Members of opposite sex mutually regulate gonadal recrudescence in the lizard *Calotes versicolor* (Agamidae)

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Adult males and females of the seasonally breeding lizard *Calotes versicolor* were subjected to various social situations under semi-natural conditions to explain the role of socio-sexual factors in gonadal recrudescence. They were grouped as: (i) males and females, (ii) males and females separated by a wire mesh, (iii) same sex groups of males or females, (iv) castrated males with intact females and (v) ovariectomized (OvX) females with intact males from postbreeding to breeding phase. Specimens collected from the wild during breeding season served as the control group. Plasma sex steroid levels (testosterone in male and 17*b*-estradiol in female), spermatogenetic activity and vitellogenesis were the criteria to judge gonadal recrudescence. In intact males and females that were kept together, gonadal recrudescence and plasma sex steroids levels were comparable to those in wild-caught individuals. Gonadal recrudescence was at its least in all male and all female groups, and plasma sex steroids were at basal levels. Association with OvX females initiated testicular recrudescence but spermatogenetic activity progressed only up to the spermatid stage while males separated from females by wire mesh showed spermatogenetic activity for a shorter period. Females grouped with castrated males and those separated from males by wire mesh produced vitellogenic follicles. However, the total number and diameter of vitellogenic follicles, and plasma estradiol levels were lower than in the females grouped with intact males. The findings indicate that association with members of the opposite sex with progressively rising titers of sex steroids is crucial in both initiating and sustaining gonadal recrudescence in the lizard. Thus, members of the opposite sex mutually regulate gonadal recrudescence in the *C. versicolor*.

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1. Introduction

Reproduction in vertebrates involves successful interaction between external (physical, climatic and social environment) and internal (neuroendocrine) factors. There are many studies on the pattern of annual/seasonal cycles of gonads in nonmammalian vertebrates (Licht 1984; Whittier and Crews 1987; Saidapur 1989; Vitt 1992). These studies generally show that a gonadal cycle typically involves recrudescence, breeding and postbreeding quiescent phases. The control of gonadal cycles through endocrine mechanisms and proximate factors is reasonably well understood in lizards. However, little is known about the role of social cues governing reproduction. The integration of social cues may be mediated through optic, olfactory and vomeronasal systems with specific inputs being sent to specific brain nuclei. The relevant nuclei in the limbic system that concentrate steroid hormones receive inputs from these sensory areas. In turn these inputs are projected to the hypothalamus where both internal and external cues are integrated (Whittier and Crews 1987). Studies dealing with the role of socio-sexual factors in reptilian reproduction have been carried out mainly by two schools of researchers: Crews (Crews 1974, 1975; Crews *et al* 1974; Crews and Garrick 1980; Gustafson and Crews 1981; Greenberg and Crews 1990; Hartman and Crews 1996) and Summers (Summers 1995; Andrews and Summers 1996; Summers *et al* 1995, 1997).

Keywords. Gonads; lizard; reproduction; socio-sexual factors

These studies have focussed on the genus *Anolis* (family: Iguanidae) and, in general, show that the sexual composition of conspecifics of opposite sex, social experience, and dominance-subordinance hierarchy influence gonadal recrudescence and mating behaviour of the cagemates.

Calotes versicolor (family: Agamidae) is a seasonally breeding lizard, widely found all over India. It breeds from late May to October coinciding with the south-west monsoon. Its reproductive cycle can be divided into recrudescence, breeding and postbreeding phases as in other lizards. The males are spermatogenetically active from April–September (Gouder and Nadkarni 1979; Radder *et al* 2001) and gravid lizards may be seen from May–October. The *C. versicolor* is a polyautochronic and multiclutched lizard (Shanbhag and Prasad 1993; Shanbhag *et al* 2000a; Radder *et al* 2001). Phenological data and experimental manipulation studies have provided information on the role of certain proximate (temperature and rainfall) and endocrinal factors that control the onset and termination of gonadal recrudescence in the *C. versicolor* (Gouder and Nadkarni 1979; Shanbhag and Prasad 1993; Shanbhag *et al* 2000b). However, the influence of psychobiological factors and conspecifics on gonadal recrudescence in the same or opposite sex are unknown. Hence, the present study was undertaken to investigate the role of socio-sexual factors on gonadal recrudescence in *C. versicolor*. Therefore, in their postbreeding resting phase (November), these lizards were subjected to diverse situations such as intact males and females housed together or isolated by a wire mesh, or as all male and all female groups, groups of castrated males housed with intact females, and ovariectomized (OvX) females with intact males. The gonadal recrudescence and plasma sex steroid profiles were assessed in June, the peak breeding season. Such studies that explain the importance of interaction with conspecifics of the same or opposite sex in controlling or affecting the gonadal recrudescence in lizards are needed.

2. Materials and methods

Adult male and female (snout-vent length, SVL = 9·7–11·3 cm) *C. versicolor* were collected from the surrounding areas of Dharwad city (15°17′N and 75°3′E) during the third week of November 1999. They were acclimated to indoor terraria for one week and then categorized into seven groups.

Group I – Initial control: Lizards autopsied on day 1 of the experiment (n = 5 each sex).
Group II – All Male (devoid of tactile/visual/chemical cues from members of opposite sex, n = 5).
Group III – All female (devoid of tactile/visual/chemical cues from males, n = 5).
Group IV – Male–female housed together (1 : 1 ratio; 5 lizards of each sex).
Group V – Sexually isolated rearing: Males (n = 5) were separated from females (n = 5) by a wire mesh.
Group VI – OvX female (n = 5) with intact males (n = 5).
Group VII – Castrated male (n = 5) with intact female (n = 5).
Group VIII – Wild caught lizards at termination of experiment (final control).

The wild caught lizards provided the measure of gonadal activity in nature. The terraria (120 H × 90 L × 90 W cm) used in this experiment were made of Syntex plastic wire mesh (hole size 5 mm hexagonal) on all sides except the base. The substratum contained black soil (15 cm height), potted plants, twigs, hiding places, bricks and a water dish. During the rainy season water was sprayed to mimic the moisture as in the wild. The lizards were provided a mixed diet of live cockroaches, grasshoppers, crickets, and larvae of the silk worm or *Castor semilooper* every other day. Water was provided *ad libitum*.

The experiment ended after six months to coincide with the peak breeding phase (June 2000) of the lizards in the wild. At autopsy, SVL (cm), and mass of body, gonads, and oviducts (g) for each lizard were recorded. Gonads and epididymis were fixed in Bouin's fluid and preserved in 70% ethanol for routine histology. They were cut at 5 μm thickness and stained with hematoxylene-eosin. The diameter of the testis, seminiferous tubule, and Leydig cell nucleus were measured as described earlier (Sharma and Shanbhag 1992) and spermatogenetic activity visually assessed. The epithelial height of mid-epididymal tubule was measured from 25 tubule cross sections/lizard using an ocular micrometer. The data are expressed as mean ± SEM for each group.

The number and diameter of the largest follicles were recorded from the left ovary of each lizard. The ovarian follicles < 1·0 mm diameter were measured using a ocular micrometer, while those > 1·0 mm were measured using a micrometer. Follicles > 2·5 mm diameter represent the vitellogenic status (Shanbhag and Prasad 1993).

2.1 *ELISA for plasma sex steroids*

Serum was collected from the lizards as described previously (Shanbhag *et al* 2000b) and used to measure levels of testosterone (T) in males and 17b-estradiol (E_2) in females as per the protocol supplied with the respective ELISA kits (Bio source Europe S A, Belgium).

Plasma T: The minimum detectable concentration (MDC) was 0·05 ± 0·02 ng/ml according to the manufacturer's

protocol. Precision for intrarun assay had a coefficient of variation of 4·88%. Recovery rate for 1·2, 4·0, and 14·0 ng/ml *T* added to the serum was 105%, 103% and 101·4% respectively.

Plasma E_2: The MDC for E_2 in our assay was 5 ± 2 pg/ml. Precision for intrarun assay had a coefficient of variation of 4·2%. Recovery for 50, 100, 270 and 900 pg/ml E_2 added to the serum solution was 102%, 98%, 96·3% and 100·2% respectively.

2.2 *Statistical analysis*

Means and standard errors for all recorded variables were computed from untransformed data and are represented in tables 1 and 2. For other analyses data were log transformed. Residuals for body, gonad and oviduct masses, diameters of testis, seminiferous tubule and Leydig cell nucleus, and the number of largest follicles were generated by regressing each trait on SVL. Variations in the above mentioned parameters, plasma *T* and E_2 levels, epithelial height of the epididymis and diameter of largest ovarian follicles (LFD) among experimental groups were analysed by one way ANOVA followed by Tukey's HSD multiple range test. Significance was accepted at $P < 0{\cdot}05$ level. Analyses were performed using SPSS (version 6.1.3 for Windows).

3. Results

3.1 *Testicular recrudescence in the presence/absence of females*

Residual testis mass (ANOVA $df_{5,24}$, $F = 101{\cdot}07$, $P < 0{\cdot}0001$) and diameter of testis (ANOVA $df_{5,24}$, $F = 624{\cdot}0$, $P < 0{\cdot}0001$), seminiferous tubule (ANOVA $df_{5,24}$, $F = 359{\cdot}93$, $P < 0{\cdot}0001$) and Leydig cell nucleus (ANOVA $df_{5,24}$, $F = 294{\cdot}75$, $P < 0{\cdot}0001$), epithelial height of the epididymal tubule (ANOVA $df_{5,24}$, $F = 163{\cdot}62$, $P < 0{\cdot}0001$), and plasma *T* levels (ANOVA $df_{5,24}$, $F = 24{\cdot}05$, $P < 0{\cdot}0001$) varied significantly among different experimental groups (table 1 and figure 1) while residual body mass did not (ANOVA $df_{5,24}$, $F = 1{\cdot}24$, $P > 0{\cdot}05$).

Table 1. Effect of group composition on testis mass, diameter of testis, seminiferous tubule and Leydig cell nucleus, and epithelial height (mean ± SE) of the epididymis in male *C. versicolor* ($n = 5$ for each group).

Group	Testis mass (g)	Diameter of			Epithelium height (μm)
		Testis (mm)	S. Tubules (μm)	Leydig cell nucleus (μm)	
Initial control	$0{\cdot}03 \pm 0{\cdot}01^a$	$1{\cdot}16 \pm 0{\cdot}07^a$	$61{\cdot}71 \pm 1{\cdot}46^a$	$1{\cdot}46 \pm 0{\cdot}02^a$	$10{\cdot}47 \pm 0{\cdot}51^a$
With intact female	$0{\cdot}74 \pm 0{\cdot}20^b$	$6{\cdot}19 \pm 0{\cdot}27^b$	$303.24 \pm 9{\cdot}25^b$	$5{\cdot}96 \pm 0{\cdot}23^b$	$46{\cdot}56 \pm 0{\cdot}86^b$
With OvX female	$0{\cdot}39 \pm 0{\cdot}04^c$	$3{\cdot}85 \pm 0{\cdot}21^c$	$211{\cdot}48 \pm 11{\cdot}46^c$	$4{\cdot}27 \pm 0{\cdot}14^c$	$32{\cdot}17 \pm 1{\cdot}33^c$
Separated by wire mesh	$0{\cdot}40 \pm 0{\cdot}10^c$	$3{\cdot}77 \pm 0{\cdot}13^c$	$206{\cdot}79 \pm 10{\cdot}48^c$	$4{\cdot}01 \pm 0{\cdot}12^c$	$25{\cdot}83 \pm 2{\cdot}15^d$
All males	$0{\cdot}11 \pm 0{\cdot}01^d$	$1{\cdot}94 \pm 0{\cdot}10^d$	$79{\cdot}15 \pm 2{\cdot}41^d$	$2{\cdot}62 \pm 0{\cdot}07^d$	$11{\cdot}68 \pm 0{\cdot}51^a$
Wild-caught*	$0{\cdot}89 \pm 0{\cdot}16^b$	$6{\cdot}36 \pm 0{\cdot}32^b$	$325{\cdot}44 \pm 13{\cdot}16^b$	$6{\cdot}26 \pm 0{\cdot}22^b$	$43{\cdot}90 \pm 1{\cdot}58^b$

Non-identical superscripts indicate a significant difference between groups at $P < 0{\cdot}05$.
*Wild-caught lizards at the termination of experiment.

Table 2. Effect of group composition on body mass, ovarian mass, number of vitellogenic follicles in left ovary and range of largest follicular diameter (LFD) in female *C. versicolor* ($n = 5$ for each group).

Group	Body mass (g)	Ovarian mass (g)	Vitellogenic follicles (number)	LFD (mm)
Initial control	$36{\cdot}00 \pm 1{\cdot}92^a$	$0{\cdot}08 \pm 0{\cdot}01^a$	–	$0{\cdot}59 - 0{\cdot}80^a$
With intact male	$46{\cdot}60 \pm 2{\cdot}18^b$	$3{\cdot}12 \pm 0{\cdot}90^b$	$12{\cdot}20 \pm 0{\cdot}37^a$	$3{\cdot}80 - 7{\cdot}96^b$
With aX male	$42{\cdot}60 \pm 1{\cdot}29^b$	$1{\cdot}12 \pm 0{\cdot}30^c$	$9{\cdot}20 \pm 0{\cdot}58^b$	$3{\cdot}17 - 5{\cdot}41^c$
Separated by wire mesh	$40{\cdot}80 \pm 1{\cdot}88^b$	$0{\cdot}48 \pm 0{\cdot}09^d$	$6{\cdot}80 \pm 0{\cdot}58^c$	$2{\cdot}78 - 3{\cdot}92^c$
All females	$48{\cdot}00 \pm 4{\cdot}58^b$	$0{\cdot}13 \pm 0{\cdot}01^d$	–	$1{\cdot}00 - 2{\cdot}29^d$
Wild-caught*	$53{\cdot}00 \pm 2{\cdot}88^b$	$2{\cdot}05 \pm 0{\cdot}90^b$	$10{\cdot}60 \pm 0{\cdot}93^a$	$2{\cdot}40 - 7{\cdot}52^b$

Non-identical superscripts indicate a significant difference between groups at $P < 0{\cdot}05$ based on the residuals.
*Wild-caught lizards at termination of experiment.

3.1a *Initial control*: The testis mass and diameter of testis and seminiferous tubule were lowest in the initial controls (table 1). Plasma *T* level was also minimal in this group (figure 1). The testis contained small seminiferous tubules lined by Sertoli cells and a few spermatogonia (figure 2). The interstitial areas were large and Leydig cells were indistinct from fibroblasts (figure 2). Epididymal epithelial height was significantly low in these lizards compared to all other groups (table 1 and figure 3).

3.1b *All male group*: An increase in the mass and diameter of testis, seminiferous tubule and Leydig cell nucleus over the initial controls was evident in this group, but plasma *T* levels and epithelial height of the epididymis were low as in the initial controls (table 1 and figure 1). All the recorded traits measured were significantly lower than in the corresponding males of the other remaining groups (table 1 and figure 1). The seminiferous tubules contained only spermatogonia and Sertoli cells. The interstitial area was less, in comparison to that in the initial controls (figure 4). The diameter of epididymal tubules increased over the initial controls (figure 5).

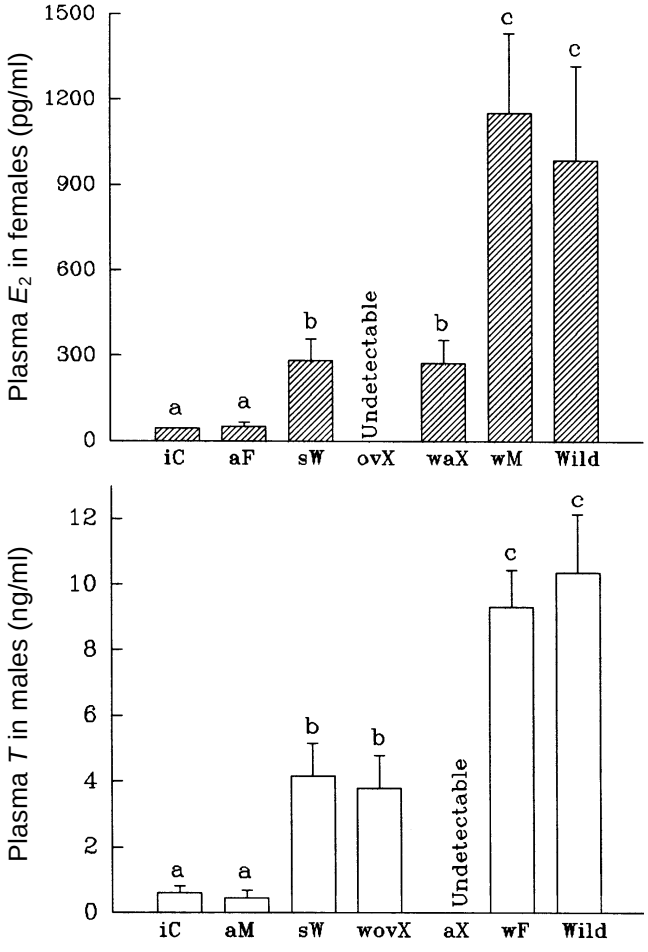


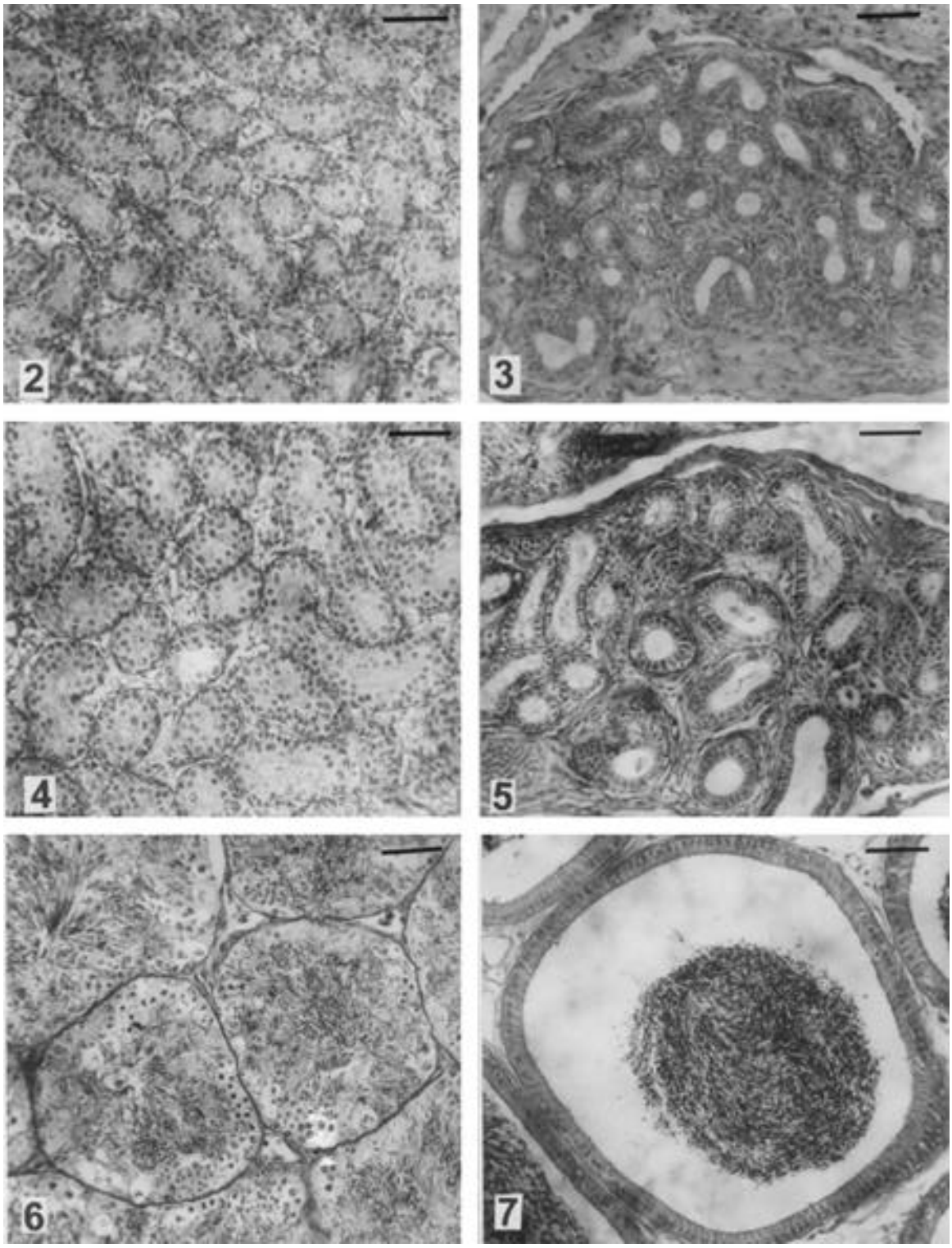
Figure 1. Effect of group composition on plasma *T* (males) and E_2 (females) in *C. versicolor*. The values are mean ± SE ($n = 5$ for each group). Non-identical alphabets above the bar indicate significant difference among groups. (iC, initial control; aM, all males; aF, all females; sW, separated from opposite sex by wire mesh; waX, kept with castrated males; wovX, maintained with OvX females; wM/wF, kept with intact males and females; ovX, overiectomized females; aX, castrated males; Wild, wild-caught male/females).

3.1c *Males separated from females by wire mesh or housed with OvX females*: The testis mass and diameter, diameters of seminiferous tubule and Leydig cell nucleus, and plasma *T* levels were comparable in the two groups. However, epithelial height of the epididymis was significantly greater in specimens housed with OvX females compared to those separated from the females by a wire mesh (table 1). In both groups all the parameters mentioned above registered significantly higher values over the initial control and the males reared in isolation from the females. Yet the values were significantly lower than in the males housed with intact females and wild-caught specimens (table 1, figure 1).

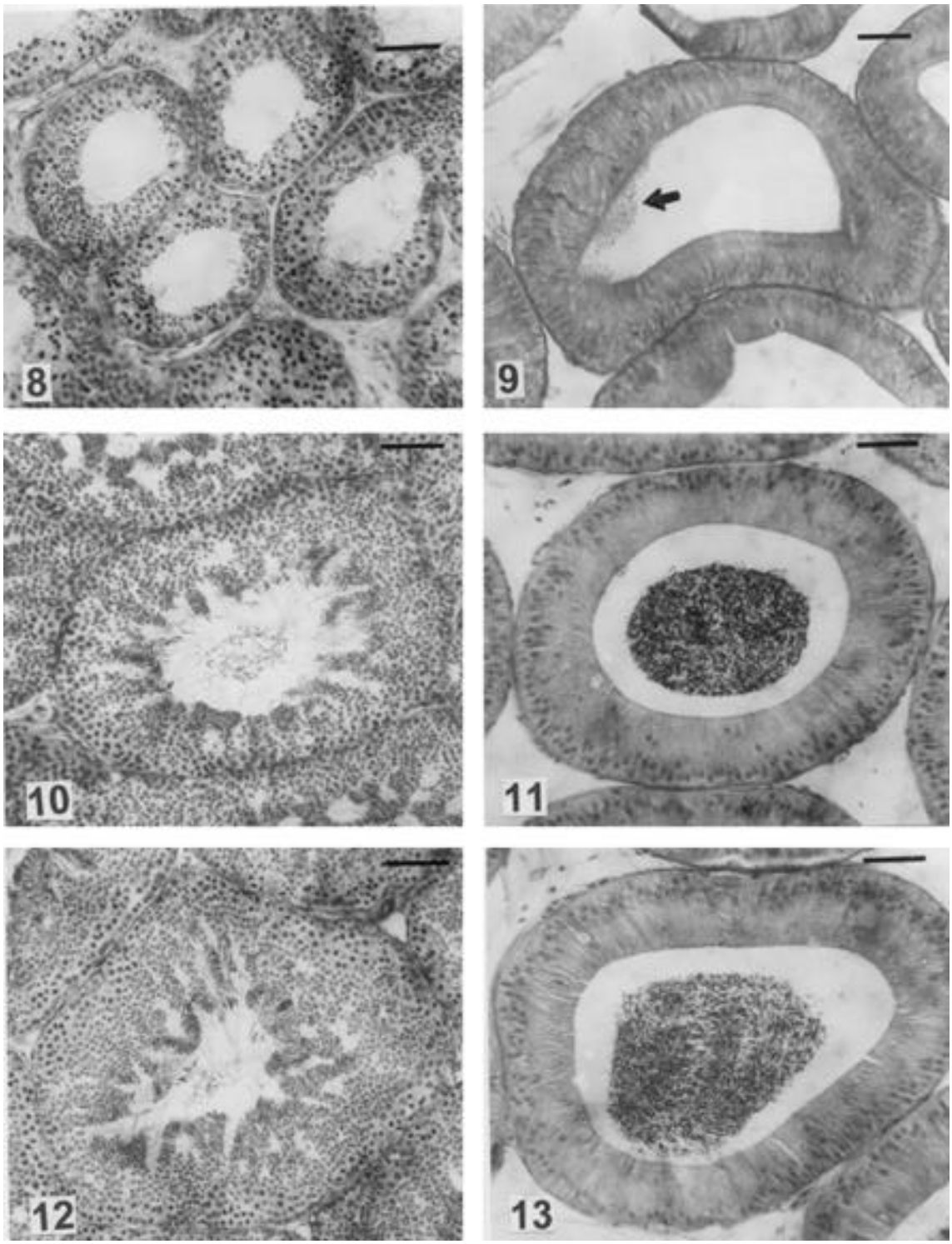
The testis of males separated from females by a wire mesh contained all stages of spermatogenesis, a few degenerating spermatids and sperm (figure 6). Epididymal tubules contained sperm. Though the epididymal tubules were larger in size epithelial cells appeared non-secretory, devoid of granular material and exhibited early signs of regression (figure 7). Further, the epididymal epithelial cell height varied among individuals of this group. In one of the lizards, the height of epithelial cells was very low, contributing to a low mean value for this group.

In males housed with OvX females, spermatogenesis progressed up to the spermatid stage (figure 8). No sperm were seen in the epididymis. The epithelial cells of the epididymis were typically columnar and exhibited secretory granules (figure 9). The epithelial cell height was significantly greater than in the initial control and all male groups as well as of those separated from females by a wire mesh.

3.1d *Males housed with intact females*: The testis mass and diameter, seminiferous tubule and Leydig cell nuclear diameters, epididymal epithelial height were greatest in these lizards compared to the corresponding males of other groups (table 1). Plasma *T* levels were high in these lizards (figure 1). Seminiferous tubules were large and densely populated with all stages of spermatogenesis with abundant spermatids and sperm. The epididymal epithelial cells were columnar and exhibited secretory granules at their free ends (figure 10 to 13). Epididymal tubules contained sperm. All the parameters examined were comparable with wild-caught males (June 2000, table 1, figure 1).



Figures 2–7. (**2, 4, 6**) Transverse sections of testis of *C. versicolor*. (**2**) Showing small seminiferous tubules containing a few spermatogonia and Sertoli cells of Initial control group. (**4**) All male group exhibiting slightly enlarged seminiferous tubules and reduced interstitial space. (**6**) Showing seminiferous tubules exhibiting degenerating spermatogenetic stages in males separated from female by wire mesh. (Scale line 50 μm.) (**3, 5, 7**) Effect of group composition on epididymis of *C. versicolor* showing small epididymal tubules with small epithelial cells in initial control group (**3**) and all male group (**5**). (**7**) Larger epididymal tubules in males separated from females by wire mesh. Note the presence of sperm in the lumen of epididymal tubules. (Scale line = 50 μm.)



Figures 8–13. (**8, 10, 12**) Effect of group composition on testis, spermatogenesis and Leydig cells in *C. versicolor*. (**8**) Production of a few spermatogonia and spermatocytes. In lizards housed with OvX female. (**10**) Enlarged seminiferous tubules and all stages of spermatogenesis including sperm in lizards housed with intact females. (**12**) Spermatogenetic activity and size of seminiferous tubules in wild-caught specimens are comparable to that in **10**. (Scale line 50 μm.) (**9, 11, 13**) Effect of group composition on epididymis of *C. versicolor*. (**9**) Epididymal tubules with taller epithelial cell height with secretory material at the free end of the cells (arrow) in males housed OvX female. Note the absence of sperm. (**11**) Showing epididymal tubules with tall columnar epithelial cells in males housed with intact female. Note the presence of sperm. (**13**) Showing epididymal tubules in wild-caught male comparable to that in **11**. (Scale line = 50 μm.)

3.2 *Ovarian recrudescence in the presence/absence of males*

Mean residual body mass varied significantly among different groups (ANOVA $df_{5,24}$, $F = 14{\cdot}36$, $P < 0{\cdot}05$, table 2). The mean body mass of females in all the experimental groups was comparable with and higher than the initial control group. Significant variations were observed in residual ovarian mass (ANOVA $df_{5,24}$, $F = 24{\cdot}06$, $P < 0{\cdot}001$), oviduct mass (ANOVA $df_{5,24}$, $F = 3{\cdot}76$, $P < 0{\cdot}05$), number and size of the largest follicles (ANOVA $df_{5,24}$, $F = 11{\cdot}32$, $P < 0{\cdot}001$ and $df_{5,24}$, $F = 38{\cdot}10$, $P < 0{\cdot}001$ respectively, table 2), and plasma levels of E_2 (ANOVA $df_{5,24}$, $F = 27{\cdot}62$, $P < 0{\cdot}0001$, figure 1) among the experimental groups.

3.2a *Initial control and all female group*: The mean residual ovarian mass and oviduct mass of the initial control and all female group lizards that exhibited small previtellogenic follicles were significantly lower compared to that of the other groups (table 2). Plasma E_2 level was basal in both the groups (figure 1).

3.2b *Females separated from males by wire mesh, or housed with castrated males*: Ovarian mass was significantly greater in these two groups compared to that in the initial control and all female groups. Further, between the two groups, the ovarian mass of females kept with castrated males was significantly greater than in those separated from males by wire mesh. In both the groups, the ovaries contained vitellogenic follicles of various sizes. The largest follicles were of comparable size in the two groups (table 2). However, the number of vitellogenic follicles was greater in females maintained with castrated males than in the lizards isolated from males by wire mesh (table 2). Plasma E_2 level was comparable between the two groups (figure 1). Though the diameter of follicles and plasma E_2 levels in these two groups registered significantly higher values than in the initial control and all female groups, the values were significantly lower than in the group in which females were maintained with intact males and wild caught females (table 2 and figure 1).

3.2c *Females housed with intact males*: Residual ovarian and oviduct mass, the number and size of vitellogenic follicles and plasma E_2 levels were comparable to those in wild caught females i.e. significantly greater than in the lizards of all other groups (table 2, figure 1).

4. Discussion

Though endocrine and proximate factors regulating seasonal gonadal cycles are reasonably better understood in lizards, information on the role of socio-sexual factors in gonadal recrudescence is limited to a few species, mainly to *Anolis* and *Cnemidophorus*. The studies showed that in the female *Anolis carolinensis*, isolation, housing with a castrated male or with another female slowed down ovarian growth, and they laid eggs, devoid of shells (Crews *et al* 1974). Similarly, the male *A. carolinensis* when housed with females exhibited heavier testes and more advanced spermatogenesis than those that were isolated from females (Crews and Garrick 1980). In this species, the appearance of pink dewlap in males is believed to induce enhanced gonadotropin secretion in females. *Cnemidophorus ironatus* females reared in the absence of males or housed with castrated males ovulated fewer eggs compared to those housed with castrated males treated with exogenous *T* (Crews *et al* 1986). In *Cnemidophorus uniparens*, a parthenogentic species, the probability of ovulation fluctuated depending upon social conditions. For instance, fewer individuals ovulated when housed with OvX cagemates compared to those housed with intact females or with OvX individuals treated with dehydrotestosterone or implanted with silastic capsules containing progesterone (Gustafson and Crews 1981; Crews *et al* 1986). The above studies suggest that the sex steroid millieu of conspecifics of opposite sex/cagemates facilitates gonadal development in these species. Summers and his group proposed a neuroendocrine mechanism to explain regulation of reproductive status in *A. carolinensis* maintained under different socio-sexual setups. The social interactions between and within sexes among these lizards seems to result in a dominant-subordinate hierarchy in sexual behaviour (Summers *et al* 1995; Andrews and Summers 1996; Summers *et al* 1997) that is mediated neurochemically. For instance, when the female *A. carolinensis* is housed in a group of 5 and competing for one mate, only one or two exhibits recrudescing ovaries. The individuals with recrudescing ovaries happened to be the dominant individuals. An individual female when paired with a male exhibited normal ovarian growth while an isolated female had quiescent ovaries (Summers *et al* 1995, 1997). Region specific activation of monoamine systems in the brains is linked to social hierarchy and reproductive status in *Anolis*. A rise in monoamines may be related to aggressive interaction with other females or submissive behaviour towards dominant males. Also, reproductively dominant females had greater dopaminergic activation in telencephalon than in the subordinate females (Summers *et al* 1997). In contrast, in subordinate males (Summers and Greenberg 1995) and females (Summers *et al* 1997) serotonergic activation was elevated in the brainstem. Therefore, establishment of dominant social status plays an important role in gonadal recrudescence, and access to mates in *A. carolinensis*.

In *C. versicolor* we did not come across any aggressive behaviour among or between the sexes. Moreover, in our

studies terraria used were large in size and males and females were housed in a 1 : 1 ratio. This might have led to abolition of competition for space or a mate. In *C. versicolor* there is no evidence of social hierarchy *vis-a-vis* dominant and subordinate individuals that can account for failure of gonadal recrudescence in the present experimental set up. In fact, within a given group of individuals, the degree of gonadal development was identical, thereby ruling out the effects of dominant individuals, if any, over the subordinate ones. Even the *T* and E_2 levels were comparable in individuals of a group (after correction for body size). Thus effects of social hierarchy on gonadal recrudescence as reported in *A. carolinensis* are not applicable to *C. versicolor*. Ideally, initial and final control groups of lizards belonging to the same reproductive age should have been used. Although it was not possible to establish the reproductive age as they were wild-caught, lizards of comparable SVL were chosen assuming they were of a comparable age-group. Since lizards have indeterminate growth, individuals of comparable SVL of the same sex are generally of a comparable age-group (Patnaik and Behara 1981). *C. versicolor* are seasonal breeders and individuals more than 8·6 cm in SVL are reproductively active during the breeding season (Shanbhag *et al* 2000a).

In the present study, the lizards in all male or all female groups did not undergo gonadal recrudescence thereby suggesting that some cues, may be tactile/ optic/chemical (olfactory or by tongue flicking), from that opposite sex are needed for normal gonadal recrudescence in *C. versicolor*. Association of *C. versicolor* with gonadectomized conspecifics of the opposite sex resulted in partial recrudescence of gonads. This suggests that tactile and optic cues alone are not enough to bring about complete gonadal recrudescence and cues (chemical/ olfactory) derived from sexual composition of members of opposite sex are also needed. The significance of tactile cues is evident in males separated from females by a wire mesh. In these males, all the stages of spermatogenesis were produced but they showed signs of widespread degeneration. Furthermore, in these lizards sperm were found in the epididymal tubules thereby suggesting completion of at least one cycle of spermatogenesis. These males could view the females and also perceive chemical stimuli if any, emanating from the females housed on the other side of the wire mesh. Yet deprivation of physical contact with members of the opposite sex led to low *T* levels, failure of subsequent spermatogenetic waves and regression of epididymal epithelial cells. Thus, it is apparent that the visual and chemical cues from females may induce completion of spermatogenesis, but sustenance of spermatogenetic activity requires physical contact with females who have recrudescing ovaries that secrete high levels of E_2. Possibly, elevated E_2 levels are needed for production of pheromonal signals in the females. Perception of these chemicals by the males seems to play an important role in sustaining the qualitative and quantitative aspects of spermatogenetic activity. Similarly, sub-optimal ovarian growth in females that were separated from intact males by wire mesh suggests that the visual and tactile stimuli as well as the chemical (olfactory) cues associated with rising *T* levels in males in breeding condition are needed for recruitment and growth of optimal number of eggs and for production of high E_2 levels in conspecific females. It is likely that high *T* levels in males induce production of cues that influence ovarian recrudescence in females. Normal spermatogenetic and ovarian activity, plasma *T* and E_2 levels in intact male and female lizards respectively when housed together, as in the wild-caught lizards, further support the view that socio-sexual cues are required for normal gonadal recrudescence in *C. versicolor*. The ability to perceive intraspecific odour is reported in *Sceloporus occidentalis* (Duvall 1979), *Iguana iguana* (Werner *et al* 1987), *Cordylus cordylus* (Cooper *et al* 1996), *Lacerta monticola* (Aragon *et al* 2000, 2001) and in several species of skinks (review by Cooper 1994; Olsson and Shine 1998). Tongue-flicking serves to sense (through vomeronasal organs) the chemical signals from faeces/dropping and exudates from proctodeal and femoral glands of conspecifics, and is a mode of social communication in these lizards. Incidently, tongue-flicking behaviour is uncommon among agamids including *C. versicolor* that rarely exhibit tongue-flicking behaviour. Findings suggest, albeit indirectly, that *C. versicolor* possess the ability to detect conspecifics of the opposite sex through olfaction.

The present study on *C. versicolor* shows that mechanisms controlling gonadal recrudescence in lizards are complex and involve endocrine and proximate factors as well as socio-sexual interaction between conspecifics of the opposite sex. Further, it shows that besides visual and tactile cues, rising sex steroid levels, *T* in males and E_2 in females are essential for production of pheromonal signals that induce optimal gonadal recrudescence in the members of the opposite sex. The absence of any one or all (visual, tactile and chemical) cues from the conspecific members of the opposite sex impair gonadal recrudescence in *C. versicolor*. However, sensory modulators involved in the regulation of gonadal recrudescence in *C. versicolor* are not clear at present. It is noteworthy that the production of gametes is an expensive phenomenon and their production in the absence of mates in a seasonally breeding species would be a wasteful process. The present study shows that natural selection has shaped the perception of socio-sexual cues from the members of opposite sex for regulating their own gonadal recrudescence *vis-a-vis* reproductive effort in *C. versicolor*.

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