Data in Brief 8 (2016) 404–410

Contents lists available at ScienceDirect

Data in Brief

journal homepage: www.elsevier.com/locate/dib

Data Article

Data on the characterization of follicle-stimulating hormone monoclonal antibodies and localization in Japanese eel pituitary

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ARTICLE INFO

Article history:
Received 11 May 2016
Received in revised form 20 May 2016
Accepted 27 May 2016
Available online 3 June 2016

Keywords:
Japanese eel
FSH
Monoclonal Antibody

ABSTRACT

Monoclonal antibodies were generated against recombinant follicle-stimulating hormone (rec-FSH) from Japanese eel *Anguilla japonica*; rec-FSH was produced in *Escherichia coli* and purified using Ni-NTA Sepharose column chromatography.

In support of our recent publication, "Production and characterization of monoclonal antibodies against recombinant tethered follicle-stimulating hormone from Japanese eel *Anguilla japonica*" [1], it was important to characterize the specificity of eel follicle-stimulating hormone antibodies. Here, the production and ELISA system of these monoclonal antibodies are presented. The affinity-purified monoclonal antibodies specifically detected eel rec-FSH in ELISA and on western blots of rec-FSH produced from CHO cells. Immunohistochemical analysis revealed that FSH staining was specifically localized in the eel pituitary.

DOI of original article: http://dx.doi.org/10.1016/j.ygcen.2016.04.030
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http://dx.doi.org/10.1016/j.dib.2016.05.069
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Specifications Table

Subject area	*Biology*
More specific subject area	*Eel FSH antibody*
Type of data	*Figures, graphs, tables and Western blots*
How data was acquired	*ELISA, Western blotting and immunohistochemistry*
Data format	*Analyzed*
Experimental factors	*Immunization of mice with rec-eel FSHβ/α, antibody purification and isotype determination*
Experimental features	*Characterization of monoclonal antibody and ELISA analysis using purified antibody, western blotting and confocal microscopy to determine the localization of FSH in the pituitary*
Data source location	*Anseong and Busan, Korea*
Data accessibility	*Data presented in this article*

Value of the data

- The antibody generated can serve as a tool for basic research in the field of eel FSH biology.
- FSH localization in the pituitary suggests a potential FSH role during oocyte- maturation.
- ELSIA system can analyze the quantity of rec-eel FSH hormone and be used in investigations in reproductive endocrinology *in vitro* and *in vivo*.

1. Data

The pRSET expression vector encoding a putative protein containing 220 amino acids was constructed (Fig. 1A). The protein in *Escherichia coli* was purified using a 1st Ni-NTA Sepharose column and a 2nd Sepharose column (Fig. 1B,C). After mice were immunized with the antigen, the supernatants of the hybridoma cells were analyzed by using indirect ELISA (Fig. 2). To establish a sandwich-ELISA system, the intersection method was used with HRP-labeled antibodies (Table 1). The quantities of rec-FSHβ/α and luteinizing hormone (LH) β/α and the selected stable cell lines, and western blot result were described **(**Fig. 3**).** The FSHβ-subunit antibody (eFB-C14) was used for examining FSH localization in the pituitary during oocyte maturation **(**Fig. 4**).**

2. Experimental design, materials and methods

2.1. Experimental design

A cDNA encoding eel FSHβ/α was cloned into the vector pRSET, one *E. coli* strain expressing FSHβ/α was selected and cultivated in large-volume cultures. The protein was purified and immunized, and then spleen cells were fused with Sp2/0 cells. Subsequently, hybridoma cells were selected. The reactivity of the culture supernatant was tested using indirect ELISA, and the antibodies were purified using Hi-Trap Protein G columns. The antibodies were tested for specificity by performing sandwich-ELISA analysis, western blotting, and immunohistochemical analysis.

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