

# Distinct role of insulin and IGF-I and its receptors in white skeletal muscle during the compensatory growth of gilthead sea bream (*Sparus aurata*)

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## Abstract

In order to examine compensatory growth (CG) in gilthead sea bream (*Sparus aurata*), we used an experimental protocol involving 1-, 3- or 4-week fasting followed by a one- or two-month re-feeding period. After one month of re-feeding, final body weight gain and growth acceleration in groups fasted for 1 or 3 weeks (W1 and W3) were comparable to the control group values (C), indicating that partial CG was achieved. Fasting had a marked effect on depressing circulating glucose plasma levels, which were recovered to C group values after one month re-feeding. Glycogen content in liver and muscle decreased during fasting and was completely recovered after two month re-feeding. FFA plasma levels increased during fasting trial for all the different experimental groups, whereas lipid content in liver and muscle was depressed, indicating that lipid depot utilization contributes to the energy metabolism of gilthead sea bream. After re-feeding for 1 or 2 months, FFA and lipid content in liver and muscle were comparable to C group values. Plasma insulin and IGF-I values decreased during the fasting trial, while normal levels were restored after 1 month of re-feeding. IGF-I and insulin binding assays in semi-purifications of soluble proteins from white skeletal muscle showed that IGF-I binding increased with fasting while insulin binding decreased. However, the specific binding for these two peptides was comparable to C group values after re-feeding for 1 or 2 months.

In conclusion, after re-feeding insulin and IGF-I recovery was parallel with increases in weight gain and growth acceleration in those groups which showed diminutions in growth during the period of food restriction. Furthermore, the differential regulation of the specific binding for insulin and IGF-I in white skeletal muscle during fasting, indicates that these two receptors play distinct roles in response to the nutritional status of the animal.

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*Keywords:* Compensatory growth; Insulin; Insulin like growth factor I (IGF-I); White muscle; Hormonal regulation

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## 1. Introduction

The culturing of gilthead sea bream (*Sparus aurata*) has been widely improved in Southern Europe in the last

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decade. Several works reported the effects of dietary composition and ration size on growth efficiency in this species (Pérez-Sánchez and Le Bail, 1999; Metón et al., 2003). Nevertheless, studies on different fish species have shown that nutritional status provokes profound effects on the growth and development of somatic tissues, particularly in skeletal muscle (Navarro and Gutiérrez, 1995; Power et al., 2000; Chauvigné et al., 2003). Although information is available for salmonids, few studies have addressed the effects of food deprivation on the metabolism and the endocrine system of *Sparus aurata* (Pérez-Sánchez et al., 1994; Company et al., 2001; Power et al., 2000; Albalat et al., 2005).

The natural life cycle of many fish species includes a fasting period (Navarro and Gutiérrez, 1995). Moreover, compensatory growth (CG) of fish implies a readjustment of growth acceleration to minimize the discrepancy between achieved and desired growth, caused by a period of under-nutrition or unfavourable environmental conditions (Hubbell, 1971; Ali et al., 2003). From a practical point of view, this compensatory strategy is of great interest to the aquaculture industry because feeding programmes can be designed to improve growth rates while minimizing costs (Hayward et al., 1997). However, few studies in gilthead sea bream have reported data on the effects of fasting and subsequent re-feeding on morphologic parameters changes neither on glycogen or lipid body storage mobilization (Company et al., 1999; Power et al., 2000; Grigorakis and Alexis, 2005).

Fish liver is one of the first organs to be affected by food deprivation (Power et al., 2000), and plays a major role in glucose homeostasis. In gilthead sea bream, this organ exhibits a sensitive control to regulate glucose mobilization or production through cycling substrates and products in the glycolytic/gluconeogenic pathway (Metón et al., 2003). The maintenance of glycaemia during food deprivation is directly related to the capacity of hepatic glycogen mobilization, which occurs early in fasting (Love, 1970). Glucose homeostasis during fasting also depends on the subsequent activation of hepatic gluconeogenesis and the reduction in the rate of glucose utilization (Navarro and Gutiérrez, 1995). Information regarding metabolic changes under conditions of food deprivation in gilthead sea bream is also limited to skeletal muscle (Power et al., 2000; Grigorakis and Alexis, 2005; Sangiao-Alvarellos et al., 2005).

Circulating levels of growth hormone (GH) and insulin-like growth factors (IGFs) provide a useful tool to monitor growth performance and nutritional status (Pérez-Sánchez et al., 1994; Pérez-Sánchez and Le Bail,

1999). In the same manner, it is well established that insulin and thyroid hormone are strongly affected by nutrition (Navarro and Gutiérrez, 1995; Cerdà-Reverter et al., 1996; Power et al., 2000). Extended fasting normally causes a reduction in plasmatic insulin levels (Navarro and Gutiérrez, 1995), and a strong decrease in IGF-I plasma levels (Pérez-Sánchez and Le Bail, 1999).

To our knowledge, there is no information available on IGF-I or insulin binding in white skeletal muscle under a range of nutritional conditions in the gilthead sea bream. However, several authors have reported the physiological and endocrine mechanisms used by this species to recover from food deprivation by means of re-feeding. Power et al. (2000) studied changes in lipid and glycogen content in liver and the variations in thyroid hormone levels after fasting and re-feeding. More recently, Grigorakis and Alexis (2005) reported changes in muscle glycogen and lipid content during fasting in this species, and interestingly, Sangiao-Alvarellos et al. (2005) reported changes on carbohydrate metabolism in several tissues of gilthead sea bream after 14 days fasting. Whereas the endocrine response to feed deprivation has been well studied, the subsequent CG response remains poorly understood in teleost and this is especially evident for species such as the gilthead sea bream. Nevertheless, although salmonids show distinct degrees of CG mechanisms, little is known about those underlying CG in terms of endocrine responses (Miglav and Jobling, 1989; Quinton and Blake, 1990; Jobling and Koskela, 1996; Bilton and Robins, 1998; Damsgård and Dill, 1998).

Due to the importance of understanding the metabolic and endocrine mechanisms underlying the compensatory growth strategy in fish, in the present work we studied the variations in circulating concentrations of glucose and FFA together with the mobilization of glycogen and lipid body energy reserves during this process. We investigated changes on plasma circulating insulin and IGF-I peptides, and for the first time in this species, we reported data of receptor binding in white skeletal muscle of juvenile gilthead sea bream, a Mediterranean teleost of high commercial value in Europe.

## 2. Materials and methods

### 2.1. Animals and experimental conditions

Three hundred and fifty sexually immature gilthead sea breams (*Sparus aurata*) of 3 month age were acclimatized to laboratory conditions at the Aquaculture Centre-IRTA (Sant Carles de la Ràpita, Tarragona,

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