

pink bollworm moths, *Pectinophora gossypiella* (Saunders), eliciting a copulatory response in laboratory tests and luring large numbers to field traps. As such, it becomes the first sex attractant to be discovered by empirical means. The attractiveness of this compound, which we have named hexalure, is highly unusual, since propylure<sup>4</sup>, the natural pink bollworm sex pheromone, is a C<sub>16</sub> alkadienol acetate which has a branched chain and *trans* configuration.

Hexalure is far more attractive than propylure, which requires admixture with an activator<sup>5</sup> before it can lure males in the field. In tests with several species of insects, hexalure attracted only the pink bollworm, and this insect was not attracted to the *trans* isomer of hexalure.

Hexalure was synthesized by first condensing 2-(7-octynyloxy)-tetrahydropyran<sup>6</sup> with 1-bromooctane, according to previously published procedures<sup>7</sup>, to give a 40% yield of 2-(7-hexadecynyloxy)-tetrahydropyran (bp, 140–145°C at 0.001 mm Hg;  $n_D^{25}$ , 1.4636). This product was refluxed with acetic acid-acetyl chloride<sup>8,7</sup> to give a quantitative yield of 7-hexadecyn-1-ol acetate (bp, 117–121°C at 0.001 mm;  $n_D^{25}$ , 1.4532), which was semihydrogenated to the desired product. Hexalure (bp, 100–104°C at 0.001 mm, 121.5–124.5°C at 0.08–0.14 mm;  $n_D^{25}$ , 1.4484) is a clear, colorless liquid with a mild odor reminiscent of freshly cut grass. Analysis of one lot by a new gas chromatographic method<sup>8</sup> showed the isomeric composition to be roughly 80% *cis* and 20% *trans*.

The efficacy of *cis*-7-hexadecen-1-ol acetate as an attractant for pink bollworm moths was evaluated in laboratory and field tests at Brownsville, Texas, and Phoenix, Arizona. In a preliminary test, a trap baited with 6 mg of hexalure caught, during 7 days, 51% of the male moths caught by the natural sex attractant extracted from 25 virgin females. Over a 14-day period the synthetic/natural catch ratio was 42%. In extensive field tests 60 mg of hexalure was initially about equal

to 50 female equivalents of natural lure but after 5–7 days the synthetic lure at all test dosages was superior to either natural lure or to live caged virgin female moths.

Hexalure is now being used by the U.S. Department of Agriculture's Plant Pest Control Division to combat the pink bollworm in Florida and several southwestern states. It has been found to be more stable, more convenient to use, and much more economical than the previously used lure, which is a crude methylene chloride extract of the terminal abdominal segments of virgin female moths.

*Zusammenfassung.* Empirisch wurde ein Sexuallockstoff – *cis*-7-Hexadecen-1-ol-acetat – gefunden, der auf Männchen der roten Baumwollkapselmotte, *Pectinophora gossypiella* (Saunders), auffallend stark wirkt.

N. GREEN, M. JACOBSON and J. C. KELLER

Entomology Research Division,  
U.S. Department of Agriculture,  
Beltsville (Maryland 20705, USA) and  
Entomology Research Division,  
U.S. Department of Agriculture,  
Phoenix (Arizona 85040, USA), 21 January 1969.

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## Nephrocalcinosis in Rats After Forced Weaning

After forced weaning of the young, certain biochemical changes appear in the mammary glands, bones<sup>1</sup>, blood<sup>2</sup> and urine<sup>3</sup> of the suckling rat mothers. Rats fed on the so-called Larsen diet were also found to have morphological changes in the aorta<sup>2</sup>. A further manifestation of this period were changes found in the kidneys having the character of nephrocalcinosis.

Thirteen rat mothers of the Wistar strain, aged 3–4 months, were allowed to suckle 12 young each after their first gestation. This group, together with a control group of virgin females, were fed on a Larsen diet<sup>2</sup> containing 1.4% calcium and 0.5% phosphorus. A second group of 21 rat mothers reared under the same conditions and a control group, were fed on a Larsen diet enriched with calcium (1.8% calcium and 0.5% phosphorus). On the day of weaning, which was always on the twenty-first day of lactation, 6 mothers from the first group were killed and 10 from the second. The remaining rats of both groups were killed 24 h after weaning. Blood for determining calcium and phosphorus was obtained by opening the heart. An excision was taken from one or both kidneys for histological examination. The renal calcium, phosphorus and citrate content was determined quantitatively in most animals.

At post mortem examination urolithiasis was observed in the form of gravel (composed of calcium carbonate),

most often in the bladder. Nearly all these cases showed varying degrees of dilatation of the ureters and pelves. Acute suppurative pyelonephritis accompanying the above 2 changes was a frequent finding in the second group. In most animals killed 24 h after weaning the surface of the kidneys showed a typical form of yellowish red marbling. It was only in these animals that nephrocalcinosis was found histologically (Table I) as follows: calcification was found in the epithelium of numerous proximal tubules and in the basement membrane and sometimes in Bowman's capsule. The tubules were always affected in the area of the cortex corticis, but changes reached into the deeper layers of the cortex between the medullary processes. No differences were found between the experimental animals and the controls in the PAS-reaction and on staining with Alcian blue.

The biochemical findings show good correlation (Table II) with the occurrence of nephrocalcinosis. Changes in the calcium and phosphorus levels in the serum

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Table I. Macroscopical and microscopical findings in the kidneys of rats after weaning

		Macroscopical changes			Microscopical changes		No. of animals
		Uro-lithiasis	Pyelo-nephritis	Nephro-calcinosis	Nephro-calcinosis	Pyelo-nephritis	
Larsen diet	Weaning	0	0	0	0	0	6
	Day after weaning	2	0	5	5	0	7
Larsen diet + calcium	Weaning	4	2	0	0	2	10
	Day after weaning	7	4	8	8	4	11
Controls		0	0	0	0	0	12

Table II. Biochemical changes in the kidneys of rats after weaning

	Serum (given in mg/100 ml)		Kidneys (given in mg/g wet wt.)			Calcium	No. of animals	Nephro-calcinosis
	Calcium	Phosphorus	wet wt.	Citrate	Phosphorus			
Larsen diet								
Controls	12.7 ± 0.67	10.4 ± 1.3	1.55 ± 0.34	0.033 ± 0.012	0.345 ± 0.01	0.083 ± 0.028	5	0
Weaning	14.2 ± 0.57 <sup>a</sup>	6.7 ± 1.2 <sup>a</sup>	2.17 ± 0.07 <sup>a</sup>	0.076 ± 0.028 <sup>a</sup>	0.332 ± 0.041	0.110 ± 0.04	4	0
Day after weaning	15.6 ± 1.2 <sup>a</sup>	19.7 ± 1.3 <sup>a</sup>	2.20 ± 0.33 <sup>a</sup>	0.095 ± 0.029 <sup>a</sup>	1.30 ± 0.390 <sup>a</sup>	2.22 ± 1.1 <sup>a</sup>	5	+
	15.3	11.5	1.9	0.084	0.340	—	1	0
Larsen diet + calcium								
Controls	14.2 ± 0.72	11.1 ± 1.7	1.62 ± 0.16	0.039 ± 0.005	0.340 ± 0.009	0.078 ± 0.016	6	0
Weaning	15.9 ± 1.44 <sup>a</sup>	4.4 ± 0.8 <sup>a</sup>	2.20 ± 0.17 <sup>a</sup>	0.110 ± 0.024 <sup>a</sup>	0.321 ± 0.016	0.105 ± 0.005 <sup>a</sup>	6	0
Day after weaning	18.1 ± 1.67 <sup>a</sup>	14.8 ± 3.2 <sup>a</sup>	2.22 ± 0.37 <sup>a</sup>	0.393 ± 0.166 <sup>a</sup>	1.06 ± 0.555 <sup>a</sup>	1.70 ± 1.2 <sup>a</sup>	6	+
	16.1 ± 0.2	11.5 ± 1.2	2.1 ± 0.10	0.100 ± 0.036	0.385 ± 0.02	0.220 ± 0.03	2	0

The mean differences were verified by the Student *t*-test taking into account the condition of whether  $s_1^2 = s_2^2$  or  $s_1^2 \neq s_2^2$ . <sup>a</sup> The difference was found to be at least at the 5% level.

are striking. After weaning, lactational hypophosphataemia gave place to hyperphosphataemia and the calcium level increased on both diets used. We have already pointed to this finding on our previous work<sup>1</sup> where we interpreted it as an expression of activation of the parathyroid glands during lactation. This explanation is supported by the increased renal citrate content on the day of weaning<sup>4</sup>. Nephrocalcinosis developed very quickly within 24 h with marked deposition of minerals in the outer layers of the renal cortex. These layers are usually most affected after the injection of substances causing hypercalcaemia<sup>5</sup> or sex hormones<sup>6</sup>. After forced weaning the rats must excrete the increased calcium load absorbed from the mammary glands and the calcium level rises in the blood and urine<sup>3</sup>. Histological findings indicate that in most of our animals the kidneys were unable to ensure the steady excretion of the calcium load, leading to accumulation at the beginning of the nephrones. We consider that the increased renal citrate content may play a role here since it was a primary finding and its affinity for calcium is well known. Citrate is also reabsorbed in the proximal tubules, that is at the site of the anatomical changes.

It must finally be stressed that post-lactational nephrocalcinosis has so far been observed only after the so-called Larsen diet. The same diet enriched with calcium led to an increased incidence of post-lactational urolithiasis and subsequent pyelonephritis. However, the incidence of nephrocalcinosis, i.e. exclusively postlactational changes, remained approximately the same in both groups. Recently COUSINS and GEARY<sup>7</sup> and a number of older workers, drew attention to the effect of diet. Our control group, which can be compared with the findings of the above authors, showed no pathological changes in the kidneys, however. Other work deals with

the relationship between increased phosphorus intake in the diet and the incidence of nephrolithiasis. Our experience suggests that other mechanisms are probably involved in these cases. We are in agreement with the opinion of COUSINS that the dietary factor does not depend solely on the absolute content of calcium, phosphorus, magnesium, Vitamin D, etc. Not merely the nutritional factor, but very probably also certain hormonal factors are involved in postlactational nephrocalcinosis.

*Zusammenfassung.* Nach dem Abstillen wurde bei Rattenmüttern eine ziemlich schnell sich entwickelnde Nephrocalcinosis festgestellt, die hauptsächlich den Cortex corticis betraf. Bei der biochemischen Analyse der Nieren (Calcium, Phosphor und Zitronensäure) wurde eine gute Korrelation der biochemischen und morphologischen Veränderungen gefunden. Der Zitronensäuregehalt war aber auch in den intakten Nieren erhöht.

A. KOMÁRKOVÁ, Z. ZÁHOŘ  
and V. CZABANOVÁ

Charles University, Central Biochemical Laboratories  
of the Faculty Hospital and  
Institute of Pathological Anatomy,  
Praha 2 (Czechoslovakia), 28 February 1969.

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