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A Novel Female Influences Δ^9-THC Effects on Plasma Hormone Levels in Male Mice

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DALTERIO, S., A. BARTKE AND D. MAYFIELD. *A novel female influences Δ^9-THC effects on plasma hormone levels in male mice.* PHARMAC. BIOCHEM. BEHAV. 15(2) 281–284, 1981.—Exposure to Δ^9-tetrahydrocannabinol (THC) (50 mg/kg) alters the endocrine responsivity of male mice to female-related exteroceptive stimuli. Exposure to a novel female prevents or delays the THC-induced decrease in plasma testosterone (T) and luteinizing hormone (LH) levels. These hormonal alterations are apparently not due to the LH-releasing effects of female-related pheromonal or tactile cues, since administration of luteinizing hormone releasing factor (LRF) did not mimic the effects of a novel female on plasma T levels in THC-treated males. Exposure to a much lower dose of THC (0.5 mg/kg) did augment the LRF-induced increases in plasma T levels suggesting a possible synergism between gonadotropins and THC on androgen production. The present findings suggest that THC-induced alterations in hormonal status may be influenced by complex social or environmental factors.

Oral Δ^9-THC Testosterone Luteinizing hormone Luteinizing hormone releasing factor
Female-related exteroceptive stimuli Hypothalamic-pituitary-gonadal axis Endocrine responsivity
Pheromones Social experience

MARIHUANA, particularly the main psychoactive component Δ^9-tetrahydrocannabinol (THC), can influence male reproductive functions in laboratory animals and in men. Several studies present evidence of reduced plasma testosterone (T) levels [9, 17, 18], suppressed pituitary-gonadotropin release [18, 22, 26, 29] and sexual dysfunction [7, 8, 17, 23] subsequent to cannabinoid exposure in males. However, there is still much that remains unclear regarding cannabinoid effects on the endocrine system, and the mechanisms of the observed cannabinoid actions remain to be elucidated.

In some of our earlier studies we have noted that exposure to female conspecifics could either attenuate [10] or enhance [12] cannabinoid effects on reproductive functions in male mice. Thus, we observed that cannabinoid exposure not only affected the behavioral repertoire of the male [9,10], but also influenced his endocrine responsivity to female-related exteroceptive stimuli. In earlier studies we have characterized the responses of plasma T levels in male mice to housing with other males, aggressive interactions, pairing with a female [1] and exposure to a novel female [19]. We observed that in a male mouse paired with a female for one week, the introduction of a novel female resulted in a pronounced increase in plasma T levels. It was subsequently demonstrated that this hormonal response was independent of copulatory activity, sexual receptivity, or endocrine state of the novel female, though it could not be elicited by another male [6, 20, 21]. Further evidence suggests that the rise in peripheral T is produced by a luteinizing hormone (LH) spike, which occurs in the male in response to female-related pheromonal or behavioral stimuli [4, 15, 16, 20, 21].

The present studies were conducted to examine the effects of a single dose of THC on the characteristic endocrine response of male mice to a novel female. We also attempted to relate these THC actions to the dynamics involved in the mediation of female-induced hormonal influences by studying the interactive effects of luteinizing hormone releasing factor (LRF) and THC.

METHOD

Animals

Random-bred adult male mice were obtained from our breeding colony at 60–80 days of age. Animals were maintained on a 14 hr L:10 hr D lighting schedule and provided access to Wayne Breeder Blox and tap water ad lib. For the experiments involving exposure to a female, male mice that had previously been housed 4 males/cage were individually housed with a female for one week and then exposed to a novel female as indicated. These females were adult, previously housed 4/cage, and were not examined for the stage of the estrous cycle or evidence of mating. For the other studies, adult mice were obtained from stocks of males housed 4/cage since weaning.

Procedures

The THC was administered by oral feeding in sesame oil (20 μl) using a blunted 23 gauge needle at a dose of 50 mg/kg

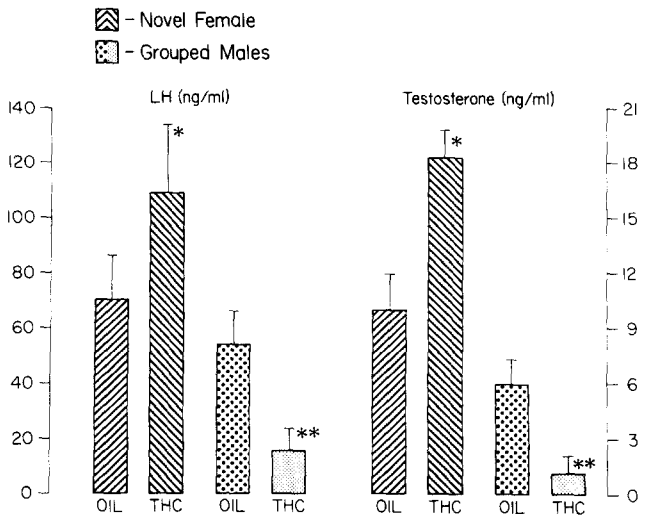

FIG. 1. The effects of Δ^9-tetrahydrocannabinol (THC; 50 mg/kg) and/or exposure to a novel female on plasma LH and testosterone levels in male mice. The THC was administered 1 hr prior to the introduction of a novel female, 30 min after which blood was collected. In grouped males, THC was given 1.5 hr prior to bleeding. (Means ± SE, n=20); *significantly different from controls ($p<0.05$); **($p<0.01$).

body weight. This route of administration and dose have been used in several previous studies [9, 10, 11]. In one experiment THC was administered 1 hr prior to the introduction of a novel female, 30 minutes after which blood was collected. In the second study, the introduction of the novel female and treatment with THC occurred concomitantly, with blood collections at 15 minute intervals for up to 2 hr after exposure.

An additional experiment was conducted to characterize THC interactions with pharmacologically-induced gonadotropin release. In this experiment plasma levels of T and LH were measured after concomitant administration of THC (0.5 or 5 mg/kg) and 5 ng LRF, a dose of LRF which produces levels of plasma T quite comparable to those produced by the stimulation of female exposure.

In a recent study [13] we have determined that there is no difference between the effects of the two highest doses of THC employed in these studies, i.e. 5 or 50 mg/kg body weight on plasma T, LH or FSH during a one-hour period after a single exposure. However, the effects of the lowest dose of THC, i.e. 0.5 mg/kg, did produce consistently different effects on these hormones [13].

Radioimmunoassays

In all experiments, blood was collected between 0930–1130 hr by cardiac puncture under light ether anesthesia in a room adjacent to the mouse housing facility to minimize transfer time and disturbance. Plasma was stored frozen for radioimmunoassay determination of T [2], and LH using the Niswender anti-ovine serum and NIAMDD rat LH kit which has been validated for measuring mouse gonadotropin [3].

Statistics

The values for the hormonal data were not normally distributed, therefore the non-parametric Mann-Whitney U test was employed in the analysis of the data [28].

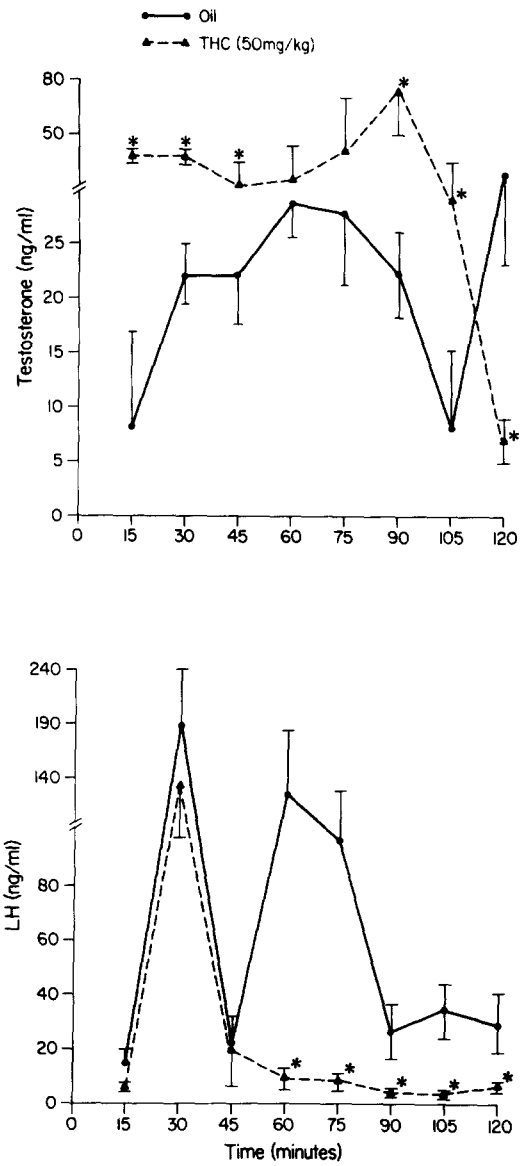

FIG. 2. Plasma levels of testosterone (top) and LH (bottom) in male mice after concomitant exposure to Δ^9-tetrahydrocannabinol (THC, 50 mg/kg) and a novel female. (Means ± SE, n=4 animals per point); *significantly from controls ($p<0.05$).

RESULTS

In male mice pretreated for one hour with THC, plasma levels of LH and T were significantly elevated after exposure to a novel female for thirty minutes. This contrasts with the THC-induced suppression of the levels of both of these hormones in male mice maintained in all-male groups (Fig. 1). In the second study, concomitant novel female exposure and THC administration resulted in higher plasma T concentrations than those produced by the female alone, except at the 2 hr sampling time (Fig. 2, top). In both THC-treated and

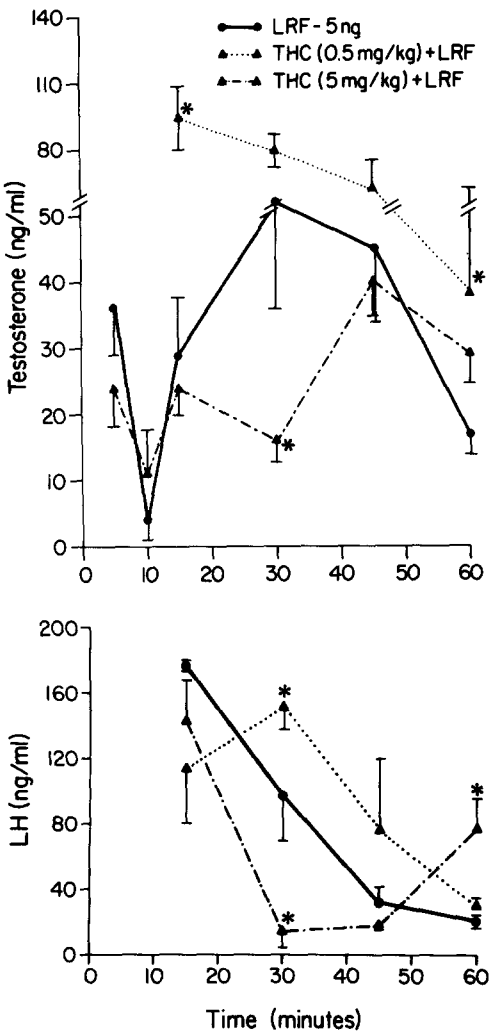

FIG. 3. Effects of concomitant treatment with Δ⁹-tetrahydrocannabinol (THC 0.5 or 5 mg/kg) and LRF (5 ng, IP) on plasma testosterone and LH levels in male mice. (Means ± SE; n=3–6 mice per point); *significantly different from controls ($p<0.05$).

vehicle controls, an initial LH peak occurred. However, in the controls there was a second peak of even longer duration, and LH levels were overall higher than in THC-exposed males during the 2 hr sampling period (Fig. 2, bottom).

In the experiment concerned with interactions of THC and stimulated release of endogenous gonadotropins, the concomitant administration of 0.5 mg THC/kg augmented the effect of 5 ng LRF on plasma T levels, while the higher dose of THC significantly ($p<0.05$) suppressed LRF-induced stimulation of plasma T levels at 30 min, although it had no effect at other sampling intervals (Fig. 3, top). There were no significant differences in plasma FSH levels among any treatment groups at any time during the sampling (data not shown).

DISCUSSION

Administration of THC alters the endocrine response of male mice to exteroceptive cues from a female conspecific. Although it is well known that THC can reduce plasma levels of both T and LH [5], we have recently observed that these effects can be biphasic in nature, with higher THC doses (5 or 50 mg/kg) producing rapid simultaneous increases in plasma T and LH concentrations persisting up to 20 min followed by sustained suppression [13]. Thus, it might seem that the stimulatory effects of THC on the endocrine responses to the novel female may simply be due to THC alone. However, we have shown that plasma T and LH levels are decreased significantly 1.5 hr post-THC treatment in the absence of a female, while exposure to the novel female appears capable of preventing or delaying these depressive effects of THC (Figs. 1 and 2). It is conceivable that the LH release-promoting effects of pheromonal and/or tactile cues emanating from female conspecifics [6,18] may synergize with the early stimulatory action of THC on the pituitary and the testis, resulting in sustained elevations in plasma androgen levels. However, since injection of LRF does not mimic the effects of a novel female in THC-treated males, other effects of social experience may be involved.

It is doubtful that the well-documented effects of THC in altering sensory perception or behavior [14, 24, 25] explain the alterations in endocrine responsivity to female stimuli observed in the present studies. Pretreatment and concomitant administration of THC produced increased plasma T levels after the introduction of the novel female. However, plasma LH levels were increased only in those males pretreated with THC. We did observe that symptoms of behavioral intoxication were more apparent in pretreated males, while they were delayed or less obvious in those males exposed to the novel female at the same time as THC. Thus, behavioral arousal, involving both neural and endocrine stimulation, may attenuate the usually depressive effects of THC on gonadal function, which may then affect androgen-dependent behavioral responses, such as sexual activity. The present results indicate that social and environmental factors may play a critical role in the determination of endocrine responses of the hypothalamic-pituitary-gonadal axis to THC in male mice.

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