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Review

Osmoregulatory actions of the GH/IGF axis in non-salmonid teleosts

Juan Miguel Mancera [a,b,*], Stephen D. McCormick [b]

[a] *Departamento de Biología Animal, Facultad de Ciencias del Mar, Universidad de Cádiz, 11510 Puerto Real, Cadiz, Spain*
[b] *Conte Anadromous Fish Research Center, Biological Resources Division, USGS, P.O. Box 796, Turners Falls, MA 01376, USA*

Abstract

Salmonid fishes provided the first findings on the influence of the growth hormone (GH)/insulin-like growth factor I (IGF-I) axis on osmoregulation in teleost fishes. Recent studies on non-salmonid species, however, indicate that this physiological action of the GH/IGF-I axis is not restricted to salmonids or anadromous fishes. GH-producing cells in the pituitary of fish acclimated to different salinities show different degrees of activation depending on the species studied. Plasma GH levels either increase or do not change after transfer of fish from freshwater to seawater. Treatment with GH or IGF-I increases salinity tolerance and/or increases gill Na^+,K^+-ATPase activity of killifish (*Fundulus heteroclitus*), tilapia (*Oreochromis mossambicus* and *Oreochromis niloticus*) and striped bass (*Morone saxatilis*). As in salmonids, a positive interaction between GH and cortisol for improving hypoosmoregulatory capacity has been described in tilapia (*O. mossambicus*). Research on the osmoregulatory role of the GH/IGF-I axis is derived from a small number of teleost species. The study of more species with different osmoregulary patterns will be necessary to fully clarify the osmoregulatory role of GH/IGF-I axis in fish. The available data does suggest, however, that the influence of the GH/IGF-I axis on osmoregulation may be a common feature of euryhalinity in teleosts. © 1998 Elsevier Science Inc. All rights reserved.

1. Introduction

The somatomedin hypothesis suggests that growth hormone (GH) stimulates the synthesis of insulin-like growth factor I (IGF-I), predominantly from the liver, which carries out some or all of the physiological actions of GH [11,19,21]. Research over the last 2 decades indicates that the GH/IGF-I axis, in addition to its somatotropic actions, also promotes seawater acclimation in salmonids species [9,23] (for review see Refs. [31,45]). Since seawater entry in anadromous salmonids is associated with a period of rapid growth, it was thought that this osmoregulatory function of GH

and IGF-I may be restricted to anadromous species or even just salmonids. Research in the last few years indicates that this is not the case. In this mini-review we will analyze the recent data on the hypoosmoregulatory actions of the GH/IGF-I axis in non-salmonid teleosts.

2. GH/IGFs and environmental salinities

Morphological data on GH-producing cells (somatotropes) in the pituitary of fish adapted to different salinities show different patterns of activation depending of the species studied (for review see [4,36,38]). In tilapia, *Oreochromis mossambicus*, in situ studies show similar pattern of mRNA expression in pituitary GH-cells of freshwater (FW) and seawater (SW) adapted

* Corresponding author. Tel.: + 34 56 470834; fax: + 34 56 470811; e-mail: juanmiguel.mancera@uca.es

fish [37]. In the same species, no significant difference in pituitary GH mRNA levels were observed between FW- and SW-acclimated larvae [3]. However, morphological studies indicate greater pituitary GH cell activity in SW-acclimated compared to FW-acclimated juvenile *O. mossambicus* [6].

Plasma GH levels show different patterns of change after transfer from FW to SW. Eel (*Anguilla japonica*) transferred to SW had no change in plasma GH levels [51,52] or metabolic clearance rate [12]. Nor did in vitro osmotic pressure affect GH secretion from the eel pituitary [51]. In the Nile tilapia (*Oreochromis niloticus*) plasma GH did not change upon transfer from FW to brackish water (BW) [1]. Tilapia had similar plasma GH levels in fish acclimated for 3–4 weeks to FW and SW (for *O. mossambicus*) or 50% SW (for *O. niloticus*) [2]. Other studies with *O. mossambicus* report modifications in plasma GH levels in males specimens after transfer to a hyperosmotic environment [56]. In addition, increased plasma GH in *O. mossambicus* after 4 and 14 days in SW suggests a role of this hormone in the acclimation phase [34,53].

Plasma IGF-I levels in fish have been evaluated using heterologous and homologous radioimmunoassay. Using these techniques it has been demonstrated that GH treatment increases IGF-I levels in salmonid and non salmonids species [5,35,37,41]. In salmonids, plasma IGF-I increases during the parr-smolt transformation and SW-acclimation [24] (McCormick and Moriyama, unpublished observations). We are not aware of any published information on osmoregulation-related changes in circulating IGF-I in non-salmonid fishes.

The expression of IGF-I and IGF-II mRNA has been reported in different tissues of salmonids and non-salmonids species [8,13–15,39]. In salmonids, the IGF-I mRNA expression in gills is GH dependent [14] and this expression increases in the gills following SW exposure [44] and during the parr-smolt transformation [46]. In the gilthead seabream (*Sparus aurata*), however, GH treatment did not increase IGF-I or IGF-II mRNA expression in gills [15]. Relatively high levels of IGF-II mRNA were found in gills of *Oncorhynchus mykiss* [8] and *S. aurata* [15]. However, information on modifications of IGF-I and IGF-II mRNA expression in osmoregulatory organs in relation to osmoregulatory processes is completely lacking in non-salmonids species.

IGF-bindings proteins (IGF-BPs), with several biological actions, have been described in plasma of different species [22]. In teleosts several circulating IGFBPs have also been identified (for review see Ref. [50]). However, there is no report about the possible osmoregulatory functions of IGFBPs in salmonid or non-salmonid teleosts.

3. Effects of GH treatment

The osmoregulatory effects of GH have been analyzed in only a few non-salmonids species. To date, only two tilapia species (*O. mossambicus* and *O. niloticus*), the anadromous striped bass (*Morone saxatilis*) and the euryhaline mummichog (*Fundulus heteroclitus*) have been examined.

Long-term treatment of tilapia with recombinant tilapia GH increased chloride cell density in the opercular membrane [17]. Treatment with ovine GH (oGH) or tilapia GH (tGH) increased salinity tolerance and stimulated gill Na^+,K^+-ATPase activity of tilapia [6,47,48]. In accordance with these results, treatment with tGH decreased plasma osmolality and increased gill Na^+,K^+-ATPase activity of tilapia after transfer to SW [47]. In a less euryhaline FW species of tilapia (*O. niloticus*), however, treatment with recombinant tGH did not improve the adaptability of this species to BW [1]. However, Xu et al. [57] reported an osmoregulatory actions of recombinant eel GH (reGH) in *O. niloticus*. In this study treatment with reGH enhanced SW adaptation and stimulated the differentiation of chloride cells. The different source of hormone or experimental design could explain the different results of GH treatment on the osmoregulatory system of *O. niloticus*.

The striped bass, *M. saxatilis*, is an anadromous percichthyid teleost with well developed SW tolerance [26]. Treatment of hypophysectomized fish maintained in FW with recombinant striped bass GH (rsbGH) improved gill Na^+,K^+-ATPase activity and the capacity of the fish to maintain plasma osmolality. In addition, a weak hypoosmoregulatory effect was observed after treatment with rsbGH in SW-acclimated fish [28].The euryhaline mummichog, *F. heteroclitus*, was the subject of some of the earliest studies on the endocrine control of ion transport in fish (e.g. [42,43]; for review see Ref. [55]) but the role of GH and IGFs in SW-acclimation has not been examined until recently. In *F. heteroclitus* a single injection of oGH improved their ability to maintain plasma osmolality and to increase gill Na^+,K^+-ATPase activity after transfer from BW (1.0% salinity) to SW (3.5% salinity) [29]. Similarly, 10 days of oGH treatment, using a slow-release system (vegetable oil), also improved the hypoosmoregulatory capacity of *F. heteroclitus* after transfer to high salinity (Mancera and McCormick, unpublished results).

4. Effects of IGF treatment

Several studies have shown that exogenous IGF-I increases salinity tolerance and hypoosmoregulatory mechanisms of salmonids [27,32,33]. To our knowledge osmoregulatory effect of IGF-I on non-salmonids spe-

cies has only been tested in the anadromous percichthyid *M. saxatilis* [28] and the euryhaline cyprinodont *F. heteroclitus* [29]. In spite of the positive effects of GH treatment on hypoosmoregulation in striped bass, treatment of this species with recombinant bovine IGF-I (rbIGF-I) resulted in an osmoregulatory imbalance after transfer to SW that was greater than that of control fish. An unfavorable metabolic effect of rbIGF-I could explain this negative effect on SW acclimation [28].

In *F. heteroclitus* a single injection of rbIGF-I improved hypoosmotic capacity (decreased plasma osmolality and increased gill Na^+,K^+-ATPase activity) in a dose-dependent manner [29]. However, 10 days of rbIGF-I treatment, using a slow-release system (vegetable oil) did not improve hypoosmoregulatory capacity after transfer from BW to SW (Mancera and McCormick, unpublished results). A similar inability of long-term rbIGF-I treatment to duplicate the effects of oGH was found for *Salmo salar*, indicating that IGF-I may carry out only some of the osmoregulatory actions of GH [32].

IGF-II is another member of the insulin-like growth factor family present in the gill and other tissues [7,8]. Treatment of *F. heteroclitus* with recombinant human IGF-II (rhIGF-II) or with insulin had no effect on plasma osmolality and gill Na^+,K^+-ATPase activity after transfer from BW to SW [29]. The failure of this hormone to improve hypoosmoregulatory capacity suggests that rhIGF-II has no effect on monovalent ion secretion in *F. heteroclitus*. Additional experiment will be necessary, however, to confirm this hypothesis.

5. Interactions between GH, IGF-I and other endocrine factors

The pathway for the osmoregulatory effects of GH and IGF-I in non-salmonid teleost is not known. The available data about this possible pathway come mainly from salmonid species where several options have been proposed. GH could act by itself on osmoregulatory organs or, more likely and in accord with the somatomedin hypothesis, GH could stimulate IGF-I production in the liver and osmoregulatory organs and IGF-I could mediate some of the physiological actions of GH [31,45]. Interaction of GH and IGF-I with other endocrine systems such as cortisol and prolactin is also likely.

A cooperation between GH and IGF-I for increasing gill Na^+,K^+-ATPase activity and ion secretory capacity has been proposed [31,45]. *F. heteroclitus* treated simultaneously with oGH and rbIGF-I (a single injection over 3 days of treatment) showed higher gill Na^+,K^+-ATPase activity and better salinity tol-

erance compared to either hormone alone [29]. These results suggest a cooperation between GH and IGF-I and agree with the data observed in salmonid species. Compared to either hormone alone, 10 days of oGH plus rbIGF-I treatment using a slow-release system (vegetable oil) improved salinity tolerance after transfer from BW to SW but did not affect gill Na^+,K^+-ATPase activity (Mancera and McCormick, unpublished results).

The non-correlation between gill Na^+,K^+-ATPase activity and salinity tolerance after GH plus IGF-I treatment suggest an effect of these hormones on other osmoregulatory organs (e.g. kidney, intestine). In addition, an effect of GH and IGF-I on gill osmoregulatory function (e.g. chloride channel activity, co-transporter activity) other than gill Na^+,K^+-ATPase activity (e.g. chloride channel activity, co-transporter activity) also could explain this difference.

In addition to the GH/IGF-I axis other hormones also have hypoosmoregulatory activity and may interact with GH and IGF-I. Cortisol is the main corticosteroid released from the adrenal gland and has a well known role in promoting SW-acclimation in salmonids and non-salmonids [10,30,43] (see Ref. [31] for review). In salmonid species a positive interaction between GH and cortisol has been shown [25,32]. A possible mechanism for this interaction is the upregulation of gill cortisol receptors by GH reported in coho salmon (*Oncorhynchus kisutch*) [49]. In *O. mossambicus* a positive cooperation between cortisol and oGH was observed for increasing gill Na^+,K^+-ATPase activity (Mancera and Bern, unpublished data). In *F. heteroclitus*, however, this cooperation was not observed after 10 days of treatment using a slow-release system (Mancera and McCormick, unpublished data). In addition, *F. heteroclitus*, treated with oGH plus thriiodothyronine (T3) did not show increased gill Na^+,K^+-ATPase activity or improved salinity tolerance relative to either hormone alone (Mancera and McCormick, unpublished data).

In salmonid and non-salmonid species a GH receptor has been characterized in liver and other organs. In salmonids, the data on dynamics of GH receptors during adaptation to SW suggest a osmoregulatory role for this receptor [44]. In non-salmonid species there are no data concerning modifications of GH receptors in relation to osmoregulatory processes. In addition IGF-I receptor has been shown in several tissues of salmonids and non-salmonids fish (e.g. brain, liver, gastrointestinal tract, skeletal and heart muscle, gill and ovaries) [16,18,20,40]. However, further studies on expression of IGF-I receptor in osmoregulatory organs and hormonal control of this expression are necessary to understand the role of IGF-I in osmoregulation.

6. Conclusions and future research

The available data on the osmoregulatory role of the GH/IGF-I axis in teleosts come from a very small number of species (salmonids, *O. mossambicus*, *O. niloticus*, *M. saxatilis* and *F. heteroclitus*). The study of more species, with different osmoregulatory pattern, will be necessary to fully clarify the osmoregulatory role of the GH/IGF-I axis in non-salmonids species.

Nonetheless, the wide phylogenetic separation of these species indicates that the role of the GH/IGF axis on osmoregulation may be a relatively common feature of euryhalinity in teleosts. It would be of interest to examine non-teleosts fishes such as Acipenseriformes (sturgeon) and Petromyzontiformes (lamprey) to examine how early in vertebrate history the osmoregulatory function of GH evolved. In a recent analysis of the structural evolution of vertebrate GH Wallis [54] found that the changes in its primary structure were subject to 'bursts of rapid evolution' and suggested that these were related to osmoregulation and other physiological actions of GH, not related to growth regulation. Examination of the role of GH in osmoregulation in more teleost species may allow us to analyze the relationship between the structural and functional evolution of GH.

In addition to an evolutionary approach, we need a greater understanding of the mechanisms involved in the osmoregulatory function of GH and IGF-I. Unlike prolactin, the secretion of GH is apparently not altered by changes in plasma osmotic concentration, so the regulation of GH (and IGF-I) secretion in response to salinity changes is likely due to 'indirect' neuroendocrine factors. There is currently little information on changes in tissue receptors and binding proteins in teleost in response to salinity. We must resolve the relative roles of GH and IGF-I in osmoregulation and their interactions with other endocrine systems such as prolactin and cortisol in osmoregulation. Potential conflicts exist for action of the GH/IGF-I axis in regulating growth, metabolism and osmotic balance within an organism. Discovering how GH and IGF-I are able to carry out all of these functions will be an additional challenge to researchers in this field.

Acknowledgements

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