

Japan-US Brain Research Cooperation Program

Group Joint Study Report

[field: emotionality & memory]

1. The Representative of Group Joint Study:

Yasuo Sakuma, M.D., Ph.D., Professor, Nippon Medical School

2. Project Title:

Conditional knock-out of GnRH neurons and its effects on development, emotionality and behavior in mice

3. Japanese Investigator's Name, Title, Affiliation and Phone Number:

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4. U.S. Investigator's Name, Title, and Affiliation:

Chief: Donald W. Pfaff, Professor, The Rockefeller University

Collaborator: Sonoko Ogawa, Associate Professor, The Rockefeller University

5. The Term of Research: From 2000/4/1 To 2003/3/31 (3 Years)

6. Abstract, Result and Significance of Research (300 Words):

Detection of the odor of receptive females is a key step for the onset of male sexual behaviors. Previously, we have reported that estrogen receptor beta knockout (bERKO) mice exhibited normal levels of male sexual behaviors such as mounts and intromissions (Ogawa et al., 1999). However, it is not known whether they show any behavioral alterations in terms of sexual odor discrimination and preference. We tested gonadally intact male bERKO and wild type (WT) mice in a three-chamber apparatus for nose poking toward the odors of sexually receptive female mouse and gonadally intact male mouse. Odor preference indexes (PI) as responses toward the female odor over the male odor were calculated for each mouse in terms of cumulative duration (PID) and frequency (PIF) of nose poking during 20min test sessions. Each mouse was tested twice (one week apart) and between the two tests, a half of mice from each genotype were exposed overnight in their home cage to a sexually receptive female mouse through a perforated transparent plexiglass board. In the first test, both genotypes of mice showed similar levels of preference toward the female odor. However, overnight exposure to receptive female mice affected WT and bERKO mice differently. The two groups of WT mice showed similar preference toward the female odor in both tests. In bERKO mice, overnight exposure increased PIF and PID, while non-exposed males showed decreased preference to the female odor in the second tests. These findings suggest that ER- β activation may be responsible for fine-tuning of social recognition in male mice.

7. The Others (Practical Issues, Special Mention Matters): NA

参考資料 1

第 32 回北米神経科学学会 (2002) 抄録

Program Number: 482.15 Day / Time: Tuesday, Nov. 5, 10:00 AM - 11:00 AM

FACILITATION OF MOUNT BEHAVIOR IN FEMALE RATS BY CAUDAL DEEFFERENTATION OF THE VENTROMEDIAL HYPOTHALAMUS

H.Ohnishi; Y.Kondo*; Y.Sakuma

Dept. of Physiology, Nippon Medical School, Tokyo, Japan

Exposure to sex steroids at the early stage produces sex differences, both functional and morphological, of mammalian brain. Testosterone makes brain masculinized (and defeminized), whereas a lack of sex steroids makes brain feminized. However, it has been reported that a destruction of the ventromedial hypothalamus (VMH) induced mount, a typical male sexual behavior, in female rats when ovariectomized and treated with testosterone. This suggests that female rat brain possesses the neural circuit for expressing male sexual behavior, and also has the inhibitory system for male sexual behavior, presumably in the VMH. In the present experiment, we examined the effect of a caudal defferentation of the VMH on sexual behavior and partner preference in female rats. Ovariectomized Wistar rats were subjected to a coronal transection caudally located in the VMH with a razor blade (1 mm or 2 mm in width). After recovery, the partner preference, female sexual behavior and male sexual behavior were tested after a single injection of 2 g estradiol benzoate 48 hrs prior to the testing. The transection significantly enhanced mount behavior when placed with highly receptive females, while no effect was found in female sexual behavior, lordosis and soliciting behavior. It was also ineffective on the partner preference which is indicated by time spent to explore sexually active male odors over estrous female odors. The results indicate that the VMH may exert an inhibitory influence on neural circuits regulating masculine behavior, but not partner preference. After the disinhibition, a low dose of estrogen is sufficient to induce mount behavior in female rats. Citation: H.Ohnishi, Y.Kondo, Y.Sakuma. FACILITATION OF MOUNT BEHAVIOR IN FEMALE RATS BY CAUDAL DEEFFERENTATION OF THE VENTROMEDIAL HYPOTHALAMUS. Program No. 482.15. 2002 Abstract Viewer/Itinerary Planner. Washington, DC: Society for Neuroscience, 2002. CD-ROM

Program Number: 288.6 Day / Time: Monday, Nov. 4, 9:00 AM - 10:00 AM

SPECIES DIFFERENCES AND EFFECTS OF ESTROGEN RECEPTOR (Er)-alpha GENE DISRUPTION IN SEXUALLY DIMORPHIC EXPRESSION OF Er-beta IN THE PREOPTIC AREA

Y.Sakuma1*; Y.Nanasaki2; M.Nomura2; C.Orikasa1; D.W.Pfaff2; S.Ogawa2

1. Dept Physiology I, Nippon Medical School, Tokyo, Japan; 2. Lab. Neurobiology and Behavior, The Rockefeller University, New York, NY, USA

Female rats have a significantly larger number of estrogen receptor (ER)-beta positive cells than males in the anterior medial preoptic area (MPOA), particularly in the medial-most portion of the anteroventral periventricular nucleus (AVPV; Orikasa et al., 2002). In the present study, we examined whether a similar sex difference in the expression of ER-beta mRNA and protein might be detected in the mouse brain and whether it might be affected by ER-alpha gene disruption. In situ hybridization histochemistry (ISHH) revealed that unlike in rats, ER-beta gene expression in the MPOA was significantly higher in gonadectomized male mice than in females ($p < 0.05$). Furthermore, in the MPOA, ER-alpha knockout male mice had significantly reduced levels of ER-beta mRNA ($p < 0.05$ vs wild type). Consistent with the ISHH results, male mice had significantly higher number of ER-beta immunoreactive (IR) cells compared to female mice in the MPOA ($p < 0.01$), whereas in the ventromedial nucleus of the hypothalamus, an opposite sex difference was found ($p < 0.01$). Detailed immunocytochemical analyses performed in parallel in gonadectomized rats and mice revealed that the number of ER-beta IR cells was significantly higher in female rats compared to male rats ($p < 0.01$) not only in the AVPV but also throughout the anterior MPOA. In mice, there was no apparent localization of ER-beta IR cells in the AVPV, regardless of sex. These findings suggest that there is a clear species difference in the sexually dimorphic expression of ER-beta in the MPOA, which may be modified by ER-alpha activation in mice. Supported by: MH 62147 to SO Citation: Y.Sakuma, Y.Nanasaki, M.Nomura, C.Orikasa, D.W.Pfaff, S.Ogawa. SPECIES DIFFERENCES AND EFFECTS OF ESTROGEN RECEPTOR (Er)-alpha GENE DISRUPTION IN SEXUALLY DIMORPHIC EXPRESSION OF Er-beta IN THE PREOPTIC AREA. Program No. 288.6. 2002 Abstract Viewer/Itinerary Planner. Washington, DC: Society for Neuroscience, 2002. CD-ROM.

Program Number: 383.1 Day / Time: Monday, Nov. 4, 1:00 PM - 2:00 PM

EFFECTS OF ESTROGEN RECEPTOR-beta GENE DISRUPTION ON SEXUAL ODOR PREFERENCE IN MALE MICE

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1. Lab. Neurobiol. & Behav., The Rockefeller Univ., New York, NY, USA; 2. Dept. Psychol., Univ. of Kagoshima,

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Detection of the odor of receptive females is a key step for the onset of male sexual behaviors. Previously, we have reported that estrogen receptor beta knockout (bERKO) mice exhibited normal levels of male sexual behaviors such as mounts and intromissions (Ogawa et al., 1999). However, it is not known whether they show any behavioral alterations in terms of sexual odor discrimination and preference. We tested gonadally intact male bERKO and wild type (WT) mice in a three-chamber apparatus for nose poking toward the odors of sexually receptive female mouse and gonadally intact male mouse. Odor preference indexes (PI) as responses toward the female odor over the male odor were calculated for each mouse in terms of cumulative duration (PID) and frequency (PIF) of nose poking during 20min test sessions. Each mouse was tested twice (one week apart) and between the two tests, a half of mice from each genotype were exposed overnight in their home cage to a sexually receptive female mouse through a perforated transparent plexiglass board. In the first test, both genotypes of mice showed similar levels of preference toward the female odor. However, overnight exposure to receptive female mice affected WT and bERKO mice differently. The two groups of WT mice showed similar preference toward the female odor in both tests. In bERKO mice, overnight exposure increased PIF and PID, while non-exposed males showed decreased preference to the female odor in the second tests. These findings suggest that ER- β activation may be responsible for fine-tuning of social recognition in male mice. Supported by: MH 62147 to SO Citation: Y.Nanasaki, K.Tomihara, D.W.Pfaff, S.Ogawa. EFFECTS OF ESTROGEN RECEPTOR-beta GENE DISRUPTION ON SEXUAL ODOR PREFERENCE IN MALE MICE. Program No. 383.1. 2002 Abstract Viewer/Itinerary Planner. Washington, DC: Society for Neuroscience, 2002. CD-ROM.

Program Number: 383.2 Day / Time: Monday, Nov. 4, 2:00 PM - 3:00 PM

SEXUAL ODOR PREFERENCE IN FEMALE WILD - TYPE AND ESTROGEN RECEPTOR KNOCKOUT MICE.

K.Tomihara^{1*}; Y.Nanasaki²; D.W.Pfaff²; S.Ogawa²

1. Dept. Psychol., Kagoshima Univ, Kagoshima, Japan; 2. Lab. Neurobiol. & Behav., Rockefeller Univ., New York, NY, USA

We have previously demonstrated that female mice showed significantly higher lordosis quotient when they approached to a male mouse compared to the cases when the male mouse approached them (Tomihara and Makino, 1991). This finding suggests male olfactory cues may be important when female mice choose mating partners. In the present study, we examined the roles of two types of estrogen receptors (ER), alpha and beta, in the odor preference between intact and castrated male mice using a three-chamber apparatus, which was designed to deliver two kinds of odors separately. We found that unlike its strongly destructive effects on lordosis, ER-alpha gene disruption did not affect odor preference. Gonadally intact ER-alpha knockout (aERKO) and wild type (aWT) mice equally showed a strong preference toward castrated male odors as measured as frequency and duration of nose poking during 20min tests. Priming with estradiol benzoate (10ug/0.1ml) and progesterone (500ug/0.1ml) in gonadectomized mice reduced preference toward castrated male odors ($p < 0.01$ vs oil treated control) in both aERKO and aWT mice. Preference toward castrated male odors over intact male odors was also observed in gonadectomized ER-beta knockout (bERKO) and their WT (bWT) control mice. However, steroid priming did not significantly affect the odor preference in bERKO mice, although it tended to decrease preference toward castrated male odors in bWT mice. These results suggest that ER-alpha activation may be crucial for the expression of lordosis but not for sexual odor preference by female mice. Supported by: MH 62147 to SO. Citation: K.Tomihara, Y.Nanasaki, D.W.Pfaff, S.Ogawa. SEXUAL ODOR PREFERENCE IN FEMALE WILD-TYPE AND ESTROGEN RECEPTOR KNOCKOUT MICE. Program No. 383.2. 2002 Abstract Viewer/Itinerary Planner. Washington, DC: Society for Neuroscience, 2002. CD-ROM.

参考資料 2

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26 February 2003

Ms E2K3-825

"Estrogen-induced vigorous mounting in female rats carrying hypothalamic knife cuts"

By H. Ohnishi, Y. Kondo, Y. Sakuma

Received in Edinburgh Office: 7th January 2003

Dear Dr Kondo,

Your paper was sent for review to two independent expert referees whose reports I enclose. I am pleased to inform you that both referees complement the authors on an interesting study and a very clearly presented paper. I have read your manuscript myself with interest and enclose an annotated copy with minor modifications to improve readability and to conform to Journal style.

In your resubmission please pay careful attention to the 'Instructions to Authors' and "Style Notes" (<http://www.blackwell-science.com/jne>). I would be pleased if you could send three copies of a revised manuscript, together with a letter explaining how you have responded to each of the points raised by the referees. Could I also ask you to complete the enclosed copyright declaration form, and to enclose a disk copy of your revised manuscript, with a completed file description form. I must ask that you put the date of your final revision on the cover page of the disk version of your revised manuscript. If the top copy of your final manuscript does not bear the same date as that on the disk version, then the publishers will assume that the disk version is incorrect and will ask you to provide a new disk that does match the accepted copy, with a likely delay in publication.

Thank you for submitting your work to The Journal, and I hope that we may receive further contributions from you in the future.

Yours sincerely

Professor Gareth Leng
Editor-in-Chief