peduncle of the first antenna, the compact body, and the smooth cuticle and lanceolate uropods (Fig. 1, C, B, and D, respectively) are satisfactory evidence (7). No doubt many undescribed Lysianassidae still remain in the deep sea, which makes closer identification impossible. However, this species apparently lacks the strongly ridged dorsal surface of the common bathyal and abyssal Eurythenes gryllus [figure 132a in (7)] and has some features, such as the shape of the basis of the last pereiopod, the dorsal surface of the urosome and pleon, and the telson (visible in Fig. 1, D, B, and E, respectively), similar to those of Alicella gigantea (3).

Evidence of the habits of these large amphipods is scanty. In general, Lysianassidae are ubiquitous, strongly swimming carnivores or omnivores. Eurythenes gryllus has diffuse eyes (3,4) occupying a large part of the side of the head. The adults are red in color (5) and have oily bodies. These features suggest a bathypelagic life, although "mineral particles" have been found in the gut (4), and specimens have been collected from such diverse sources as benthic fish traps and the stomach (3) of a sea bird (the fulmar, Fulmarus glacialis), which indicate that this species may feed from the surface of the sea (probably at night) to the abyssal bottom on occasion. Alicella gigantea has large eyes (3), and our species probably has extensive ocular areas (see Fig. 1C); both have whitish or light-colored bodies like obligate benthic species. It is likely that both swim extensively but spend more time on the bottom than Eurythenes gryllus.

These large Lysianassidae appear to be versatile omnivores and scavengers, swimming through a great depth range and having adaptations for reduced specific gravity (for example, the oily body of Eurythenes gryllus) and photoreception in the photic zone, but retaining features, such as the compact bodies, relatively short simple pereiopods, antennae, and uropods, necessary for opportunistic scavenging and carnivory on the bottom. The features of Eurythenes gryllus indicate that its primary habitat is the bathyal region of the water; Alicella and our species probably spend much of their time on or near the bottom but venture occasionally into the photic zone.

Chevreux (3) commented on the difficulty or impossibility of obtaining large deep-sea Lysianassidae by trawl-

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ing or dredging. His specimens of *Eurythenes gryllus* and *Alicella gigantea* were taken from large fish traps containing smaller traps, so in many ways his method of collection was similar to our bait can method. The apparent alacrity with which these scarce, mobile creatures approach the bait suggests that trapping of the attracted animals may be a potent technique for obtaining otherwise uncatchable organisms in the deep sea.

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References and Notes

- 1. M. H. Sessions, J. D. Isaacs, R. A. Schwartzlose, "A camera system for the observation of deep-sea marine life," paper presented at the Underwater Photo-Optical Instrumentation Applications Seminar in San Diego, Calif., 1968.
- 2. L. A. Zenkevich, Ed., Biology of the Pacific Ocean, vol. 2, The Deep-Sea Bottom Fauna, Pleuston (Nauka, Moscow, 1969).
- 3. E. Chevreux, Bull. Soc. Zool. Fr. 24, 152 (1889); Result. Camp. Sci. Prince Albert I 90, 42 (1935).
- 4. J. L. Barnard, Galathea Rep. 5, 23 (1961).
- 5. J. A. Birstein and M. E. Vingradov, Tr. Inst. Okeanol. Akad. Nauk SSSR 12, 210 (1955).
- 6. E. Dahl, Galathea Rep. 1, 211 (1959).
- 7. For a complete diagnosis and figures of Lysianassidae see J. L. Barnard, U.S. Nat. Mus. Bull. 271 (1969), pp. 294–295 and figures 117–132.
- 8. Supported by NSF grant GB 14488 (to R.R.H.), AEC contract AT (11-1)-34, project 127 (to J.D.I.), and National Research Council of Canada grant A-1900 (to E.L.M.). We thank Dr. J. L. Barnard, Division of Crustacea, Smithsonian Institution, for helpful comments.

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Ovulation in Hamster: Induction by β Subunit of Ovine Interstitial Cell Stimulating Hormone

Abstract. As little as 5 micrograms of interstitial cell stimulating hormone (ICSH) or 20 micrograms of ICSH- β is effective for the induction of ovulation in 100 percent of hamsters treated at 0500 hours on day 4 after lordosis, whereas as much as 800 micrograms of ICSH- α is ineffective. Both ICSH and ICSH- β are also effective for induction of ovulation in hypophysectomized animals. Thus, the ovulation-inducing activity of the ICSH molecule resides in its β subunit.

Pituitary interstitial cell stimulating hormone [ICSH or luteinizing hormone (LH)] induces ovulation in the rat (1)and hamster (2). Highly purified ICSH isolated from sheep pituitary glands (3)is composed of two chemically dissimilar subunits (4, 5), and the primary structures of both subunits (ICSH- α and ICSH- β) have recently been elucidated (6). We now report the ovulation-inducing activity of ICSH, ICSH- α , and ICSH- β in both normal and hypophysectomized hamsters.

Mature female golden hamsters (Mes-

Table 1. Induction of ovulation in the intact hamster by a single intraperitoneal injection of ovine-ICSH or its subunits at proestrous stage at 0500 on day 4 after lordosis. Animals were examined for the number of ruptured follicles and newly ovulated tubal eggs 17 hours later. The hormone was administered in 0.2 ml of water; S.E., standard error.

| Single dose (µg/hamster) | Animals | | | Duntund | |
|-----------------------------|------------------|-----------------------------------|---------------------------|--------------------------------|---------------------------|
| | Treated (No.) | With ruptured follicles (%) | With tubal eggs (%) | follicles (Mean \pm S.E.) | Tubal eggs (Mean±S.E.) |
| | | I | CSH | | |
| 1.25 | 9 | 0 | 0 | 0 | 0 |
| 2.5 | 9 | 6 (67) | 5 (56) | 3.1 ± 1.0 | 2.6 ± 1.0 |
| 5.0 | 9 | 9 (100) | 9 (100) | 8.0 ± 0.8 | 8.0 ± 0.8 |
| | | IC | SH-a | | |
| 8.0 | 9 | 0 | 0 | 0 | 0 |
| 80.0 | 10 | 0 | 0 | 0 | 0 |
| 800.0 | 9 | 0 | 0 | 0 | 0 |
| | | IC | SH-B | | |
| 2.5 | 9 | 0 | 0 | 0 | 0 |
| 5.0 | 9 | 1 (11) | 1 (11) | 0.4 ± 0.4 | 0.2 ± 0.2 |
| 10.0 | 13 | 9 (69) | 9 (69) | 5.9 ± 1.1 | 4.6 ± 1.0 |
| 20.0 | 10 | 10 (100) | 10 (100) | 8.7 ± 0.8 | 8.7 ± 0.8 |
| 40.0 | 5 | 5 (100) | 5 (100) | 9.6 ± 0.5 | 9.6 ± 0.5 |
| | | Pro | lactin | | |
| 800.0 | 7 | 0 | 0 | 0 | 0 |
| | | | H,O | | |
| 0.0 | 8 | 0 | ~0 | 0 | 0 |

Table 2. Induction of ovulation in the hypophysectomized hamster by a single intraperitoneal injection of ovine ICSH or its *B* subunit immediately after the hypophysectomy at proestrous stage between 0400 to 0600 hours on day 4 after lordosis. Animals were examined for the number of ruptured follicles and newly ovulated tubal eggs 17 hours later. The hormone was administered in 0.2 ml of water; S.E., standard error.

| Single dose (µg/hamster) | Animals | | | ~ | |
|--|---------------|-----------------------------------|---------------------------|--|-----------------------------|
| | Treated (No.) | With ruptured follicles (%) | With tubal eggs (%) | Ruptured follicles (Mean ± S.E.) | Tubal eggs (Mean ± S.E.) |
| | | IC | CSH | | |
| 10 | 10 | 5 (50) | 5 (50) | 4.5 ± 1.4 | 4.3 ± 1.0 |
| 20 | 8 | 8 (100) | 8 (100) | 8.6 ± 1.4 | 8.6 ± 1.4 |
| | | IC | SH-B | | |
| 10 | 8 | 0 | Ó | 0 | 0 |
| 20 | 8 | 2 (25) | 2 (25) | 1.2 ± 1.0 | 1.1 ± 1.0 |
| 40 | 6 | 5 (83) | 5 (83) | 6.7 ± 1.4 | 6.5 ± 1.3 |
| 80 | 9 | 9 (100) | 9 (100) | 8.9 ± 0.6 | 8.9 ± 0.6 |
| | | h | I2O | | |
| 0 | 7 | 0 | 0 | 0 | 0 |
| ************************************** | | | | | |

ocricetus auratus) weighing from 90 to 130 g (8 to 12 weeks old) were kept in a 10.5-hour light and 13.5-hour dark schedule with the light on from 0730 to 1800 hours. Females with the characteristic vaginal discharge that occurs after ovulation (7) were selected each morning, and thereafter were segregated into four groups so that they could be used for testing on the first, second, third, and fourth day after lordosis; (these groups were designated PLD-1, PLD-2, PLD-3, and PLD-4, respectively). For examination of ovulation-inducing activity, only those females that showed more than three consecutive 4-day cycles were used. Ovine ICSH was isolated by the method of Papkoff et al. (8), and its subunits were isolated by procedures described previously (4). A single injection of various doses of ICSH, ICSH- α , or ICSH- β was given intraperitoneally to intact females at 0500 hours on PLD-4 (9) with human chorionic gonadotropin (HCG). Similar treatments were also performed in hypophysectomized animals immediately after the operation between 0400 and 0600 hours on animals of the PLD-4 group. Hypophysectomy was performed by a parapharyngeal approach after a tracheotomy under intraperitoneal injection of Nembutal for anesthesia. All treated animals were killed and autopsied 17 hours after the injection. The ovaries were separated from the periovarian sacs, and newly ovulated eggs were taken from the punctured Fallopian tubes and examined with reflected light (i) for the number of ruptured follicles and (ii) with transparent light under a stereomicroscope for the number of eggs. The result obtained by counting of ruptured follicles agreed with that obtained by numeration of tubal eggs. The minimum effective doses of ICSH and ICSH- β for in-

duction of ovulation in 100 percent of the treated intact animals were 5 μg and 20 µg, respectively (Table 1); whereas the minimum doses for the hypophysectomized females were 20 μ g and 80 μ g, respectively (Table 2). Although a dose of 10 μ g of ICSH- β was effective for induction of ovulation, a considerably higher dose (800 μ g) of ICSH- α was unable to induce ovulation in the hamster. In the controls, the injection of either ovine prolactin (10) (adjusted to pH 8.3 with sodium hydroxide) or water was not effective for induction of ovulation. As a control of spontaneous ovulation that occurred physiologically in this lighting schedule, 100 females were examined for the onset of lordosis and the commencement of ovulation after adaptation for three estrous cycles in this laboratory environment. Females on PLD-4 were put into the same cage with the active males from the early afternoon to the late evening for observation. None of the tested animals showed lordosis to the advent of males before 1730 hours. Thereafter, 18, 33, 23, 17, and 9 percent of the observed females began to manifest the characteristic sexual response during the following time intervals: 1730 to 1800, 1800 to 1830, 1830 to 1900, 1900 to 1930, and 1930 to 2100 hours, respectively. When those tested animals were killed randomly at 7-hour intervals from 2300 to 0300 of the following morning, none of 20 females had either tubal eggs or ruptured follicles at 2300 hours. However, 5, 20, 60, and 95 percent of the animals of each group (20 females) were ovulated by the time of examination at 2400, 0100, 0200, and 0300 hours, respectively.

Since the interval between the treatment of HCG and the onset of ovulation was about 13 hours (9), it may be expected that the pituitary secretion of ICSH for spontaneous ovulation in this lighting schedule may have started as early as 1100 hours in some animals. In view of the average 8-hour interval between the onset of lordosis and the beginning of ovulation (9, 11), and also in view of the result of autopsy in the control animals, it is unlikely that spontaneous ovulation could occur before 2200 hours, the time set for the examination of induced ovulation in these experiments. Thus, it is evident that the β subunit of ICSH was potent in inducing ovulation. The fact that ICSH and ICSH- β were both effective for induction of ovulation in either intact or hypophysectomized animals may be taken to show that the mode of action of both preparations is mediated by their direct effect on the ovary. Since the biological activity of ICSH- α and ICSH- β is negligible in comparison with that of the ICSH molecule in the ovarian ascorbic acid depletion test (12), it is significant that a marked difference in ovulation-inducing activity of ICSH- α and ICSH- β has been demonstrated in experiments herein reported. Thus, the ovulation-inducing activity of the ICSH molecule is a function of its β subunit.

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References and Notes

- References and Notes
 A. J. Lostroh and R. E. Johnson, Endocrinology 79, 991 (1966); S.-Y. Ying and R. O. Greep, ibid. 89, 294 (1971).
 G. S. Greenwald, ibid. 88, 671 (1971).
 P. G. Squire and C. H. Li, Science 127, 32 (1958); J. Biol. Chem. 234, 520 (1959); D. N. Ward, R. F. McGregor, A. C. Griffin, Biochim. Biophys. Acta 32, 305 (1959).
 H. Papkoff and T. S. A. Samy, Biochim. Biophys. Acta 147, 175 (1967).
 C. H. Li and B. Starman, Nature 202, 291 (1964); D. N. Ward, M. Fujino, W. M. Lamkin, Fed. Proc. 25, 348 (1966); P. de la Llosa, C. Courte, M. Jutisz, Biochem. Biophys. Res. Commun. 26, 411 (1967).
 W.-K. Liu, C. M. Sweeney, H. S. Nahm, G. N. Holcomb, D. N. Ward, Res. Commun. Chem. Pathol. Pharmacol. 1, 463 (1970); H. Papkoff, M. R. Sairam, C. H. Li, J. Amer. Chem. Soc. 93, 1531 (1971); W.-K. Liu, H. S. Nahm, C. M. Sweeney, H. N. Baker, W. M. Lamkin, D. N. Ward, Res. Commun. Chem. Pathol. Pharmacol. 2, 168 (1971).
 R. Deanesley, Proc. Zool. Soc. Ser. A 108, 31 (1938).
 H. Papkoff, D. Gospodarowicz, C. H. Li
- (1938).
- (1938).
 H. Papkoff, D. Gospodarowicz, C. H. Li, Arch. Biochem. Biophys. 111, 431 (1965).
 W. H. Yang, Endocrinology 89, 287 (1971).
 C. H. Li, J. S. Dixon, T.-B. Lo, K. D. Schmidt, Y. A. Pankov, Arch. Biochem. Biophys. 141, 705 (1970).
 H. B. Warray, B. Yanginachi, M. C. Chang 10. C.
- Biophys. 141, 705 (1970).
 E. B. Harvey, R. Yanagimachi, M. C. Chang, J. Exp. Zool. 146, 231 (1961).
 H. Papkoff, J. Solis-Wallckermann, M. Martin, C. H. Li, Arch. Biochem. Biophys. 143, 236 (1971) tin, C. H. 226 (1971).
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