LETTER

Reply to Thornton and Carroll: Lamprey possess a highly specific corticosteroid signaling system

Close et al. (1) provided direct evidence that 11-deoxycortisol (S) is the corticosteroid hormone in the sea lamprey, *Petromyzon marinus*. Although two putative corticosteroids, S and 11-deoxycorticosterone (DOC), were directly identified in circulation, DOC was shown to be nonfunctional in the lamprey. Our radio-ligand binding studies revealed a single receptor population highly specific to only S, with characteristic binding constants found in other glucocorticoid receptors in vertebrates. Determination of S as a corticosteroid hormone in the lamprey was further supported by a variety of physiological experiments demonstrating that S levels in plasma responded upon exposure to acute stress and that S is under control of the hypothalamus pituitary interrenal axis. These results strongly indicate that S is the only corticosteroid that can bind and activate lamprey corticosteroid receptor (CR).

We made no conceptual error in our article, as claimed by Thornton and Carroll (2). Although unlikely, it is certainly possible that the endocrine features that we have reported in lamprey represent a derived state. We should not ignore the more likely possibility that the endocrinology of the lamprey is similar to that of basal vertebrates, as it is in many aspects of its morphology, physiology, and genetics. We were careful in our article to indicate that if the end extant lamprey is representative of basal vertebrates, then our findings have important implications for the evolution of corticosteroid hormone signaling (1).

The approaches we used in our study indicate that S is a hormone in lamprey with a highly specific receptor. These results contrast with the heterologous transactivation studies by Thornton's group (3), which found broad corticosteroid binding for the lamprey CR. Therefore, one can legitimately question the validity of heterologous expression approaches for investigating basal vertebrate CR function. Such promiscuous activation found in Bridgham et al. (3) may be an artifact because of the absence of the whole receptor or region outside of ligand binding domain (4), the lack of lamprey chaperone proteins (5), or the result of assay conditions (6). Although the results of Close et al. (1) indicate that a highly specific corticosteroid signaling pathway is present in lamprey, further in vivo and in vitro experiments can determine whether promiscuous activation is present or is an experimental artifact.

In summary, we identified a corticosteroid (S) with a highly specific receptor in the lamprey by adopting multiple approaches. Our data raise questions about the validity of promiscuous activation of the lamprey CR by multiple corticosteroids reported by Bridgham et al. (3). In any event, a variety of approaches will be necessary to demonstrate an endocrine pathway, and no single approach should be accepted as definitive, especially when it is as indirect as the approach in Bridgham et al. (3).

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