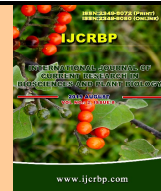




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## Original Research Article

### The Post Ovarian Genital Complex of the Dragonfly *Crocothemis servilia* (Drury 1773), (Odonata: Libellulidae)

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Abstract	Keywords
<p>In the dragonfly, <i>Crocothemis servilia</i> (Drury 1773), the post ovarian genital complex (POGC) is composed of a pair of very large, prominent, bean-shaped spermathecae, a small bursa copulatrix and a large ventrally placed vagina. The right spermathecae is substantially larger than the left. The wall of the POGC is basically composed of an outer muscle layer, middle epithelial layer resting on a basement membrane and an internal layer of cuticle. The bursa copulatrix is a small, oval chamber with a triangular lumen since its dorsal region is sunk between the two spermathecae and lie just above the junction of median oviduct and vagina. The spermathecae wall is composed of a thin layer of indistinct epithelial cells followed by a very thin cuticular intima. The bursa copulatrix opens into the vagina through the <i>Bursa Communis</i> and fertilization pore. The <i>Bursa Communis</i> contains short and sharp denticles all along its inner surface and around the inner and outer tips. The fertilization pore is a short tunnel, formed of three plates, hanging from the roof of the vagina and internally lined with spines. The vagina is short and squat with a thick covering of muscle bands. The vagina is divided into anterior, middle and posterior regions. The anterior region of the vagina is amalgamated with the descending median oviduct. The lumen is circular so that the egg can slip inside the vagina from the oviduct. The middle region of vagina exhibits prominent modification in its cuticular intima so as to support and form the <i>Bursa Communis</i>. The cuticular intima of posterior region is stubbed and this region tapers into the female gonopore.</p>	<p>Bursa copulatrix <i>Crocothemis servilia</i> Odonata Post ovarian genital complex Spermathecae</p>

## Introduction

The female reproductive system in insect usually consists of a pair of ovaries and their oviducts which open into a post ovarian genital complex (POGC). The

POGC consist of the sperm storage organs i.e. bursa copulatrix and spermatheca and while the vagina is the extension of the median oviduct or the involution of the female gonopore. The male deposits his sperms in the sperm storage organs during copulation, where it is

stored until needed. The spermathecal gland along with other sex glands produces nutrients in order to keep the sperm viable in the spermatheca, where it can survive for weeks, months or even years depending upon the reproductive biology of the species. The vagina is generally the site of egg fertilization.

Tillyard (1917), Asahina (1954), Prasad and Srivastava (1961) and Bjanes (1974) initiated the study of the POGC in Odonata. Recent studies on the POGC indicate that it comprises typically of the spermathecae, bursa copulatrix and vagina on the eighth abdominal sternum and the sex glands are wanting in all libellulid and gomphid dragonflies (Midttun, 1976; Miller, 1982a, b, 1984, 1987; Waage, 1982, 1984, 1986; Siva-Jothy, 1987; Michiels, 1989; Andrew and Tembhare, 1994, 96, 97).

The gross structure of the female genitalia was described by Miller (1982a, 1984) in *Brachythemis lacustris*, *Nesciothemis farinose*, *Nesciothemis farinose* and *Orthetrum chrysostigma* with reference to possible copulatory event and mechanism. Waage (1986) described the female genital complex of *Celithemis elisa* and *Erythemis simplicicollis* while studying the phenomena of sperm displacement and copulatory duration. Siva-Jothy (1984) studied the post ovarian genital complex in *Crocothemis erythraea* and *Orthetrum cancellatum*. Srivastava and Srivastava (1988) studied it in the damselfly, *Ischnura rufostigma* while Andrew and Tembhare (1994, 96, 97) detailed the POGC of the libellulid, *Tramea virginia*, aeshnid *Anax guttatus* and zygopteran *Ischnura aurora*.

The available literature on odonate genitalia, moreover, reveals that various morphological and physiological aspects of the POGC are still unexplored. Therefore the present work was undertaken to study in details the structure of the POGC of the dragonfly, *Crocothemis servilia*.

## Materials and methods

*Crocothemis servilia* (Drury 1773) females (Fig. A) were collected from the water bodies around the campus of Hislop College, Nagpur and dissected in Ringer's saline solution under the stereoscopic binocular stereo microscope (Magnus- MS 24). The post ovarian genital complex (POGC) was removed from the abdomen and immediately fixed in aqueous Bouin's fixative for 24 h. The POGC was washed in running water for half an

hour and later dehydrated in a series of ascending grades of aqueous alcohol, cleared in xylene, embedded in molten paraffin wax at 62°C for block preparation. The paraffin blocks were mounted on the block-holder of a Rocking microtome (Radical Cambridge type) and sections of 6 µm–10 µm thicknesses were cut in longitudinal and transverse sections and spread on glass slides. The sections were double stained with Delafield Haematoxylin – Eosin (Tembhare, 2008).

Selected sections were photographed at required magnifications with the help of Cat Cam Microscope Eyepiece digital camera (CC 130) under the Carl Zeiss microscope (Primostar- 37081).

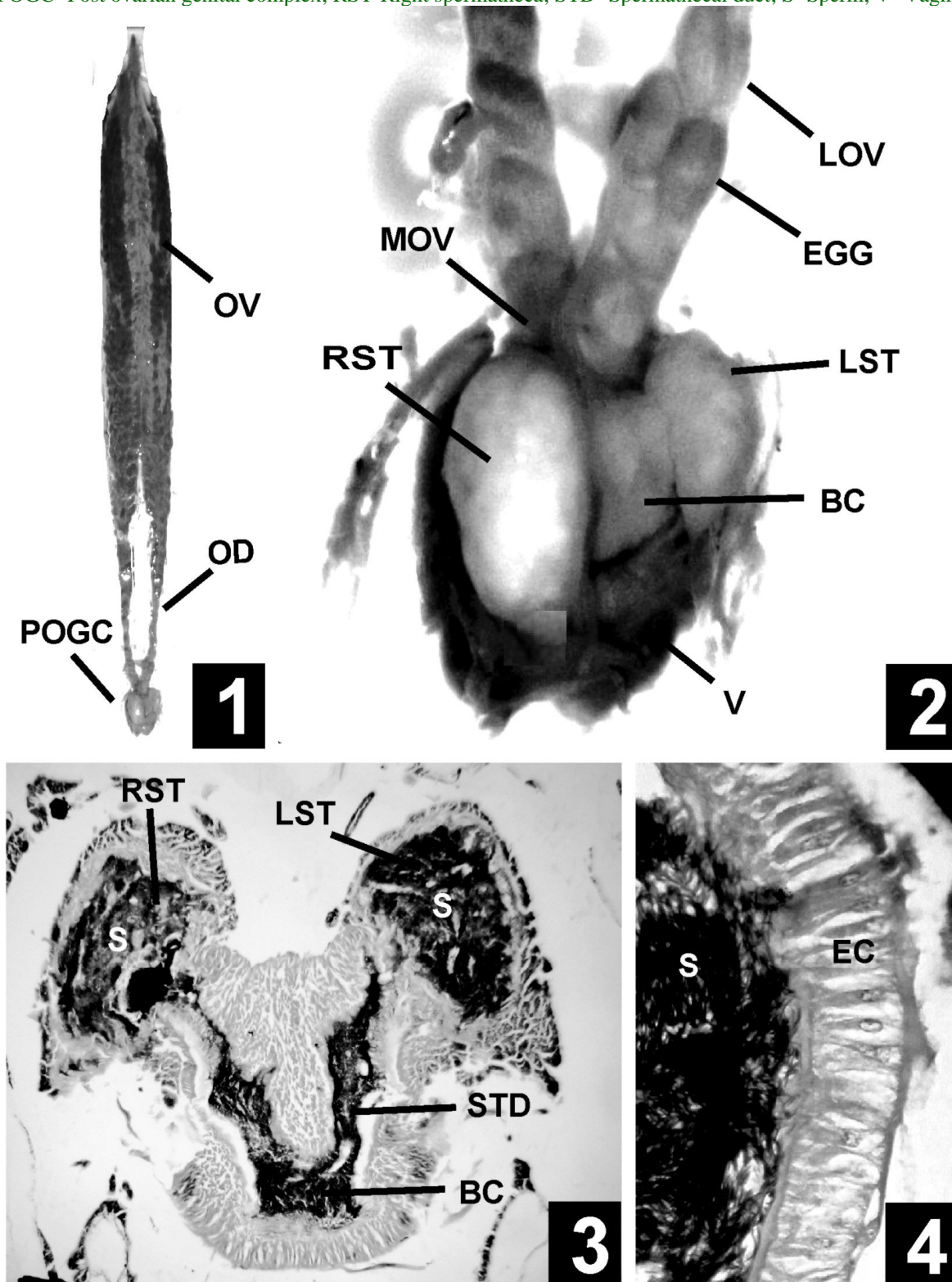
Fig. A: *Crocothemis servilia* female.



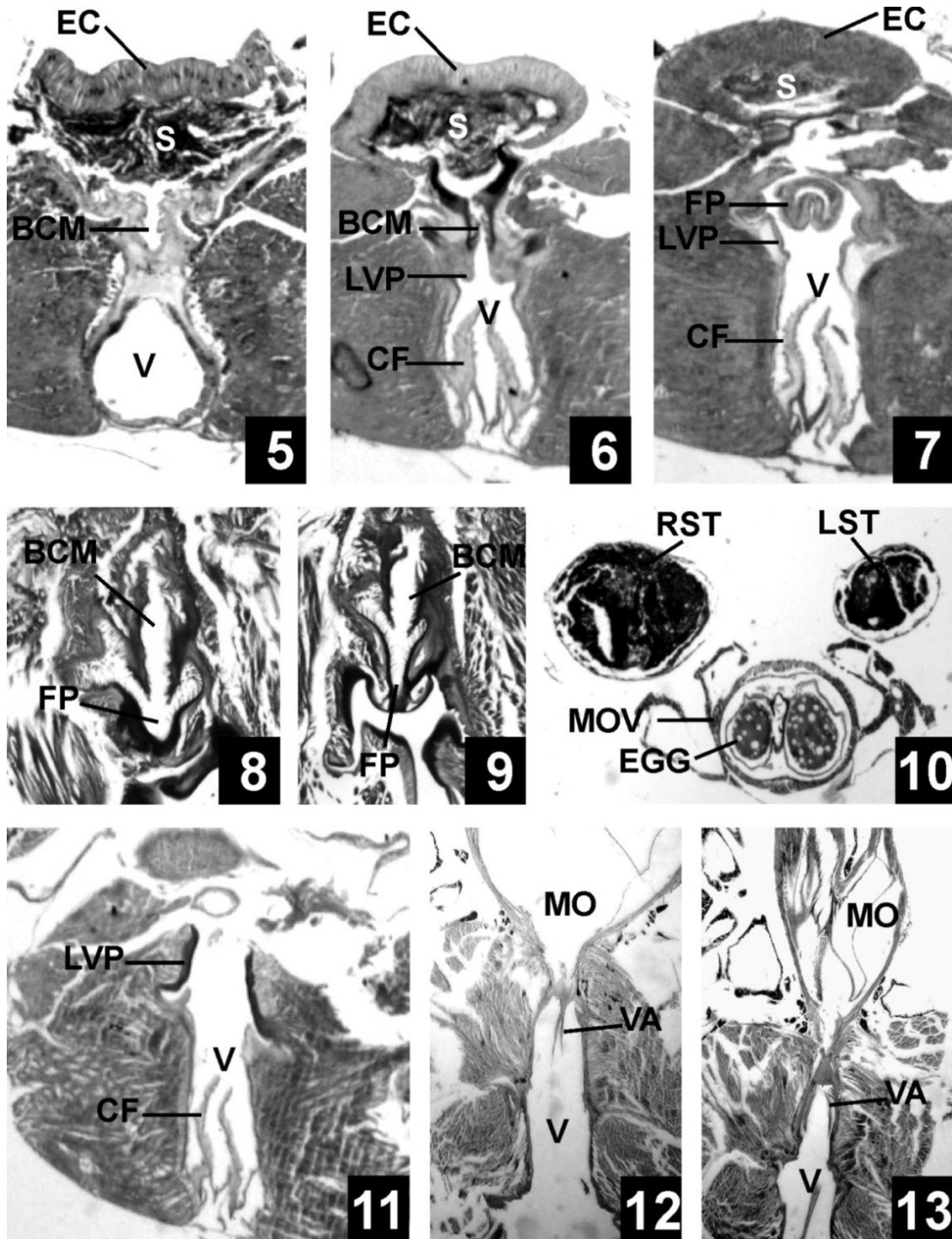
## Results

In *Crocothemis servilia*, the post ovarian genital complex (POGC) lies in the mid-region of the eight sternum below the hind-gut vestibule just behind the last abdominal ganglion. The complex is composed of a pair of very large prominent bean shaped spermathecae present along the antero lateral region which opens into a small bursa copulatrix through a very short spermathecal duct and a large ventrally placed vagina. The right spermatheca is almost double the size (720×420µm) of the left one (465×230µm). A median cuticular plate with arms embedded in muscles is present mid-dorsally on the POGC. The vagina occupies a large part of the complex and is firmly attached to the eight sternum by prominent muscle bands. The vagina receives the median oviduct anteriorly and posteriorly, it opens outside through a well defined female gonopore [Fig. B (1-2)].

**Fig. B (1-4):** 1: Female reproductive system of *Crocothemis servilia* i.e. ovary, oviduct and post ovarian genital complex (POGC) (x22). 2: Magnified view of POGC showing lateral oviduct, median oviduct, right spermatheca, left spermatheca, bursa copulatrix and vagina (x40). 3: Longitudinal section showing right and left spermatheca, spermathecal duct and bursa copulatrix filled with sperm (x45). 4: Transverse section showing epithelial cells and sperm filled bursa copulatrix (x330). [BC- Bursa copulatrix; EC- Epithelial cells; LOV- Lateral oviduct ; LST- Left spermatheca; MOV- Median oviduct; OV- Ovary; OD- Oviduct; POGC- Post ovarian genital complex; RST-Right spermatheca; STD- Spermathecal duct; S- Sperm; V- Vagina].



**Fig. B (5-13): Section passing through the post ovarian genital complex of the dragonfly *Crocothemis servilia*.** 5-7: Transverse sections showing the *Bursa Communis*, fertilization pore and vagina. Note the tall epithelial cells of the bursa copulatrix and sperm filled lumen, formation of *Bursa Communis* and lateral vaginal plates. Also note the cuticular folds of the vagina and hanging fertilization pore (x52). 8-9: Longitudinal section showing spiny *Bursa Communis* and fertilization pore (x160). 10: Transverse sections showing the unequal spermathecae and median oviduct filled with eggs (x40). 11: Transverse sections showing lateral vaginal plate and cuticular folds of the vagina (x40). 12-13: Longitudinal section of the vagina showing valves guarding the opening of median oviduct and vagina (x90). [BCM- *Bursa Communis*; CF- Cuticular folds; FP- Fertilization pore; LST- Left spermatheca; LVP- Lateral vaginal plate MOV- Median oviduct; RST- Right spermatheca; V- Vagina; VA- Valve].



The wall of the POGC is basically composed of an outer muscle layer, middle epithelial layer resting on a basement membrane and an internal layer of cuticle [Fig. B (3-13)].

### Spermathecae

The unequal sized spermathecae are very large prominent and oval in shape. The wall is composed of a thin indistinct epithelial cells followed by a thinner cuticular intima. The intrinsic outer muscle layer is also thin and scattered but thickens along the lateral side. The cuticular intima is thin and lightly striated longitudinally. Each open into the median bursa copulatrix antero-laterally by a well-defined short spermathecal duct which is covered by thicker intrinsic muscles. In the copulated females, the spermathecae appears bulbous and is filled with variable quantity of spermatozoa [Fig. B (3, 10)].

### Bursa copulatrix

The bursa copulatrix is a small, oval chamber with a triangular lumen since the dorsal region is sunk between the two spermathecae and lie just above the junction of the median oviduct and vagina. It has a small lumen measuring 220  $\mu\text{m}$  in length and 115  $\mu\text{m}$  in height. The dorsal wall of the bursa copulatrix is composed of very tall epithelial cells ( $54 \pm 8 \mu\text{m}$ ) [Fig. B (4, 5)] and the cells gradually shorten and become cuboidal along the latero-ventral region. The bursa copulatrix tapers along the postero-lateral margin to form the *Bursa Communis* [Fig. B (5, 6)]. The bursa copulatrix is externally covered with thick muscles of intrinsic origin [Fig. B (3)].

### *Bursa Communis* and fertilization pore

The opening between the bursa copulatrix and vagina is in the form of a cuticular *Bursa Communis* followed by a spiny tunnel shaped fertilization pore [Fig. B (5-9)]. The *Bursa Communis* contains short and sharp denticles all along its inner surface and around the inner and outer tips [Fig. B (8, 9)]. The fertilization pore is made up of three plates, a median-dorsal plate and paired lateral plates formed due to the thickening of the cuticle at the posterior tip of the *Bursa Communis* [Fig. B (7-9)]. The tunnel shaped fertilization pore extends downwards and opens along the roof of vagina. The initial region of the tunnel is inlaid with denticles but the distal region is highly modified into an inverted 'U' shaped tunnel [Fig. B (7)].

### Vagina

It is a short, tubular, cylindrical, chamber composed of outer thick muscular, middle epithelial and inner thin cuticular layers. The vagina is divided into the anterior, middle and posterior regions [Fig. B (5-7)]. The anterior region of the vagina is amalgamated with the descending median oviduct. In this region the epithelial layer of the median oviduct continues as the ventral wall of the vagina. The lumen is circular so that the egg can slip inside the vagina from the oviduct [Fig. B (5)]. The middle region of vagina exhibits prominent modification in its cuticular intima. At the base of this region the intima folds and forms a pair of cuticular undulating membrane which helps to hold the egg [Fig. B (6, 7)]. The cuticular of the lateral wall the lateral vaginal plates from are studded with mechanoreceptors and also support the hanging fertilization pore [Fig. B (6,7,11)]. The cuticular intima of posterior region is stubbed with microscopic pegs. It opens posteriorly in the sub genital plate of the ninth segment. The opening of the median oviduct and vagina is guarded by a pair of cuticular valves [Fig. B (11-13)].

### Discussion

Rathke (1832) for the first time observed and noted the post ovarian genital complex (POGC) in Odonata. Later, Siebold (1940) and Marshall (1914) described the POGC in some dragonflies. Tillyard (1917) described a pair of accessory glands on either side of the genital complex of *Petalura*. Asahina (1954) reported that in *Epiophlebia superstes*, the spermathecae is absent and the spermathecae glands are attached to the vagina which later identified as fused spermathecae (Siva-Jothy, 1987). In *Crocothemis servilia*, the POGC consists of a vagina, a single bursa copulatrix occupying the antero-dorsal position and receiving the short ducts of the paired lateral bulbous spermathecae. Richards and Davies (1977) mentioned that in Odonata the two oviducts are very short and open into a large pouch-like spermathecae in the eight segment but in *Crocothemis servilia* as in most dragonflies (Mastuda, 1976) both the lateral oviducts join to form a median oviduct before opening into the vagina.

The spermathecae are generally paired in Anisoptera and single or absent in Zygoptera (Tembhare and Andrew, 1986; Srivastava and Srivastava, 1986, 88; Siva-Jothy, 1987). In the dragonfly *Brachythemis leucosticte* and *B. lacustris* the paired spermathecae unite to form a

common duct which opens into the bursa copulatrix (Miller, 1982a). In *Crocothemis servilia*, the spermathecae are paired, very bulky and prominent. Each spermathecae is provided with independent short duct opening into the lateral region of the bursa copulatrix. In all the odonates the spermathecae if present in pairs are of equal size. The unequal size of the spermatheca as reported in *Crocothemis servilia* is unique and may probably help in the retention of pre-deposited sperms during sperm removal by preceding males (Corbet, 1999).

In anisopteran dragonflies, the bursa copulatrix differs in shape and size. It may be large as in *Trithemis arteriosa*, *Crocothemis erytraea*, *Sympetrum rubicundalum* (Waage, 1984), *Nesciothemis farinosa* (Miller, 1984) and *Sympetrum depressiusculum* (Siva-Jothy, 1987) of median size as in *Brachythemis leucoticta*, *Celithemis eponina* and *Brachythemis lacustris* (Miller, 1982a) or tiny as in *Orthetrum cancellatum* and *O. chrysostigma* or small but globular as found in *Tramea virginia* (Andrew and Tembhare, 1994). In Zygoptera, moreover the bursa copulatrix is quite large, triangular or spherical in shape (Waage, 1982; Miller, 1987). In *Crocothemis servilia* the bursa copulatrix is small and sunken between large spermathecae.

The median oviduct opens into the vagina anteriorly as found in *Brachythemis lacustris*, *Nesciothemis farinosa* (Miller, 1982a, b), *Lestes vigilax* (Waage, 1982), *Orthetrum chrysostigma* (Miller, 1984), *Ischnura elegans* (Miller, 1987) and *Orthetrum cancellatum* (Siva-Jothy, 1987). In *Crocothemis servilia*, the median oviduct opens anteriorly, although in *Tramea virginia*, *Somatochlora arctica* and *Celithemis elisa* it runs below and opens ventrally into the vagina (Midttun, 1976; Waage, 1984; Andrew and Tembhare, 1994). The bursa copulatrix of *Somatochlora arctica* is lined by cuticle with intricate folds (Midttun, 1976). Similar folding is observed in the bursa copulatrix of *Crocothemis servilia*. In *Somatochlora arctica*, Midttun (1976) stated "occupying the central area of the floor are two elevated structures lying close together and separated by a groove, which receives furrows from these structures. The groove and furrows, in addition to the ducts bursae, represents the connection with the vagina". This is termed as the ventral slit by Miller (1984) and *Bursa Communis* by Siva-Jothy (1987). In *Crocothemis servilia*, the *Bursa Communis* and the fertilization pore are well-distinct and these structures contain cuticular spines resembling that of other dragonflies (Midttun,

1976; Miller, 1984; Siva-Jothy, 1987; Andrew and Tembhare, 1994, 96, 97).

Siva-Jothy (1987) reported the presence of an extension of the vagina anteriorly under the bursa copulatrix in *Orthetrum cancellatum* and similar situation is also found in *Crocothemis servilia*. In *Somatochlora arctica*, the median oviduct opens into the mid-vagina through a slit like opening encircled with a cuticular rim (Midttun, 1976). This opening is situated just below the fertilization pore. In *Crocothemis servilia*, the vagina receives the median oviduct anteriorly and this opening too is guarded by a cuticular valve.

## Conclusion

The post ovarian genital complex (POGC) of the dragonfly, *Crocothemis servilia* (Drury 1773) is located in the eighth abdominal segment. It consists of a pair of very large and bulbous but unequal spermatheca, a small, sunken, dorsal bursa copulatrix and a large ventrally placed vagina. The POGC is ectodermal in origin with outer muscle layer, middle epithelial layer resting on a basement membrane and an internal layer of highly modified cuticle. The POGC is externally covered with intrinsic and prominent muscle bands. The opening between the bursa and vagina is in the form of a cuticular *Bursa Communis* and a spiny tunnel-shaped fertilization pore. The vagina is divided into anterior and posterior regions showing variation in their cuticular intima.

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