The Table shows that formation of labelled thyroxine gradually increased proportionately from 90 to 120 min and further increase in the labelled thyroxine synthesis was not observed in 130 min incubated sample. When the amount of head kidney soluble protein was increased to double and incubated for the same periods, the quantity of thyroxine formed in almost double quantity (Table), which indicates that formation of thyroxine is an enzymatically catalyzed process. This is again evident from the fact that when head kidney soluble supernatant was boiled at 100 °C (in a water bath, for 10 min), cooled and then incubated, no visible band was observed at the region of thyroxine and DIT. Besides, the quantity of head kidney soluble protein is a very important factor. When 1 mg of soluble supernatant was incubated for 90 min, no visible band corresponding to thyroxine marker was observed. Thus, the minimum requirement of head kidney soluble fraction to observe thyroxine formation at 90 min incubated sample was 1.5 mg/3.0 ml of incubation mixture.

Formation of thyroxine by head kidney soluble supernatant

Head kidney soluble supernatant (mg)	Incubation time (min)	^{131}I incorporated into thyroxine (%)
1.5	90	0.6
	100	0.8
	110	1.1
	120	1.4
	130	1.4
3.0	90	1.2
	100	1.5
	110	2.1
	120	2.7
	130	2.7

As thyroid follicular structure are present in the head kidney of teleost fishes[4–8], and as the head kidney soluble supernatant of this teleost fish possesses a peroxidase which significantly oxidizes the iodide into triiodide[1], the present observation of the formation of thyroxine by the same fish and by the same fraction, give further support to the possible role of teleost fish head kidney in the biosynthesis of thyroid hormone.

Zusammenfassung. In der löslichen überstehenden Fraktion (105,000 × *g*) der Kopfniere des Knochenfisches *Anabas testudineus* wurde ein Enzym nachgewiesen, welches die Synthese des Thyroxins katalysiert. Die Menge des löslichen Nierenproteins und die Inkubationszeit stellen zwei limitierende Faktoren für die in vitro-Synthese des Thyroxins dar.

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Juvenile Hormone Promotes Dominance Behavior and Ovarian Development in Social Wasps (*Polistes annularis*)

The females of a *Polistes* (Vespidae) colony have a dominance hierarchy organized with the queen (as the preeminent aggressor and egg-layer) in the α-position and the workers arranged linearly beneath her[1,2]. Even though all females are very similar externally[3], there is striking differential ovarian development which is correlated with position in the dominance hierarchy[2]. However, wasps of a colony retain their dominance status after being ovariectomized[4].

Many studies have shown the effects of gonadal hormones on vertebrate aggression and dominance hierarchies[5]. Juvenile hormone (JH), the endocrine product of the corpora allata, is the gonadotropin of most female insects[6] and topical application of JH can promote ovarian growth in *Polistes*[7]. JH is also known to affect mating behavior and pheromone production in many insects[8] and to alter caste structure and the time course of specific labors in honeybees[9]. However, this is the first report of an insect hormone affecting aggression or dominance interactions.

Colonies of *Polistes annularis* were collected in late May from one locality on Lake Travis near Austin, Texas. Each of the 16 nests was glued in the corner of a 15 cm × 15 cm plexiglass and screen observation cage. Nests were chilled at 4 °C prior to manipulation of the wasps. Wasps were provided with live caterpillars and water ad libitum during the experiment. On the day after capture the queens were determined by observers (R.H., T.K. and L.J.L.) according to behavioral criteria[1,2]. The next day 4 workers/cage were given a color-coded marking according to the treatment they were to receive and surplus workers were removed. Wasps ecdysing during the experiment were designated 'newly-emerged' and left in the cage. At noon daily for 4 days the cages were chilled and 10 µl of acetone containing 20 µg test substance (or oil) were applied to the abdomens of 3 workers/cage. The identities of the test substances were not revealed to the observers, but included JH III, 3 JH analogs, and 3 identical solutions of olive oil. Each cage was treated with 2 different hormones and oil. The JH and analogs were a gift of Zoecon Corp., Palo Alto, Cal., and were 90% pure, 80% of which was the active *trans, trans, cis*-isomer. On the 6th day wasps were sacrificed and the length of the 3 largest oocytes determined with an ocular micrometer. Additional colonies were established later as controls under the same conditions except that there were either no treatments or only acetone and oil were administered (to 3 wasps/cage). Behavioral observations of the interactions between colony members were made each day 2–3 h after treatment and on the day of sacrifice. From these observations the following system of ranking

[1] M. J. W. Eberhard, Misc. Publs Mus. Zool. Univ. Mich. *140* (1969).
[2] L. Pardi, Physiol. Zool. *21*, 1 (1948).
[3] K. Eickwort, Insectes Soc. *16*, 67 (1969).
[4] E.-P. Deleurance, C. r. Acad. Sci., Paris *227*, 866 (1948).
[5] F. A. Beach, *Hormones and Behavior* (Paul B. Hoeber Inc., New York 1948). – F. H. Bronson and C. Desjardins, in *The Physiology of Aggression and Defeat* (Eds. B. E. Eleftheriou and J. B. Scott, Plenum Press, New York 1971), p. 43. – A. M. Guhl, in *Sex and Internal Secretion*, 3rd edn. (Ed. W. C. Young, Williams & Wilkins, Baltimore 1961), p. 1240.
[6] F. Engelmann, *The Physiology of Insect Reproduction* (Pergamon Press, New York 1970).
[7] M. K. Bohm, J. Insect Physiol. *18*, 1875 (1972).
[8] R. H. Barth and L. J. Lester, A. Rev. Entomol. *18*, 445 (1973).
[9] P. Wirtz and J. Beetsma, Ent. exp. appl. *15*, 517 (1972). – H. Rembold, C. Czoppelt and P. J. Rao, J. Insect Physiol. *20*, 1193 (1974). – E. R. Jaycox, W. Skowronek and G. Guynn, Ann. ent. Soc. Am. *67*, 529 (1974).

Effects of topical application of juvenile hormone and its analogs on workers of the primitive social wasp, *Polistes annularis*

Treatment group	N	Group means [a]			Average colony rank by ovarian development	
		Longest oocytes (mm)	Mean oocyte length (mm)	Behavioral score	Queenright colonies (n)	Fallen queen colonies (n)
Queens	16	2.25	2.04	2.81	1.00 (12)	4.75 (4)
Hormone treatments						
JH III [c]	9	1.44	1.30 [b]	2.00	2.43 (7)	1.50 (2)
ZR-520 [d]	7	1.32	1.11	1.43	3.40 (5)	1.00 (2)
ZR-515 [e]	7	1.14	0.96	1.57	3.80 (5)	1.50 (2)
ZR-512 [f]	7	1.06	0.89	1.29	3.60 (5)	2.50 (2)
Combined	30	1.22	1.08	1.60	3.23 (22)	1.63 (8)
Controls						
Oil	16	1.02	0.91	1.33	3.09 (12)	3.50 (4)
Untreated	12	1.12	0.94	0.33	3.78 (9)	3.00 (3)
Newly emerged	22	0.27	0.16	0.19	6.16 (18)	6.75 (4)

[a] Significant linear regression of mean oocyte lengths on behavioral score, $p < 0.001$. [b] Significantly different from oil treated controls, $p < 0.05$. [c] Methyl-10,11-epoxy-3,7,11-trimethyl-2,6-dodecadienoate. [d] Ethyl-11-methoxy-3,7,11-trimethyl-2,4-dodecadienoate. [e] Isopropyl-11-methoxy-3,7,11-trimethyl-2,4-dodecadienoate (Altosid®). [f] Ethyl-3,7,11-trimethyl-2,4-dodecadienoate (Altozar®).

individuals' behavior was developed: holds and aggressively defends a nest-face position and usually dominates other individuals in interactions, 3; frequently aggressive in encounters and often dominant, 2; frequently aggressive in encounters but seldom dominant, 1; seldom or never aggressive in encounters and always submissive, 0.

After hormone treatment a colony's demeanor underwent a significant change. The frequency of dominance interactions rose sharply. Whereas, prior to treatment and in the control colonies all individuals spent considerable time resting on the nest, the queens of experimental colonies often aggressively repelled colony members. There was increased movement about the cage by JH treated and control individuals and for some individuals it seemed to focus on the nest face. Displacement from a nest face position, which is a good indicator of hierarchy position, happened sometimes even to queens. However, newly-emerged workers were allowed to remain on the nest whether working or resting and were seldom threatened.

Newly emerged individuals normally do not exhibit ovarian development for up to a week following ecdysis[7]. However, dissection revealed that all mature individuals in the experimental colonies had some oocyte growth. There was a statistically significant difference in oocyte size according to the Kruskal-Wallis test[10] only between the natural JH treated animals and the oil treated controls, although all treated groups showed some mean increase in size over controls (Table). Queens of 12 of 16 experimental colonies had oocytes about twice as long as those of any other adults. 4 colonies had individuals which showed queen-like behavior but possessed small degenerating oocytes. There is a tendency for infirm or displaced queens to fall rapidly in the hierarchy of the colony[2] and such a fall was indicated in these 4 individuals in that they spent little time on the nest face. In these 'fallen-queen' colonies, JH or analog-treated individuals had a clear advantage in ovary development. While the mean oocyte length averaged over all hormone-treated individuals was 1.08 mm, the value for those from fallen-queen colonies was 1.91 mm. Oocyte growth did not occur among the workers in the control colonies.

The correlation between ovarian size and position in the dominance hierarchy[2] was maintained under these experimental conditions. A regression of ovary size on dominance score for all individuals yields a highly significant equation ($p < 0.001$): dominance score $= 0.92$ (mean oocyte size) $+ 0.344$. If the hormone treatment affected vitellogenesis but not dominance one would not expect this relationship to be maintained. In fact, the correlation of mean oocyte size and aggressive behavior was stronger for the combined hormone-trated group than for the oil-treated controls. Kendall's coefficients of rank correlation[10] were 0.28 ($p < 0.05$) and 0.23 ($p < 0.10$) respectively.

The data show that hormone treatment gave an advantage both in behavior and ovarian development, though JH activity was evident in control individuals as well. Though some JH may have rubbed off treated animals onto control individuals, we propose a second hypothesis to explain the evident oocyte growth in these controls. The experimental millieu disrupted the usually stringent control of worker behavior and ovarian development by the queen. By altering the hormone titer of mature individuals, the structure of the dominance hierarchy was weakened owing to the ensuing aggressiveness of these workers. We suggest that this disruption of the hierarchy precipitated a condition of decreasing inhibition of worker corpora allata by the queen allowing activation of the worker glands via neuroendocrine pathways and resulting in vitellogenesis and dominance behavior in both treated and control individuals.

Summary. Repeated topical application of juvenile hormone to workers of the primitive social wasp, *Polistes annularis* resulted in a disruption of colony social structure as indicated by a sharp increase in the frequency of dominance interactions. Ovarian maturation was also observed, probably as both a direct and an indirect effect of hormone treatment.

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[10] R. R. SOKAL and F. J. ROHLF, *Biometry* (W. H. Freeman and Co., San Francisco 1969).

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