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The status of *Lytocestus latuensis* Kale & Kalshetty, 2020- a critical study and its placement under INCERTAE SEDIS

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Abstract-Kale and Kalshetty (2020)¹ while describing a new species of a Caryophyllaeid cestode *Lytocestus latuensis* (Lytocestidae: Caryophyllidae) from *Clarias batrachus* at Latur, Maharashtra inadvertently committed certain mistakes. The present authors have critically examined the details, pointed out the mistakes and have suggested re-examination of the species, undertake molecular characterization till then the species in question has been kept under "INCERTAE SEDIS".

Key words: *Lytocestus latuensis*, status, lacunae, 'incertae sedis'

INTRODUCTION

Caryophyllaeids are unique in having a single set of reproductive organs within a non segmented body utilizing oligochaetes as their intermediate host. These are said to have originated from acoelomate turbellarian larvae and branched out at the beginning of Paleozoic era as parasites of aquatic vertebrates Kulakovskaja and Demshin (1978)². These are having a unique morphology, evolutionary status, genetic stability and show high degree of endemism.

The cestodes are interesting because they show low fecundity yet are extremely successful showing abundance in fishes, which is on account of 'vitelline cells' which

can synthesize & store glycogen in the nuclei, a normal function- a unique phenomena in the animal kingdom Agarwal (1985)³.

The increase in glycogen without concomitant increase in the number of vitelline cells or in egg size could lead to prolonged period of infectivity & utilization of glycogen and not lipid as energy source. Glycogen being heavier than lipid, can be utilised under anerobic condition are consistent with life cycle that has benthic intermediate host Mackiewicz (1981a)⁴. Sahay *et al.* (2007)⁵ opined that higher level of glycogen in Caryophyllaeids is indicative of lower caloric value than lipid, which are the result of lower oxygen concentration in the enteric environment but low fecundity and relative abundance is due to K-strategies as an environmental condition.

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Out of the four families falling under Caryophyllidea, one of them is Lytocestidae Wardle & McLeod (1952)⁶. One of the genus belonging to Lytocestidae is *Lytocestus* Cohn (1908)⁷ to which 58 species have been reported. These are:-

1. *L.adhaerens* Cohn (1908)⁷
2. *L.filiformis* Woodland (1923)⁸
3. *L.indicus* Moghe (1925)⁹
4. *L.cunnigtoni* Fuhrmann *et al.* (1925)¹⁰
5. *L.chalmerisius* Woodland (1926)¹¹
6. *L.javanicus* Bovien (1926)¹²
7. *L.birmanicus* Lynsdale (1956)¹³
8. *L.alestes* Lynsdale (1956)¹³
9. *L.parvulus* Furtado (1963)¹⁴
10. *L.moghei* Murhar (1963)¹⁵
11. *L.longicollis* Ramadevi (1973)¹⁶
12. *L.lativitellarium* Furtado & Kim Low (1973)¹⁷
13. *L.fossilis* Singh (1975)¹⁸
14. *L.marathawadensis* Shinde & Phad (1988)¹⁹
15. *L.alii* Jadhav and Gavahne (1991)²⁰
16. *L.clariasae* Jadhav and Gavahne (1991)²⁰
17. *L.naldurgensis* Kadam, Hiware & Jadhav (1998)²¹
18. *L.chalishaonensis* Khalse & Shinde (1999)²²
19. *L.kopardaensis* Shinde & Borde (1999)²³
20. *L.teranaensis* Kolpuke, Shinde and Begum (1999)²⁴
21. *L.batrachusae* Pawar & Shinde (2002)²⁵
22. *L.clariasae* (minor) Pawar & Shinde (2002)²⁵
23. *L.govindae* Patil & Jadhav (2002)²⁶
24. *L.vishnupurensis* Shomendra *et al.* (2003)²⁷
25. *L.nagapurensis* Lakhe, Pawar & Shinde (2004)²⁸
26. *L.shindei* Khadap *et al.* (2004)²⁹
27. *L.assamensis* Tandon, Chakravorty & Das (2005)³⁰
28. *L.attenuatus* Tandon, Chakravorty & Das (2005)³⁰
29. *L.clariae* Tandon, Chakravorty & Das (2005)³⁰
30. *L.heteropneusti* Tandon, Chakravorty & Das (2005)³⁰
31. *L.bokaroensis* Poonam (2007)³¹
32. *L.majumdari* Poonam (2007)³²
33. *L.paithanensis* Shelke (2007)³³
34. *L.jagtai* Tripathi *et al.* (2007)³⁴
35. *L.punensis* Jadhav, Bhure & Padwal (2008)³⁵
36. *L.subhapradhi* Jawlikar, Pawar and Shinde (2008)³⁶
37. *L.moghei* Sharma (2009)³⁷
38. *L.murhari* Kaul & Suryavanshi (2010)³⁸
39. *L.folliculariae* Bhure *et al.* (2010)³⁹
40. *L.osmanabadensis* Bhure *et al.* (2010)³⁹
41. *L.shindei* Suryavanshi *et al.* (2010)⁴⁰
42. *L.vyasaei* Pawar & Hiware (2011)⁴¹
43. *L.purnensis* Pawar & Hiware (2011)⁴¹
44. *L.geriapinusae* Kadam *et al.* (2011)⁴²
45. *L.khami* Jawle *et al.* (2011)⁴³
46. *L.thapari* Sawarkar (2012)⁴⁴
47. *L.alii* (minor) Sawarkar (2012)⁴⁴
48. *L.manjaraensis* Solunki *et al.* (2012)⁴⁵
49. *L.rekhaensis* Nimbalkar *et al.* (2012)⁴⁶
50. *L.indica* Deshmukh (2015)⁴⁷
51. *L.godavariensis* Pawar (2016)⁴⁸
52. *L.mastacembellusi* Pardeshi (2016)⁴⁹
53. *L.ambae* Kankale (2017)⁵⁰
54. *L.paithanensis* Kale (2017)⁵¹
55. *L.bhartae* Patil (2018)⁵²
56. *L.elongatus* Barshe *et al.* (2018)⁵³
57. *L.laturensis* Kale & Kalshetty (2020)¹
58. *L.sahavi* Bhavsar (2020)⁵⁴

Hunter (1927)⁵⁵ transferred 4,5 into the genus *Monobothrioides* Gupta (1961)⁵⁶ accorded support.

Serial 8 was considered synonym of *L.birmanicus* (serial 7). Mackiewicz (1962)⁵⁷ however, considered it to be synonym of *L.filiformis* (serial 2). In *L.birmanicus* Lynsdale (1956)¹³ Mackiewicz observed 85-95 testicular follicles whereas Chakravorty & Tandon (1989)⁵⁸ recorded 170-384.

Serial nos 7,9,10,11,31 & 32 were considered by Ash (2012)⁵⁹ to be synonym of *Pseudocaryophyllaeus tenuicollis* Bovien (1926)¹². He also synonymised serial no. 12 & 27 with *Lucknowia microcephala* Bovien (1926)¹². Ash (2011a)⁶⁰ considered serial no. 14 synonym of *Pseudocaryophyllaeus ritai* Gupta & Singh (1983)⁶¹.

Most of the species are reported from the state of Maharashtra.

Ash (2011a, b, 2012)^{60,59} synonymised *L.heteropneusti* and *L.jagtai* (serial no. 30 & 34) with *Lucknowia fossilis* Gupta (1961)⁵⁶. *L.lativitellarium* Furtado & Kim Low (1973)¹⁷ & *L.assamensis* Tandon *et al.* (2005)³⁰ Serial no 12 & 27 were synonymised with *Lucknowia microcephala* Bovien (1926)¹²

Lytocestus alii Jadhav *et al.* (1991)²⁰, *Lytocestus clariasae* Jadhav *et al.* (1991)²⁰, *Lytocestus chalishaonensis* Shinde *et al.* (1999)²³, *Lytocestus kopardaensis* Shinde *et al.* (1999)²³, *Lytocestus naldurgensis* Kadam *et al.* (1998)²¹, *Lytocestus teranaensis* Kolpuke *et al.* (1999)²⁴, *Lytocestus batrachusae* Pawar & Shinde (2002)²⁵, *Lytocestus clariasae* (minor) Pawar *et al.* (2002)²⁵ (homonym), *Lytocestus govindae* Patil *et al.* (2002)²⁶, *Lytocestus nagapurensis* Lakhe *et al.* (2004)²⁸,

Lytocestus shindei Khadap *et al.* (2004)²⁹, *Lytocestus paithanensis* Shelke (2007)³³, *Lytocestus punensis* Jadhav *et al.* (2008)³⁵, *Lytocestus subhapradhi* Jawlikar *et al.* (2008)³⁶, *Lytocestus murhari* Kaul *et al.* (2010)³⁸, *Lytocestus shindei* Suryavanshi *et al.* (2010)⁴⁰ were considered synonyms of *Lytocestus indicus* Moghe (1925)⁹ by Ash (2012)⁵⁹

Sahay & Khalkho (2017)⁶² kept serial no. 49 [*L.rekhaensis* Nimbalkar *et al.* (2012)⁴⁶] under enquiry. Sahay *et al.* (2017)⁶³ revalidated *L.heteropneusti* Tandon *et al.* (2005)³⁰. Singh *et al.* (2018)⁶⁴ held *L.bishmupurensis* (serial 24) synonym of *L.indicus* Moghe (1925)⁹. Sahay *et al.* (2019)⁶⁵ kept *L.mastacembellusi* Pardeshi (2016)⁴⁹ a species under enquiry. Sahay & Ekka (2019)⁶⁶ considered *L.jagtai* Tripathi *et al.* (2007)³⁴ & *Lytocestus fossilis* Singh (1975)¹⁸ to be synonym of *Lucknowia fossilisi* Gupta (1961)⁵⁶.

The aim of the present study is to assess the status of *Lytocestus laturensis* Kale & Kalshetty (2020)¹ as the authors of the said species seem to have inadvertently committed mistakes while describing the species.

MATERIALS & METHODS

Several research papers have been consulted along with the literature of *L. laturensis* Kale *et al.* (2020)¹.

OBSERVATION & DISCUSSION

“*Lytocestus laturensis* Kale & Kalshetty (2020)¹ is supposed to show, following characters:- Head spatulate narrow than body, broad at the base narrow at the apex, neck medium in length wide narrow anteriorly, broad posteriorly with constrictions on the lateral side, testicular follicles 850-900 small oval, preovarian scattered in medullary region unevenly in 4-7 rows, not found in posterior 1/3rd region, vas deferens short, thin curved coiled. Cirrus pouch large, oval anteroposteriorly situated on left or right side & open through a genital pore tapering at anterior end curved. Cirrus thin curved slightly coiled within the cirrus pouch. Ovary bilobed butterfly shaped, medium H-shaped, lobes anteroposteriorly elongated with irregular margins, lobes spindle shaped broader in middle, tapering at both ends, uneven in size, poral lobe short, aporal lobe long, isthmus short, bag-pipe like wide, connecting lobes at 1/3rd from posterior end. Vagina thin, starts from genital pore open into ootype. Ootype large oval, antero-posteriorly elongated, **1-2 rows on each lateral side** (?). uterus tubular, 1-2 turns, wide loop shaped with 4-5 folds, narrow, open by uterine pore (oval, large double walled in middle of the worm)”.

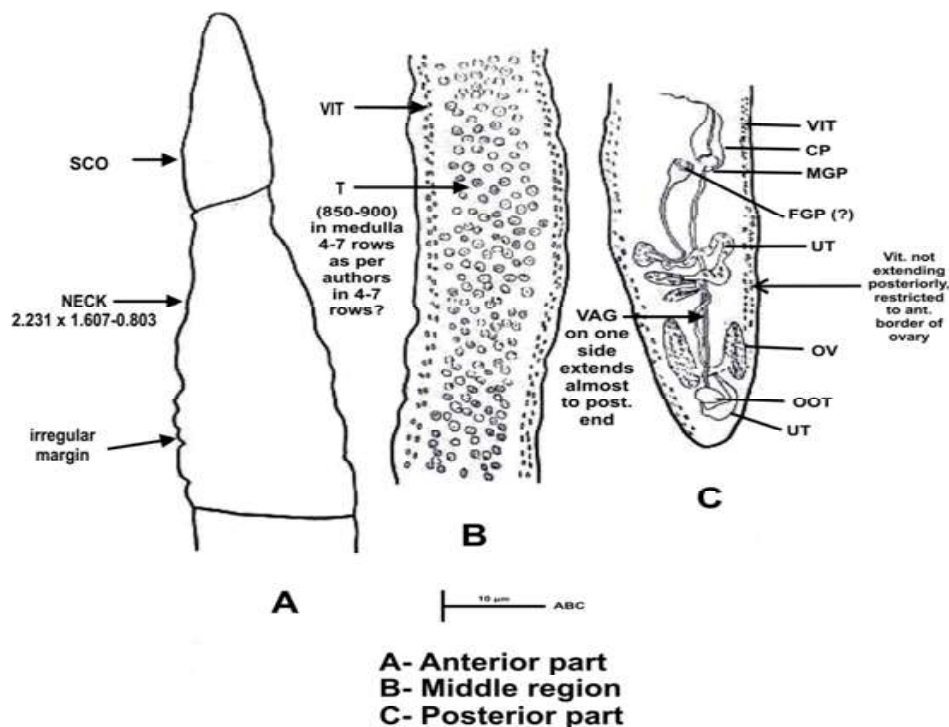


Fig.1- *Lytocestus laturensis* (Kale & Kalshetty, 2020)¹ from *International Journal of Entomology Research*. Volume 5 Issue 5 on page 63 (labeling done for convenience)

Lacunae in the paper:

There are a number of spelling mistakes in the said paper. In the abstract the authors wrote “fifteen species of cestode parasites were collected from the intestine of *Clarias batrachus*, the word “species” should have been replaced by ‘specimens’.

In case the authors were in possession of 15 specimens why there should be single measurement in the followings:

- a. Length & breadth- **4.973** x 0.446 – 0.603
- b. Head – spatulate **2.803** in length x 0.982 – 0.446
- c. Neck – **2.231** x 1.607 – 0.803
- d. Cirrus pouch – **0.893** x 0.338 – 0.178 in breadth
- e. Cirrus – **0.767** x 0.053
- f. Vas deferens – **0.535** x 0.035
- g. Ovary – **0.446** x 0.893
- h. Isthmus – at 1/3rd from post. end **0.357** x 0.089
- i. Uterus – **0.446** x 0.893

It seems that the authors studied only one specimen and the range in width in a,b,c etc in all probability was their convenience. If they had fifteen specimens they should have given range in the lengths & breadth of all the parameters, taken out the mean value, SD and CV. Because these could be used to define statistically the species. Ratios such as:

1. Commencement of vitellarial follicles and testicular follicles/ body length.
2. Position of ovary from posterior end/ body length
3. Position of cirrus sac from posterior end/ body length
4. Distance of genital pore from posterior end/ body length
5. Distance of ootype from posterior end/ body length

Of the above 5 parameters would have characterized *Lytocestus laturensis* (?) better than linear measurements.

The present authors are of the opinion that if only whole samples (of different populations of same species) are studied and quantitatively analysed, species determination would not remain entirely subjective.

The authors of the said species state “vagina is a thin tube, starts from the genital pore, runs medially and posteriorly, slightly curved, reaches and opens into the ootype and measures 0.357 in length and 0.213 in breadth.”

Mackiewicz (1972)⁶⁷ opines that- “vagina communicates between oviduct and the ventral surface (fig.

6) Caryophyllidea- a review. At its proximal end it may join the uterus to form utero-vaginal duct as in *C.laticeps* or *P.differtus*) opening posterior to the male gonopore or it

- may form the uterovaginal duct that also receives the ejaculatory duct thus forming a short distinct canal (hermaphroditic duct) that- terminates at a small, common gonopore (as in *Caryophyllaeids*, *Isoglaridacris* and *Biacetabulum*).
- A third condition (as in *Atractolytocestus* occurs when the ejaculatory duct and utero-vaginal duct open together or very close to the surface forming a shallow atrium with a single large gonopore through which one can see the male & female gonopore.)

Vagina never opens directly in ootype rather “posteriorly it drains into oviduct but it does not open independently on the ventral surface of body, usually it joins with the terminal part of the uterus to form an uterovaginal duct which communicates to the ventral surface of body as female pore”- Hafezullah (1993)⁶⁸.

Kale and Kalshetty (2020)¹ have not mentioned anything about vagina joining the uterus and forming the utero-vaginal duct which opens through uterovaginal pore is a serious lapse.

Developmental studies may reveal, which one develops first, the genital pore or the vagina. But this much is sure that vaginal’s posterior end does not open directly into the ootype rather swells to form receptaculum seminis which continues posteriorly & joins the oviduct, before joining the ootype. In *Lytocestus* species male and female genital openings opens separately.

Vitellaria: The distribution of vitellaria in *Lytocestus laturensis* (vide fig. 1c) has been shown to be unequal upto almost posterior end on one side whereas, the extension is upto anterior border of ovary. This needs clarification as regards its commencement (anteriorly) and termination (posteriorly).

Testes: The number of testicular follicles in *Lytocestus laturensis* has been shown to be 850-900 preovarian scattered in medullary region, unevenly distributed in 4-7 rows from the base of neck to the gonads, not distributed in posterior 1/3rd region of the worm.

The authors of the said species *Lytocestus laturensis* seems to have given conigence to the number of testicular follicles while separating it from other species of

Lytocestus [*L. filiformis* Woodland (1923)⁸, *L. indicus* Moghe (1925)⁹, *L. alestes* Lynsdale (1956)¹³, *L. birmanicus* Lynsdale (1956)¹³, *L. longicollis* Ramadevi (1973)¹⁶, *L. marathawadensis* Shinde & Phad (1988)¹⁹, *L. alii* Jadhav and Gavahne (1991)²⁰, *L. naldurgensis* Kadam *et al.* (1998)²¹, *L. teranaensis* Kolpuke *et al.* (1999)²⁴, *L. nagapurensis* Lakhe *et al.* (2004)²⁸, *L. assamensis* Tandon *et al.* (2005)³⁰, *L. kopardaensis* Shinde & Borde (1999)²³, *L. govindae* Patil & Jadhav (2002)²⁶, *L. murhari* Kaul & Suryavanshi (2010)³⁸, *L. geriapiusae* Kadam *et al.* (2011)⁴², *L. khami* Jawle *et al.* (2011)⁴³, *L. vyasaiei* Pawar & Hiware (2011)⁴¹ a total of 17 species.

Although it has already been pointed out by Sahay *et al.* 2019 that species identification on the basis of number of testicular follicles is questionable because the range depicts that the worm in question were not of the same age and that the worm were more than one when studied. If the worms are of the same age, the number of testicular follicles should normally remain more or less constant for a species.

It is surprising that the authors compared their specimens only with above 17 species & did not compare it with rest of the species. Not only that, out of the above 17 species

1. *Lytocestus* (*alii*, *naldurgensis*, *teranaensis*, *kopardaensis*, *murhari*, *govindae*, *assamensis*) have already been synonymized.
2. *L. marathawadensis* Shinde & Phad (1988)¹⁹, synonymized with *Pseudocaryophyllaeus ritai* Gupta & Singh (1983)⁶¹ by
3. *L. longicollis* Ramadevi (1973)¹⁶ was synonymized with *Pseudocaryophyllaeus tenuicollis* Bovein (1926)¹² by Ash (2012)⁵⁹
4. *L. khami* Jawle *et al.* (2011)⁴³, was kept by Sahay, *et al.* (2021)⁶⁹ under INCERTAE SEDIS

On the above grounds the present authors are left with the following alternatives:-

1. Invalidate *Lytocestus laturensis* as a new species.
2. Keep *Lytocestus laturensis*- a species under INCERTAE SEDIS
3. Request the authors of the paper to restudy the slides in their possession clarify the above points, provide photo-micrograph of the slides and provide cross sections. Take up proper identification of the cestode in question upto molecular level to ascertain their taxonomic status.

The molecular identification has been used for delineating species specially between cryptic and sister species- Nadler & Perez-Ponae de Leon (2011) otherwise traditional taxonomy based on phenotypes will fall into distress.

The present authors though support the 3rd option yet advocate till then that the said species be kept under "INCERTAE SEDIS".

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