (g) or *P2* (h). The cells expressing the *MOR23* endogenous gene or the TgSN-2 transgene define a zone that is dorsal to the cells expressing P2. Dorsal is to the top. All mice shown are heterozygous for the targeted alleles or hemizygous for the transgene, except (a) is a OMP-GFP homozygote, and (d) is a MOR23-GFP homozygote. Mice were P9 to P11 (a–f) or 3 weeks old (g and h).

(b) Production of gene-targeted and transgenic mice. The targeting construct was electroporated into ES cells. Southern blot analysis of G418-resistant ES cell clones identified homologous (targeted) and nonhomologous (ectopic) recombination events. Three gene-targeted mutations were generated with different cassettes. Ectopic events were further screened for single-copy (n = 1) integration of a full-length transgene, using the 5′INT and 3′INT probes. Cre-mediated site-specific recombination served in both cases to excise the *neo* selectable marker; the red triangles flanking the *neo* gene are *loxP* sites. OR, *MOR23* coding sequence; i, *IRES*.

(c) A second series of transgenic lines was generated by microinjection of the SN construct into one-cell embryos, resulting in transgenic strains (TgSN) that carry one or frequently several transgene copies.

(d–g) Expression patterns in MOR23-lacZ targeted and SN transgenic mice. Whole-mount preparations of sagitally transected mouse heads, showing the medial aspect of the turbinates and the medial face of the bulb. (d) OMP-taulacZ mouse (Mombaerts et al., 1996). Olfactory marker protein (OMP) is expressed in all mature OSNs. The taulacZ marker was revealed as a blue precipitate by incubation with Xgal. OE, olfactory epithelium; OB, olfactory bulb; CP, cribriform plate, separating nasal from cranial cavity; RE, nonneuronal respiratory epithelium. The dorsoventral (D-V) orientation of the epithelium and the anteroposterior (A-P) alignment of the nasal cavity are indicated. This view reveals endoturbinates II, II′, III, and IV. *MOR23* targeted (e) and transgenic mice (f and g) express the taulacZ marker along with MOR23. In the medial hemisphere of the bulb, labeled axons converge invariably to a single target glomerulus. Individual transgenic lines show labeled cell densities that are either below (g) or above (f) the reference pattern (e). The MOR23-lacZ (e) and ES-Tg-I (g) mice are homozygous, and the TgSN-2 mouse (f) is hemizygous. Mice were 3–3.5 weeks old. In some cases, the images were flipped along a vertical axis to point in the same direction. Scale bars, 500 μm.

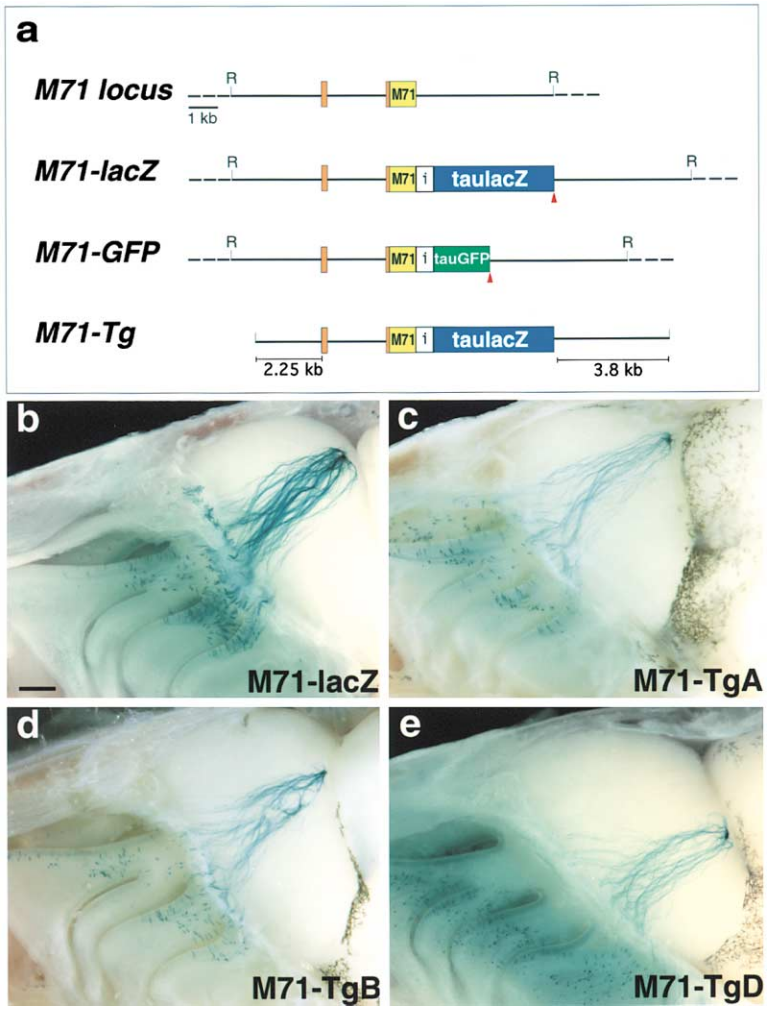

Figure 4. *M71* Transgenic Mice

(a) Structure, targeted mutagenesis, and transgenesis of *M71*. (*M71 locus*) The coding sequence (yellow box) and 5′ noncoding sequences (orange boxes) are shown. 5′RACE analysis mapped a putative TSS 2.2 kb upstream of the ATG. The 5′ noncoding exon is 175 bp and is followed by a 2.0 kb intron that ends 18 bp upstream of the start codon. R, EcoRI. (*M71-lacZ*) The targeted *M71-IRES-taulacZ* allele after Cre-mediated recombination. (*M71-GFP*) The targeted *M71-IRES-tauGFP* allele after Cre-mediated recombination. (*M71-Tg*) The transgene.

(b–e) Whole-mount specimens of a M71-lacZ heterozygous mouse (b) and hemizygous mice of various transgenic lines (c–e). In line M71-TgD (e), most of the labeled OSN cell bodies show a ventral shift compared to the reference pattern (b). Note that this correlates with a ventral shift of the medial glomerulus in the bulb. Mice were 3–4 weeks old. Scale bar, 500 μm.