UNIVERSITY GRANTS COMMISSION BAHADUR SHAH ZAFAR MARG NEW DELHI – 110 002

Title of the Project

Fine structural demonstration of olfactory neuroepithelium of *Pseudapocryptes lanceolatus* (Bloch and Schneider) with special reference to electron enzymology and X-ray microanalysis

F. No. - 41 - 161/2012 (SR) of dated 13.07.2012

[01.07.2012 - 31.06.2015 (EXTENDED UPTO 31.12.2015)]

FINAL REPORT

Submitted by

Prof. Subrata Kumar De

Principal Investigator

Department of Zoology Vidyasagar University Midnapore (West) – 721102



VIDYASAGAR UNIVE RSITY MIDNAPORE ★ WEST BENGAL ★ PIN 721102

Phone : (03222) 276554 :: 276555 :: 276557 :: 276558

Ref. No.

To Whom It May Concern

This is to certify that the grant of Rs. 11,13,809.00 (Rupees Eleven Lac Thirteen Thousand Eight Hundred and Nine only) received from the University Grants Commission under the scheme of support for Major Research Project entitled 'Fine structural demonstration of olfactory neuroepithelium of Pseudapocryptes lanceolatus (Bloch and Schneider) with special reference to electron enzymology and X-ray microanalysis' vide UGC letter No. F. 41-161/2012 (SR) dated 13.07.2012 to Prof. Subrata Kumar De, Department of Zoology, Vidyasagar University, Midnapore (West) - 721102, West Bengal has been fully utilized for the purpose for which it was sanctioned and in accordance with the terms and conditions laid down by the University Grants Commission. The Final Report of the project work is kept in the Central Library and the executive summary is placed on the website of Vidyasagar University, India.

Date: 25.7.17 Place: Midanapore

Date ..



Fax : (91) 03222, 275329, 275297 e-mail : vidya295@mail.vidyasagar.ac.in // website : url : http : // www.vidyasagar.ac.in

Annexure -III

UNIVERSITY GRANTS COMMISSION BAHADUR SHAH ZAFAR MARG NEW DELHI – 110 002

Annual Report of the work done on the Major Research Project

1. **Project report No.** 1st/2nd/3rd/Final : Final Report

2. UGC Reference No. : F. No. 41-161/2012 (SR) of

dated 13.07.2012

3. Period of report : from 01.07.2012 to 31.12.2015 (Project is extended up to 31.12.2015 as per letter Ref. No. – 41 – 161/2012 (SR) of dated 18.06.2015)

4. Title of research project : Fine structural demonstration of olfactory neuroepithelium of *Pseudapocryptes lanceolatus* (Bloch and Schneider) with special reference to electron enzymology and x-ray microanalysis.

121266251	1, 23233701, 23237731, 23234116 3, 23232317, 23236735, 23239437	FOTAR Dr. S. K. De PDW विषयविद्यालय अनुदान आयोग अहार बहादुरप्राह जफर मार्ग नई दिल्ली-110,002
F. No. 41-161/20	012 (SR)	Offices University GRANTS COMMISSION Vidyasagar University New DeLHI-110 002
The Under Secre	tary (FD-III)	time.
University Grant New Delhi-1100	s Commission 02	signature 13 JUL 2012
Sub:- UGC su Agricult	apport for the Major Res tural Sciences and Engin	earch Project in Physical Sciences, Hio-Sciences, Maths, Medical,
"Fine st	ructural damonatorit	reachers - Project entitled,

"Fine structural demonstration of olfactory neuropithelium of pseudapocryptes lanceolatus (Bloch and Schneider) with special reference to electron enzymology and X-ray microanalysis"

I am to refer to your letter forwarding the application of Dr. Subrata Kumar De of your institution for financial assistance under the above scheme and to convey the Commission's approval & sanction an on account grant of R s. 7,60,300/- (Rupees: seven lakh sixty thousand three hundred only) to the Registrar, Vidyasagar University, Midnapore-721102, WB in r/o Major Research Project of Dr. Subrata Kumar De, Department of Zoology for the period of 3 years w.e.f. 1.7.2012 as detailed below:-

A.	Non - Recurring	AMOUNT APPROVED	GRANT RELEASED AS Ist INSTALMENT	Categ ory
1.	Books & Journals Equipment (Microscope with camera attachments, digital balance)	45,000/- 2,62,000/-	3,07,000/-	GEN
5.	Kecurring Honorarium to Retd. Teacher @ Rs. 12, 000/- p.m. Project Fellow @14,000/- p.m. for initial 2 years and Rs. 16,000/- p.m. from the third year onwards. Chemical/ Glassware / Consumable Hiring Services Contingency Travel/Field Work Special Need Overhead Charges @ Rs. 10% approved recurring	nil 5,28,000/- 1,00,000/- 45,000/- 45,000/- 45,000/- nil 71:800/-	4,53,300/-	
1	Total (A + B)	11.41.800/-	7.60.3001	

The acceptance Certificate in prescribed format (Annexure-1 available on the UGC web-site) may be sent to the undersigned within one month from the issue of the award letter failing which the project may be treated as

If the terms & conditions are acceptable, as per guideline which are available on UGC web-site www.ugc.ac.in the Demand Draft/ Cheque being sent may be retained. Otherwise the same may be returned in original to the UGC by Registered Post in variably with in 15 days from the receipt of the Demand Draft/Cheque in favour of Secretary, UGC, New Delhi.

Principal Investigators should ensure that the statement of expenditure & utilization Certificate to the effect that the grant has been utilized for the purpose for which it has been sanctioned shall be furnished to the University Grants Commission in time.

The first instalment of the grant shall comprise of 100% of the Non -Recurring including Over Head Charges, and 50% of the total Recurring grant.

· · · ·
I. The sanctioned amount is debitable to the Major Hard 4 (f) a (31) Rs. 4.53,300/- & 4 (D
and is valid for payment during financial year 2012-13.
Grants Comment of the Grant shall be drawn by the Under Secretary (drawing and Disbursing Office)
Vidyasagar University Midaman 731102 While the Change Daniel Devide to the University of the Universit
3. The Grants is subject to the adjustment of the basic of Utilization Cartificate in Mail Transfer.
submitted by the University/Colleges/institution.
utilized calle and the grante with the second soft the expenditure out of the grante with
5. The Utilization Cartificate of the spenditure.
sanctioned shall be furnished to the University Great Great Great for the purpose for which it he
current financial year.
or easers acquired wholly or substantially out of University Grant Commission's event of the
sanctioned of utilized for the purposes other that those for which the grant shall not be disperse
function such assets shall forest and should, at any time the College/University
A Register of assets acquired wholly or which it is and a start commission.
8. The second state of the prescribed form
non-utilization shall ensure the utilization of grant-in-aid for which is the
unutilized amount from the simple interest @ 10% per annum as amound of
Rules of Govt. of India will be charged to the date of refund as per provisions contained in the to time to
The interest earned by the University/College/In the
10. The University in the Utilization Certificate/Statement of
from time to be furnished by grantee include the state of expenditure to be furnished by grantee include and
11. The University/Collocation of posts for Scheduled Control issued by the Government of the
Official Language Act 1962 and Complement to Official Language Policy of the Probability of the Complement of the Comple
The sanction issues in exercise of the deleast
13. An
No. F. 41-161/1012 come out the grant of
which it was sandicated has been utilized by University of Rs.
now we may enter Utilization Certificate for Rs.
No
It is also certified from the B.C.R. that the C.
ver land grant is sanctioned against the builds are available under the scheme Det
4.53 300/ 8.4 (D) Res
15. The funds to the extent of A. 3.07,000/- under the head of A. and thead of A. and thead of A. and the head of A. and thead
16. The University/Institution Calles inder the Scheme.
in Higher Educational Institutions 2000 100 following the UGC populat
the second
and lace of ragging
Copy forwarded Guine
(Kanta Bate) (Kanta Bate)
Deputy Secretary
Acknowledgement for the sector of the sector
Secretary, Finance Division III of DD / Cheque / Mail Transfer
2. Dr. Subrata Kumar De Principal
Vidyasagar University Midney Midney Midney Midney Department of the Under
3. office of the Director General of 721102, WB
Estate, New Delhi.
*. Ine Registrar, A. G. C. R. Building T. p.
or bit i P.
(h)
(Pramod Sharma)
Section Officer

5. (a) Name of the Principal Investigator	:	Dr. Subrata Kumar De			
(b) Deptt. and University/College where work has progressed:					
		Department of Zoology			
		Vidyasagar University			
		Midnapore (West) - 721102			
		West Bengal			
6. Effective date of starting of the project	:	01.07.2012			

7. Grant approved and expenditure incurred during the period of the report:

a. Total amount approved	Rs. 11, 41, 800.00/-
--------------------------	----------------------

Grant Received (First Instalment)	:	Rs. 7, 60, 300.00/-
Grant Received (Second Instalment)	:	Rs. 3, 53, 509.00/-
Total	:	Rs. 11, 13, 809.00/-

b. Total expenditure : Rs. 11, 13, 809.00/-

BANK OF INDIA 14.8.1 Relationship beyond banking... On From The Branch Manager, Office of the Registrar BAHADURSHAH ZAFAR MARG BRANCH Vidyasagar University Diais No. 2832.010. 138 Tel.: 23370617 "Hans Bhawan" 23370534 Bahadurshah Zafar Marg Tilak Bridge, New Delhi Pin-110002 Mar Monature 338 Ref.: BSZ/UGC/ 30, F. 2012 To. The Registrar/Principal gen Ur TC . Re Subject : TRANSFER OF FUNDS BY WAY OF NEFT-DEMAND DRAFT/RTGS/TRANSFER As per instruction of University Grant Commission, we have transferred a sum of MRS. Rs. by way of Transfer/NEFT/RTGS/DD No. for credit of your SB A/C No./DD No. 43101010100012829 431010100013829 Branch IFSC Code RTGS UTR No. Details of which are given below: $P.NO_{41} - 161 / 2 > 12 (SR)$ 1. UGC Sanction letter No. and Date: 13 - 7 - 2 > 122. Amount of the Grant: R. 7.60309/: 3. Purpose of the Grant: MRP-Pr. Subsch-K.De Dept. J Zoology 4. Financial year: 452012-13 Yours, Faithfully Asstt. General Manager Copy for information to : 1. Two copies for information to University Grant Commission 3. UGC Contact No. 23237721, 23234116, 23239437, 23236351

, Sukumar De, M C (Proprietor)	Com F.C.A		B-13/5	M/S Charter "N 4, Kalyani Nadi Ph. No e-mail - monhri	S. DE & CO. ed Accountants MONOROMA" a, Pin - 741235 033-25826288 de@gmail.com
	Dr. Subrata Vidya	Kumar De, Pro sagar University	f. of Zoology, Department of Zoo y, Midnapore, Pin-721102, WB.	logy	
	Receipts &	Payment Accou	ant of UGC Major Research Proj	ect.	
	(F	, No. 41-161/201	12(SR) of Dated 31.07.2012)		
eipts	Amount(Rs.)	Amount(Rs.)	Payments	Amount(Rs.)	Amount(Rs.)
Grant Received			By Non-Recurring Exp.		
from U.G.C			Books & Journals	48,637.00	
Memo No. F.No. 41	-161/2012		Microscope	2,57,896.00	2 10 022 00
(SK)			Digital Balances	11,500.00	3,18,033.00
dated 13/07/2012)			D. Desumine Fun		
Rooks & Journal	45 000 00		Honorarium to Project Fellow	2 32 581 00	
Equipments	2 62 000 00	3 07 000 00	Chemicals	19 234 00	
	2,02,000.00	3,07,000.00	Hiring Services	22 500 00	
Pocurring Crant P	aceived		Contingencies	10 972 00	
Hoporarium	2 64 000 00		Travel/Field Work	4 583 00	
Chemical	50,000,00		Overhead	71 800 00	3 91 670 00
Hiring Services	22,500.00		overnead		
Contingencies	22,500.00		By Balance c/d		50,597,00
Travel/ Field Work	22,500.00				
Overhead	71,800.00	4,53,300.00			
		7,60,300.00			7,60,300.00
Date : 14-01-2014 Place : Kalyani			Signed in terms of our utiliss For M /S S Chatered A	ation Certificate DE & CO Sountants	of even date 91 / 2014

	BOB UNIVERSITY GRA BAHADUR SHAH NEW DEL NAME OF THE Grant-in	NT COMMISSION H ZAFAR MARG HI 110002 SECTION: (SR) aid-Bill
1	Name of the Beneficiary Institution (University/ College/ Institute)	Vidyasagar University, Midnapore - 721102 (WB)
2	Sanction No. and Date	No.:- 41-161/2012/(SR) Dated:- F. D. Dy. No. 714 Dated: 12-05-2015
3	Amount being Released	(d)Sanctioned : Rs. 3,53,509/- (e)Adjusted : Rs. Nil (f)Net Release : 3,53,509/- (Rupees Three lakh fifty three thousand five hundred nine only)
4	Purpose of grant-in-aid	Major Research Project of
5	Head of Account	Dr. Subrata Rumar De, Deptt. of Zoology, 3(A)2202.03.102.10.01.31 49(a)
6	Designation and address of the Authorized Officer	The Registrar, Vidyasagar University, Midnapore - 721102 (WB)
7	Payment Details	
(a)	Bank Name & Address of Branch	UCO Bank, Vidyasagar University Branch, Midnapore Paschim Medinipore Pin- 721102
(b)	Account No	17480100003992
(c)	Type of Account : SB /Current /Cash Credit	SB
(d)	IFSC Code	UCBA0001748
(e)	MICR Code	721028303
(f)	Whether Bank Branch is RTGS or NEFT enabled : RTGS / NEFT /Both	YES
(g)	Name & Address of Account Holder	The Registrar, Vidyasagar University, Midnapore - 721102 (WB)
Recei being disbu	ved a sum of Rs.3,53,509/- (Rupees Three I amount sanctioned vide sanction letter N resement to The Registrar, Vidyasagar Univer	akh fifty three thousand five hundred nine only) 10.41-161/2012/(SR) dated (copy enclosed) for sity, Midnapore - 721102 (WB)

(

BARBHISIDSSS1042

Sukumar De, M.Com. F.C.A. M/s S. De & Co. rietor Chartered Accountants " MANORAMA" B-13/54, Kalyani, Nadia, Pin.-741235 Phone No. 033-25826288 E-mail -monhride@gmail.com Dr Subrata Kumar De, Prof. of Zoology, Department of Zoology Vidyasagar University, Midnapore (West), Pin-721102, W.B Receipt & Payment Account of UGC Major Research Project Period from 15.01.2014 to 30.06.2015 extended upto 31.12.2015. F, No. 41-161/2012 (SR) of 18.06.2015 ipt Amount (Rs.) Amount (Rs.) Payment Amount (Rs.) Amount (Rs.) rant Received From UGC lemo No. F.No. 41-161/2012/(SR) By Recurring Expenses ated. 18.06.2015 Honorarium to Project Fellow 2,48,928.00 ecurring Grant Received Chemicals 40,000.00 onor. to Project Fellow 1,91,225.00 Hiring Services 18,400.00 RA 68,284.00 2,59,509.00 Contingencies 22,500.00 hemicals 40,000.00 Traveling 21,600.00 iring Services 18,000.00 Postage and Printing & Audit 2,081.00 ontingencies 18,000.00 avel/Field Work 18,000.00 3,53,509.00 3,53,509.00 - Kalyani Sign in terms of our report of even date 16.11.2015 For M/s. S. DE & CQ Chartered Accounts MANORAM B-13/54 KALYANI (proprietors) EREDA 1 6 NOV 2015

Sukumar De, M.Com. F.C.A. M/s S. De & Co. rietor Chartered Accountants " MANORAMA" B-13/54,Kalyani,Nadia, Pin.-741235 Phone No. 033-25826288 E-mail -monhride@gmail.com Dr Subrata Kumar De, Prof. of Zoology, Department of Zoology Vidyasagar University, Midnapore (West), Pin-721102, W.B Receipt & Payment Account of UGC Major Research Project Period from 01.07.2012 to 30.06.2015 extended upto 31.12.2015 F, No. 41-161/2012 (SR) of 13.07.2012 pt Amount (Rs.) Amount (Rs.) Payment Amount (Rs.) Amount (Rs.) rant Received From UGC By Non-Recurring Expenses emo No. F.No. 41-161/2012/(SR) Books & Journal ated. 13.07.2012 48,637.00 Microscope on-Recurring Grant Received 2,57,896.00 Digital Balances ooks & Journal 11,500.00 3,18,033.00 45,000.00 quipment 2,62,000.00 3,07,000.00 ecurring Grant Received By Recurring Expenses onor. to Project Fellow 4,55,225.00 Honorarium to Project Fellow 5,23,509.00 RA 68,284.00 Chemicals emicals 89,234.00 90,000.00 Hiring Services 40,900.00 ring Services 40,500.00 Contingencies 35,993.00 ontingencies 40,500.00 Travel/Field Work 32,259,00 avel/Field Work 40,500.00 Overhead 71,800.00 erhead 71,800.00 8,06,809.00 Postage and Printing & Audit 2,081.00 7,95,776.00 11,13,809.00 11,13,809.00 Kalyani Sign in terms of our report of even date 16.11.2015 For M/s. S. DE & CO. Chartered Accounts (proprietors) 1 6 NOV 2015

SUKUMAR DE, M.Com, F.C.A. Proprietor

M/s S. DE & CO. CHARTERED ACCOUNTANTS "MONORAMA" B-13/54, Kalyani, Nadia. Pin - 741235 Phone no- (033)25826288 Mob - 9433342091 E-mail ID- monhride@gmail.com

Utilization Certificate

Certified that the grant of Rs.11,13,809.00 (Rupees Eleven Lac Thirteen Thousand Eight Hundred and Nine Only) received from the University Grants Commission under the scheme of support for Major Research Project entitled "Fine structural demonstration of olfactory neurepithelium of *Pseudapocryptes lanceolatus* (Bloch and Schneider) with special reference to electron enzymology and X-ray microanalysis" vide UGC letter No. F. No- 41-161/2012(SR) Dated 13.07.2012. The Grant has been fully utilized for the purpose for which it was sanctioned and in accordance with the terms and conditions laid down by the University Grant Commission.

Signature of the Registrar/Principal estig Statutory Auditor ator MARDE (Seal) (Seal) Registrar (Acting) VIDYASAGAR UNIVERSITY For, pology Departme ersity 721102 MIDNAPORE Vidyasagar M/S. S. DE & Midnapore (Wes Midia Chartered Accou West Bengal Signature of the Co-Investigator 1 6 NOV 2015

c. Report of the work done:

i. Brief objective of the project

Olfaction or sense of smell is important chemosensory modality of fish which is mediated through an anatomical organ *viz.*, olfactory apparatus. The olfactory apparatus can perceive variety of chemical odorants from the external environment and perform several biological functions like searching of food, recognition of sex, species identification, avoidance of predators, etc. The detail on this neurosensory organ is still hardly explored in most of the Indian teleosts including *Pseudapocryptes lanceolatus* (Bloch and Schneider), a teleostean gobiid of gangetic Bengal, India. The objective of this research work is enlisted as –

- Anatomical characterization of the olfactory apparatus of *P. lanceolatus* supported by medical analog X- ray radiograph study.
- Microanatomical study through semithin sections on the olfactory neuroepithelial cells under optical microscope.
- To study the surface topography of the olfactory components and water ventilation route using scanning electron microscope (SEM).
- Cellular component mapping with the help of transmission electron microscope (TEM).
- Histo- and electron enzymological studies to demonstate the activity of Acetyl choline esterase (AchE) in sensory cells.
- Organelle based elemental analysis of neuroepithelial sensory components and other neural derivatives of olfactory apparatus through X- ray microanalysis (EDX).

The ultimate objective of this study is to develop a feature map which proves the relevance of olfactory neuroepithelial cellular responses across the olfactory path.

ii. Work done so far and results achieved and publications, if any, resulting

from the work (Give details of the papers and names of the journals in which it

has been published or accepted for publication)

Research Abstracts:

1. Sarkar, S.K. and De, S.K. (2013) Neuroepithelial cells in olfactory apparatus of *Pseudapocryptes lanceolatus*: A mudskipper of Bengal. 24th All India Congress of Zoology & National Seminar on 'Biodiversity and its Management for Food, Livelihood & Environmental Security' & National Helminthological Congress. November 23-25, 2013. Organized by Department of Zoology, University of Kalyani, West Bengal in collaboration with Zoological Society of India, Bodh Gaya.

2. De, S. K., Acharya, A. Sarkar, S.K., Biswas, S. and Datta, N. C. (2013) Olfactory apparatus of Fish. 24th All India Congress of Zoology & National Seminar on 'Biodiversity and its Management for Food, Livelihood & Environmental Security' & National Helminthological Congress. November 23-25, 2013. Organized by Department of Zoology, University of Kalyani, West Bengal in collaboration with Zoological Society of India, Bodh Gaya.

3. Sarkar, S.K. and De, S.K. (2013) Role of rodlet cell in olfactory neuroepithelium of a fish (*Pseudapocryptes lanceolatus*). UGC- DRS Sponsored National Seminar on 'Bioprospecting of Natural Products'. December 5-6, 2013, Organized by Department of Zoology, The University of Burdwan, West Bengal. (Attended)

4. De, S.K. and Sarkar, S.K. (2014) Olfactory neuroepithelial cells use as a tool for aquatic health assessment. National conference on "Aquatic Ecosystems and their Management: Recent trends and future perspectives". March 21 – 23, 2014. Organized by Center for Environmental Sciences, Central University of Bihar, BIT Campus, Patna, Bihar.

5. De, S. K. and Sarkar, S.K. (2014) Crosstalk between olfactory sensory and nonsensory neuroepithelial cells of a mudskipper: *Pseudapocryptes lanceolatus* (Bloch and Schneider, 1801). National Seminar on Threats to Biodiversity and Ecosystems: Impacts of Developmental project and Climate Change & 25 th All India Congress of Zoology. November 17 – 19, 2014. Organized by Department of Zoology and Environmental Science, Gurukula Kangri University, Haridwar.

6. Sarkar, S.K. and De, S.K. (2014) Comparative morphoanatomy of olfactory apparatus in two different mud-dwelling teleosts. National Seminar on Threats to Biodiversity and Ecosystems: Impacts of Developmental project and Climate Change & 25 th All India Congress of Zoology. November 17 – 19, 2014. Organized by Department of Zoology and Environmental Science, Gurukula Kangri University, Haridwar.

7. De, S.K. and Sarkar, S.K. (2015) Role of macrophage in olfactory neuroepithelium of fish. National Conference on Applied Zoology in Sustainable Development: An update. January 31 – February 2, 2015. Organized by Department of Zoology, University of North Bengal (NBU), Siliguri.

8. Sarkar, S. K. and De, S.K. (2015) Anatomy and ultrastructural studies on chemosensory cells in an intertidal mudskipper (*Pseudapocryptes lanceolatus*) of South East Asia. National Conference on Applied Zoology in Sustainable Development: An update. January 31 – February 2, 2015. Organized by Department of Zoology, University of North Bengal (NBU), Siliguri.

9. Sarkar, S. K. and De, S. K. (2015) Fine structural modifications in olfactory neuroepithelium of *Pseudapocryptes lanceolatus* (Bloch and Schneider, 1801): An air breathing mudskipper of South East Asia. UGC Sponsored National Seminar on 'Zoological Research: Past, Present and Future'. March 26, 2015. Organized by Department of Zoology, Vidyasagar University, Midnapore (West) – 721102, West Bengal.

Research Papers (Published):

- Sarkar, S. K., Acharya, A., Jana, S. and De, S. K. (2014) Macroanatomical variation of the olfactory apparatus in some Indian teleosts with special reference to their ecological habitat. *Folia Morphologica (Warsz)*, **73** (2): 122 -128. [ISSN: 0015-5659] [Impact factor- 0.524].
- De, S. K. and Sarkar, S. K. (2014) Vesicular Diversity and Crowding Within the Olfactory Sensory Receptor Neuron. *Microsc. Microanal.* 20 (Suppl 3): 1272 – 1273. [Impact Factor – 2.495]
- Sarkar, S.K. and De, S.K. (2014) Functional anatomy of cellular junctions in olfactory neuroepithelium of *Pseudapocryptes lanceolatus* (Bloch and Schneider). *Indian Journal of Biological Sciences*, 20: 36-39. [ISSN: 0972-8503]
- Sarkar, S.K., Jana, S. and De, S.K. (2014) Age-specific anatomy and cytological studies on unilamellar olfactory structure of a teleosts (*Pseudapocryptes lanceolatus*). *European Journal of Experimental Biology*, 4 (6): 105-111. [ISSN: 2248-9215]
- Sarkar, S. K. and De, S. K. (2015) Acetylcholine esterase (AchE) activity in ciliated olfactory neuron of a teleostean: gobiid [*Pseudapocryptes lanceolatus* (Bloch and Schneider, 1801)]. *International Journal of Science and Nature*, 6 (3): 444 446. [ISSN: 2229 6441]
- Sarkar, S. K., Nag, T. C. and De, S. K. (2015) Ultrastructural Studies on the Nuclear Elements in Differentiating and Degenerative Ciliated Olfactory Neuron of *Pseudapocryptes lanceolatus* (Gobiidae: Oxudercinae). *Egyptian Journal of Basic and Applied Sciences*, 2 (2015): 295 – 302. [http://dx.doi.org/10.1016/j.ejbas.2015.07.004]

Research Papers (Communicated):

1. De, S. K. and Sarkar, S.K. (2015) Electron Microscope Based X-ray Microanalysis on Bioaccumulation of Heavy Metals and Neurodegenerative Dysfunctions in Mudskipper [*Pseudapocryptes lanceolatus*]. Journal of Microscopy and Ultrastructure.

iii. Has the progress been according to original plan of work and towards achieving the objective. if not, state reasons

Yes. The project is completed according to the original plan of work.

iv. Please indicate the difficulties, if any, experienced in implementing the project

No

v. If project has not been completed, please indicate the approximate time by which it is likely to be completed. A summary of the work done for the period (Annual basis) may please be sent to the Commission on a separate sheet. The project is completed within the given period of time (within 31.12. 2015).

vi. If the project has been completed, please enclose a summary of the findings of the study. Two bound copies of the final report of work done may also be sent to the Commission

This research program was focused on the advanced electron microscopical demonstration of olfactory neuroepithelium of Pseudapocryptes lanceolatus (Bloch and Schneider), a teleostean gobiid fish of South East Asia to explore macro - and microanatomical features of the neuroepithelial components in respect to various aspects of olfaction. The unilamellar olfactory apparatus of *P. lanceolatus* is located at the ethmoid region of the head. The olfactory lamella of *P. lanceolatus* along with its nasal cavity is well associated with anterior and posterior nostrils. The nasal cavity is surrounded by pseudostratified olfactory neuroepithelium. The cellular components are categorized as sensory receptor cells, supporting cells, basal cells, etc. The sensory receptor cells are bipolar neuron in nature and conveniently divided into dendron, perikaryon and axon. The tip of the dendron protrudes towards the nasal cavity to form olfactory knob. The morphoanatomy of olfactory knob denote structural variations among the sensory receptor cells. The perikaryons of different sensory receptor cells are located at the variable depth of olfactory neuroepithelium and characterized under transmission electron microscope (TEM). The axons of different sensory receptor cell travels towards the basal lamina. The variable morphometry of vesicles within the different subcellular compartments sensory receptor cells has been marked and concluded vesicular trafficking and crowding in bi-directional ends of the said neuron. These vesicles play important roles in neural signal transduction of olfactory signals. The ultrastructural features of acetylcholine esterase (AchE) activity in sensory receptor cells are also characterized under transmission electron microscope (TEM). Increasing frequencies of acetylcholine

esterase (AchE) containing vesicles at axoplasm may denote neural dysfunction of the olfactory neuron in response to aging or heavy metal toxicity. Effect of several heavy metals (e.g., iron, copper, nickel, lead, etc.) at subcellular level is analyzed through x-ray microanalyzer under transmission electron microscope (TEM). The supporting cells are columnar in nature and categorized as ciliated supporting cell and microvillous supporting cell. This fine structural detail on the microvillous supporting cell may be an indicative for neural protection of olfactory neuroepithelium. Small polygonal basal cells show various stages of cellular differentiation within the neuroepithelium and may acts as a progenitor cell of sensory receptor cell. During differentiation, the nucleus shows specific nature of chromatin condensation. The qualitative changes in condensation of euchromatin and heterochromatin structures are marked by using florescence microscope. Beneath the basal lamina of olfactory neuroepithelium, a zone of fila olfactoria is present and characterized with the presence of several axonal bundles, Schwann cells, fibroblast cells, collagen fibers, blood capillaries, etc. It is assume that the fila olfactoria is the site for axonal accumulation which leads to the formation of olfactory nerve tracts for transduction of olfactory signals.

vii. Any other information which would help in evaluation of work done on the project. At the completion of the project, the first report should indicate the output, such as

(a) Manpower trained	:	One
(b) Ph. D. awarded	:	One
(c) Publication of results	:	Research Abstracts: 09
		Research Papers (Published): 06
		Research Papers (Communicated): 01

1

(d) Other impact, if any

With the help of this financial assistance from UGC, New Delhi; I have educate the Post Graduate students in Zoology (first generation learner) and Ph.D. students of Jangal Mahal area to perform research work in advanced level on neuroscience of fish (*Pseudapocryptes lanceolatus*) and higher vertebrates respectively.

I

SIGNATURE OF THE PRINCIPAL INVESTIGATOR

DR. SUBKATA KUMAR DE PROFESSOR Department of Zoology Vidyasagar University Midnapore (West)-721102 West Bengal India

REGISTRAR/ PRINCIPAL

REGISTRAR VIDYASAGAR UNIVERSITY Midnapore

Annexure – VIII

UNIVERSITY GRANTS COMMISSION BAHADUR SHAH ZAFAR MARG NEW DELHI – 110 002

PROFORMA FOR SUBMISSION OF INFORMATION AT THE TIME OF SENDING THE FINAL REPORT OF THE WORK DONE ON THE PROJECT

1. NAME AND ADDRESS OF THE PRINCIPAL	:	Prof. Subrata Kumar De
INVESTIGATOR		
2. NAME AND ADDRESS OF THE INSTITUTION	:	Department of Zoology
		Vidyasagar University
		Midnapore (West) – 721 102
		West Bengal
3. UGC APPROVAL NO. AND DATE	:	F. No. 41 – 161/ 2012 (SR)
		Dated 13.07.2012
4. DATE OF IMPLEMENTATION	:	01.07.2012
5. TENURE OF THE PROJECT	:	01.07.2012 to 31.12.2015
		[Project is extended up to
		31.12.2015 as per letter Ref. No.
		- 41 - 161/2012 (SR) of dated
		18.06.2015]
6. TOTAL GRANT ALLOCATED	:	Rs. 11,41,800.00
7. TOTAL GRANT RECEIVED	:	Rs. 11,13,809.00
8. FINAL EXPENDITURE	:	Rs. 11,13,809.00
9. TITLE OF THE PROJECT	:	'Fine structural demonstration of
	olfact	ory neuroepithelium of
	Pseud	dapocryptes lanceolatus (Bloch and
	Schne	eider) with special reference to
	electr	on enzymology and x-ray
	micro	analysis'

10. OBJECTIVES OF THE PROJECT

Olfaction or sense of smell is important chemosensory modality of fish which is mediated through an anatomical organ *viz.*, olfactory apparatus. The olfactory apparatus can perceive variety of chemical odorants from the external environment and perform several biological functions like searching of food, recognition of sex, species identification, avoidance of predators, etc. The detail on this neurosensory organ is still hardly explored in most of the Indian teleosts including *Pseudapocryptes lanceolatus* (Bloch and Schneider), a teleostean gobiid of gangetic Bengal, India (Fig. 1). The objective of this research work is enlisted as –

2

- Anatomical characterization of the olfactory apparatus of *P. lanceolatus* supported by medical analog X- ray radiograph study.
- Microanatomical study through semithin sections on the olfactory neuroepithelial cells under optical microscope.
- To study the surface topography of the olfactory components and water ventilation route using scanning electron microscope (SEM).
- Cellular component mapping with the help of transmission electron microscope (TEM).
- Histo- and electron enzymological studies to demonstate the activity of Acetyl choline esterase (AchE) in sensory cells.
- Organelle based elemental analysis of neuroepithelial sensory components and other neural derivatives of olfactory apparatus through X- ray microanalysis (EDX).

The ultimate objective of this study is to develop a feature map which proves the relevance of olfactory neuroepithelial cellular responses across the olfactory path.

11. WHETHER OBJECTIVES WERE ACHIEVED (GIVE DETAILS)

Yes.

The proposed research program has achieved the ultimate objective within the given period of time. The detailed outcome results are furnished below -

> The macroanatomy supported by medial analog x-ray –

The olfactory apparatus of *Pseudapocryptes lanceolatus* is present at the preorbital region of the head. Anatomically the olfactory apparatus of *P. lanceolatus* is comprises of olfactory lamella, accessory nasal sacs (viz., ethmoidal sac and lachrymal sac), olfactory nerve tracts and olfactory bulb of the brain (Fig. 2 and Diagram I). P. lanceolatus shows two pairs of nostril viz., anterior nostril and posterior nostril (Diagram II and Fig. 7). In between anterior and posterior nostril the olfactory apparatus is present on either side of head. The anterior nostril is a tube-shaped structure. It is situated just behind the upper lip and projected anterior downwardly. The aperture of the anterior nostril is somewhat triangular in shape (Fig. 2). The posterior nostril is an oval shaped aperture. It is situated at the anterior edge of the eye. Apparently the diameter of the aperture of posterior nostril is much larger than the anterior nostril. The posterior nostril is bordered by a low ridge of skin (Fig. 7). Nasal flaps are absent in both pair of nostrils. The olfactory apparatus of P. lanceolatus has a single olfactory lamella at either side of the head i.e., unilamellar olfactory apparatus and olfactory rosette is absent in this species (Fig. 2 and Diagram I). The olfactory lamella is an elongated, tube-like, sickle shaped structure (Figs. 2 and Diagram I). The anterior end of the olfactory lamella is well connected with anterior nostril. The caudal part of the olfactory lamella is partly guarded by olfactory chamber. The inside of the olfactory lamella shows a canal - like, tubular nasal cavity which extends from the anterior tip to base of the olfactory lamella (Fig. 8). There are two type of accessory nasal sacs viz., ethmoidal sac and lachrymal sac are present in the olfactory apparatus of P. lanceolatus (Fig. 2). These sacs are bulbous in shape and well connected with olfactory lamella at different location viz. ventrocaudal and dorsocaudal region respectively (Diagram I). The ethmoidal sac is slightly larger than the lachrymal sac. The olfactory chambers are cup – shaped bony structure present at the ethmoid region of head. These chambers are paired and well associated with nasal bones (viz, mesethmoid, lateral ethmoidal and pre-ethmoid bones) (Figs. 3, 4, 5 and 6). The olfactory nerves are originated from the base of olfactory lamella (Fig. 2 and Diagram I). These are paired structure and run relatively long distance and connected with olfactory bulb of the fore brain (Fig. 2 and Diagram I).



Fig. 1– The photograph of *Pseudapocryptes lanceolatus* (Bloch and Schneider) (→): a teleostean: gobiid fish of coastal Bengal, India.

[Not to scale]



Fig. 2 – The macroanatomical work indicates olfactory lamella (OL), olfactory nerve tracts (ON), eye (E), olfactory bulb (OB), cerebral hemisphere (CH), optic lobe (Op L), cerebellum (CB) and medulla oblongata (MO), *etc.* in *P. lanceolatus*.



Diagram I– The diagrammatic representation of the olfactory apparatus and brain of *P. lanceolatus* showing olfactory lamella (OL), ethmoidal sac (ES), lachrymal sac (LS), olfactory nerve tracts (ON), olfactory bulbs (OB), cerebral hemisphere (CH), optic lobe (OPL), cerebellum (CB) and medulla oblongata (MO), *etc.*



- **Figs. 3 and 4** The medical analog x-ray radiograph indicates the lateral and dorsal ossified parts (arrows) *i.e.*, cranial and post cranial skeleton of *P. lanceolatus*.
- Fig. 5 The x-ray photograph shows different type of bones *viz.*, premaxilla (PM), maxilla (M), mandible (MB), lachrymal (L), parietal (P), post parietal (PP) and neurocranium (N) at the cranial region of *P. lanceolatus*.
- Fig. 6 The association of bones in the ethmoid region of *P. lanceolatus* [olfactory chambers (OC), ethmoid (ETH), lacrimal (L), premaxilla (PM), maxilla (M), neurocranium (N), parietal (P) and post parietal (PP), *etc.*]

Scanning electron microscopical study

P. lanceolatus shows two pairs of nostrils viz., anterior nostril and posterior nostril (Diagram II and Fig. 7). The anterior nostrils are short, tube like structure. The length of the anterior nostril in adult P. lanceolatus is ranges from 350 µm. - 400 µm. The average diameter of the aperture in anterior nostril is measured about 50 µm. - 100 µm. It is projected anterior and downwardly (Fig. 7). The posterior nostrils are oval shaped aperture and obliquely placed, located at the anterior edge of the eye. The diameter ranges from 200 µm. - 300 µm. (Fig. 7). In between the anterior and posterior nostril, the olfactory apparatus is present just beneath the dorsal surface of skin. The olfactory lamella of the unilamellar olfactory apparatus is an elongated, tube like structure (Fig. 8). The longitudinal sections of the olfactory lamella indicate that the tubular channel of nasal cavity at the inside of the olfactory lamella (Fig. 8). This cavity is extended from the anterior tip to the posterior part of the olfactory lamella (Fig. 8). The nasal cavity is lined by olfactory neuroepithelium (Fig. 8). The surface of the olfactory neuroepithelium of P. lanceolatus indicates the presence of numerous olfactory knob of different sensory receptor cell (Fig. 9). These cells are distributed throughout the olfactory neuroepithelium. The olfactory knobs of the ciliated sensory receptor cell possess cilia. The number of the cilia ranges from 4 to 6. The average length of the cilia is 5 µm. - 8 µm. (Fig. 9). In microvillous sensory receptor, the olfactory knob shows several microvilli (Fig. 9). The structural detail on the apical tip of the crypt cell is not well marked in the olfactory neuroepithelium of P. lanceolatus under scanning electron microscope (SEM). The tufts of cilia are radiating from the apical flat surfaces of columnar ciliated supporting cell and easily identified at the interspaces of sensory receptor cells (Fig. 9). The number of the cilia ranges from 10 to 12 and densely projected from each ciliated supporting cell. The average length of the cilia is 6 µm. - 12 µm. The density of the cilia of ciliated supporting cell is apparently greater at the lateral part of the olfactory neuroepithelium. A narrow zone of the polygonal microridges of microvillous supporting cell are present in the sensory epithelium. The average width of the microridges is 0.2 µm. In this region some mucous secretory pores are remarkably noted. The average diameters of these pores are measured as $1.5 \,\mu$ m. - $1.8 \,\mu$ m.



Diagram II - The diagrammatic representation shows the position of nostrils of *P. lanceolatus* (Anterior nostril and posterior nostril).

[Not to

scale]

Fig. 7 - The scanning electron micrograph indicates anterior nostril (AN) and posterior nostril (PN). [The anterior nostril (AN) is a tube shaped structure located just behind the upper jaw and the posterior nostril (PN) is an oval shaped structure situated at the anterior edge of the eye].

Fig. 8 - The longitudinal section of the olfactory lamella reveals that the nasal cavity (NC) is lined by olfactory neuroepithelium (*).



Fig. 9 - The surface topography of the olfactory neuroepithelium of *P. lanceolatus* [olfactory knob with cilia: ciliated sensory receptor cell (→); olfactory knob with microvilli: microvillous sensory receptor cell (>); apical flat surface with cilia: ciliated supporting cell (CSC), mucous secretory pore (MP), *etc.*]

The microanatomy through semithin sections

The transverse section of the olfactory lamella distinctly shows pseudostratified olfactory neuroepithelium and fila olfactoria (Fig. 10). This neuroepithelium (thickness: 40µm - 70µm) completely covers the nasal cavity and does not show any secondary folding (Fig. 10). The olfactory neuroepithelium of P. lanceolatus comprises of different types of cell viz, sensory receptor cells, supporting cells, rodlet cell, basal cell, etc. (Diagram III). These cells are morphologically distinct and intermingled within the olfactory neuroepithelium. Sensory receptor cells are bipolar neuron in nature and possess dendron, perikaryon and axon (Diagram III). The dendron of the sensory receptor cells reached to the lumen of the nasal cavity. The terminal part of the dendron swells to form olfactory knob. There are three different types of sensory receptor cells present within the olfactory neuroepithelium of P. lanceolatus viz., ciliated sensory receptor cell, microvillous sensory receptor cells and crypt cell (Diagram III). Apparently the length of the dendron differs among these sensory receptor cells. The olfactory knob of the ciliated sensory receptor cell possesses cilia. The perikaryon of the ciliated sensory receptor cell is located at the lower part of the olfactory epithelium (Diagram III). The shape of the nucleus is ranges from round to oval. The olfactory knob of the microvillous sensory receptor cell shows several microvilli. The length of the dendron is moderate. The perikaryon of microvillous sensory cell is present at the middle part of the olfactory neuroepithelium (Diagram III). Nucleus is deeply stained and spherical in nature. It is located at the middle and upper region of the cytoplasm. The crypt cell possesses very short dendron. The perikaryon of the crypt cell is present at the apical part of the olfactory epithelium (Diagram III). The perikaryon of sensory receptor cell is a pearshaped structure. The cytoplasm of crypt cell is remarkably electron dense in nature. Nucleus is located at the central position and spherical in shape. The axon of the sensory receptor cells including ciliated sensory receptor cells, microvillous sensory receptor cells and crypt cells respectively are runs towards the basal lamina. Supporting cells are columnar in nature (Diagram III). The apical portion of the supporting cells is generally flat and broad. Cytoplasm is faintly stained. There are two type of supporting cells has been identified viz, ciliated supporting cell and microvillous supporting cell. In ciliated supporting cell, the apical part shows several cilia (Diagram III). Nucleus is round or oval. It is placed at the central position of the cell. Cytoplasm is less granulated. The apical portion of the microvillous supporting cell bears numerous microvilli (Diagram III). Round shaped nucleus is present at the above of the central portion. Basal cells are small and polygonal in shape (Diagram III). These cells are situated just above the basal lamina *i.e.*, the basal region. The



Fig. 10 – The transverse section of olfactory lamella is showing pseudostratified olfactory neuroepithelium (OE) and fila olfactoria (FO) enclosing the nasal cavity (NC).

Diagram III - The diagram represents of the pseudostratified olfactory neuroepithelium and fila olfactoria of *P. lanceolatus*. [Not to scale] basal cells constitute 3 to 4 cellular layers from the basal lamina towards the middle of the olfactory neuroepithelium (Diagram III). Round nucleus is located centrally and occupying the maximum area of the basal cell. The differentiating stages of basal cells into sensory receptor cells are observed under light microscope (LM). At the basal region, few macrophage cells are identified (Diagram III). Rodlet cells with different morphometry are marked throughout the olfactory neuroepithelium of P. lanceolatus, (Diagram III). The rodlet cell is commonly oval to elongate in shape. Under light microscope (LM), two distinct cytoplsmic zones are clearly noticed within the rodlet cell *i.e.*, outer dense exoplasm and inner faintly stained cytoplasm. The cytoplasm contains several dark spots (rodlets). Beneath the basal lamina, the zone of fila olfactoria is present (Diagram III). This very zone is characterized with aggregation of axons of different sensory receptor cells, collagen fibers, fibroblast cells, blood vessels with erythrocyte cells, blood cells, etc. (Diagram III). The axon of different type of receptor cells are invading the basal lamina and aggregated at this region (Diagram III). The fibroblast cells are frequently observed in this ground region. Fibroblast is a polarized cell and tapering at both ends. Nucleus is elliptical in shape and centrally placed (Diagram III). Numerous collagen fibres are present at this region around the axonal bundles (Diagram III). Blood vessels and erythrocytes are well marked in this zone (Diagram III).

> Transmission electron microscopical study

The transverse section of olfactory lamella of *P. lanceolatus* shows distinct pseudostratified olfactory neuroepithelium and fila olfactoria.



A. OLFACTORY NEUROEPITHELIUM

1. SENSORY RECEPTOR CELL

The sensory receptor cells are bipolar neuron and may be conveniently divided into three distinct regions – dendron, perikaryon and axon (Fig. 12). The dendron of the sensory receptor cell is a long, slender process (Fig. 12). Generally the apical tip of the dendron is projected into the lumen of the nasal cavity by forming olfactory knob (Fig. 11). The olfactory knob is a protrusion of dendron and shows structural variation among the sensory receptor cells. On the basis of morphological demarcations, sensory receptor cell are of three types – ciliated sensory receptor cells, microvillous sensory receptor cells and crypt cell (Figs. 12, 19 and 23). The perikaryons of these sensory receptor cells are located at the different strata of the olfactory neuroepithelium of *P. lanceolatus*. The axons are also long, slender structure, arises from the perikaryon and toward the basal lamina. The subcellular characters of these distinct regions *viz.*, dendron, perikaryon and axon in three types of sensory receptor cells *i.e.*, ciliated sensory receptor cell, microvillous sensory receptor cell and crypt cell are described below –



Fig. 11 - The electron-micrograph shows the apical part of the olfactory neuroepithelium of *P. lanceolatus* which is very close to the nasal cavity (*). The ciliated sensory receptor cells are noted. The olfactory knob (\rightarrow) of the ciliated sensory receptor cell, cilia (C) with distinct basal body and prominent centriole (CE), different type of cellular junctions *i.e.*, tight junctions (TJ), desmosomes (D) are identified under transmission electron microscope (TEM)

i) CILIATED SENSORY RECEPTOR CELL

Ciliated sensory receptor cells are frequently present within the olfactory neuroepithelium of *P. lanceolatus*. The dendron of the ciliated sensory receptor cell is very long and run towards the lumen of the nasal cavity (Fig. 12). The apical tip of the dendron bulges to form the olfactory knob, projecting into the lumen of the nasal cavity (Fig. 13). The olfactory knob of ciliated sensory receptor cell is a very pronounced structure and bears 4 - 6 numbers of cilia. The average diameter of the olfactory knob of this cell is ranges from 300 nm. - 900 nm. (Fig. 13). Each cilium is a long, slender structure and supported by several microtubules (Fig. 13). The longitudinal section of the cilia indicates a large number of small vesicles along with the longitudinal axis of microtubules. The microtubules of the cilium are arranged in (9+2) pattern (Fig. 12). Each cilium has a basal body (Figs. 13). The basal body is devoid of rootlets (Fig. 13). The basal body of cilia is present in the cytoplasm of long dendron and located just beneath the plasma membrane (Figs. 13). It is an integrated structure and associated with centriole (Fig. 13). The transverse sections of the centriole shows that nine groups of microtubules are arranged in a circle. Each group is a triplet formed of three tubules that are skewed towards the centre (Fig. 13). Several neurofilaments are associated with the centriole. The neurofilaments having about 10nm of diameter are frequently present throughout the cytoplasm of dendron, perikaryon and axon of ciliated sensory receptor cell. Several small vesicles are also observed along with axis of the neurofilaments (Fig. 14). The cytoplasm is granulated. The elongated, slender mitochondria are arranged within the cytoplasm of dendron region (Fig. 14). The matrix of the mitochondria is quite dense. Clear cristae are marked. The perikaryon of the ciliated sensory receptor cells is located in the lower portion of the olfactory neuroepithelium due to presence of long dendron. The perikaryons are spindle-shaped structure. Several mitochondria are arranged at the apical part of the perikaryon (Fig. 15). Spherical nucleus present at the middle of the perikaryon (Figs. 16 and 17). Euchromatin and heterochromatin materials are well distinguishable under transmission electron microscope (TEM). Heterochromatin materials are scattered at the peripheral region of the nucleus (Fig. 16). Nucleo-pores are clear (Fig. 16). The diameter of the nucleopores ranges from 100nm. - 150nm. The peripheral cytoplasmic region of the nucleus shows several short cisternae of rough endoplasmic reticulum (rER) with numerous granulated ribosomes (Figs. 17 and 17a). Although free ribosomes are also distributed throughout the



- Fig.12 -The ciliated sensory receptor cell possesses a long dendron (*). The apical tip of the
dendron bulges to form olfactory knob (\rightarrow) associated with cilia (C).
- Fig. 13 The olfactory knob bears cilia (\rightarrow) with basal body (B) associated with centrioles (CE).
- Fig. 13 a -The cross section of each centriole is showing clockwise direction of the triplet
tubules (\rightarrow). A globular condensation is found at the tip of each triplet.
- Fig. 14 The electron micrograph shows cytoplasm of the dendron in ciliated sensory receptor cell. Elongated mitochondria (M), microtubules (MT), neurofilament (>) and small vesicles with different diameter (\rightarrow) are noted


Fig. 15 - The apical cytoplasmic region of the perikaryon in ciliated sensory receptor cell (a large aggregation of mitochondria (M), multivesicular body (\rightarrow), polyribosomes (>), *etc.*]

Fig. 16 - The electron micrograph shows elliptical nucleus (N), placed centrally in the perikaryon of ciliated sensory receptor cell. Distinct nuclear membrane (>), with characteristic nucleopore (\rightarrow) , a prominent association of free ribosomes (*) at the distal part of the perikaryon is also noted.

Fig. 17 - The perinuclear polyribosomal condensation (>), rough endoplasmic reticulum (RER) are marked in the cytoplasm of perikaryon of the ciliated sensory receptor cell in *P. lanceolatus*.

Fig. 17 a - The electron micrograph shows distinct 80S ribosomes (\rightarrow) and respective subunits in a sporadic appearance in the perikaryon of ciliated sensory receptor cell.

Fig. 18 - The photomicrograph indicates a Golgi complex (G) with its distinct axis. The different diameter of secretory vesicles with prominent condensation (\rightarrow) , coated vesicles (*) are well identified. Microtubule (MT), neurofilament (NF), polyribosomes (>) are also characterized.

cytoplasm of perikaryon but are densely found in the lower part of the perikaryon *i.e.*, the axon hillock region. In this region polyribosomes are identified (Fig. 17). The Golgi complex with prominent cis-trans axis is present in the cytoplasm of the perikaryon (Fig. 18). The Golgi complex is well associated with several secretory vesicles (Fig. 18). The axon of the ciliated sensory receptor cell runs towards the basal lamina (Figs. 28 - 30). The axon of different sensory receptor cells are characterized with several mitochondria, neurofilaments, microtubules and large number of small vesicles *etc.*. The distal part of the axon of sensory receptor cell shows a terminal swelling, known as synaptic knob (Fig. 30). The synaptic knob contains several synaptic vesicles. The diameter of these vesicles ranges from 40µm. - 50µm. (Fig. 30).

ii) MICROVILLOUS SENSORY RECEPTOR CELL

This is another type of receptor cell found in the olfactory neuroepithelium of P. lanceolatus and also possesses prominent dendron, perikaryon and axon. The length of the dendron is comparatively shorter than ciliated sensory receptor cell (Figs. 19 and 20). The dendron runs towards the free surface and projected into the lumen of the nasal cavity (Fig. 19). The olfactory knob possesses several microvilli (Fig. 19). The average length of the microvilli is measured around 500nm. It is a compact structure and represents cytoplasmic processes covered by plasma membrane (Fig. 19). The cytoplasm of the dendron is granulated. The apical part of the cytoplasm in dendron shows numerous neurofilaments and microtubules (Fig. 20). Elongated mitochondria with prominent cristea are generally observed in the cytoplasm of the dendron of microvillous sensory receptor cell. Large number of mitochondria with different morphometry is aggregated at the upper portion of the perikaryon (Fig. 20). The perikaryon of the microvillous sensory receptor cell is present at the middle part of the olfactory neuroepithelium of *P. lanceolatus*. The perikaryon possess large and spherical nucleus with two concentric membranes separated by a perinuclear space (Figs. 21 and 22). At certain points of nuclear envelop is interrupted by prominent nucleopores (Fig.). The average diameter of nucleopores is ranges from 100nm. to 150nm. The euchromatin materials are higher than the heterochromatin materials (Figs. 21 and 22). The cytoplasm of the perikaryon also shows several microtubules and neurofilaments (Fig. 20). Golgi body with several secretory vesicles is marked within the perikaryon (Fig. 20). The distal part of the perikaryon shows large number of rough endoplasmic reticulum (rER) associated



Fig. 19 - The olfactory knob of the microvillous sensory receptor cell (>) shows microvilli (\rightarrow). The cytoplasm of the dendron indicates neurofilament (NF) and microtubule (MT).

Fig. 20 - The electron micrograph shows the apical part of the perikaryon of microvillous sensory receptor cell [Golgi body (G), several secretory vesicles (\rightarrow) with different diameter, lysosome (L), multivesicular body (MVB), *etc.*]



Fig. 21 - The middle part of the perikaryon indicates neurofilament (*), polyribosomes (>) and small vesicles (\rightarrow). The ultrastructure of the nucleopore (NP) is clearly marked within the nucleus of microvillous sensory receptor cell.

Fig. 22 - The distal part of the perikaryon of microvillous sensory receptor cell shows association of rough endoplasmic reticulum (RER) numerous free ribosomes (R) and polybosomes (>).

with free and polyribosomes (Figs. 21 and 22). Small vesicles, primary and secondary lysosomes are also found in this region. The axons of the microvillous sensory receptor cells show few mitochondria, neurofilaments and several vesicles of different diameter. The axons are collectively runs towards the basal lamina.

iii) CRYPT CELL

Crypt cells are present at the apical part of the olfactory epithelium of *P. lanceolatus* (Fig. 23). These cells are ovoid or pear-shaped structure (Fig. 23). The crypt cell possesses two distinct morphological parts viz., the perikaryon and axon. The length of the dendron of the crypt cell is very short (Fig. 23). The apical tip of the perikaryon in crypt cell is equipped with microvilli and sunken cilia (Fig. 23). Crypt cell has a prominent apical invagination. The bottom and lateral parts of the invagination shows sunken cilia. The cilia show (9+2) arrangement of microtubules. The basal body of these cilia is not well marked in *P. lanceolatus*. Microvilli are present around the cilia on the apical rim of the crypt cell (Fig. 23). The lateral margin of the perikaryon shows longitudinal arrangement of numerous neurofilaments (Fig. 24). Like other type of sensory receptor cells *i.e.*, ciliated sensory receptor cell and microvillous sensory receptor cell, a large number of vesicles are associated with the axis of neurofilaments (Figs. 25 and 26). The cross sections of neurofilaments and multivesicular bodies are also observed (Fig. 25). Mitochondria are evenly distributed within the cytoplasm (Fig. 26). Mitochondria show different morphometry. Cristae are well marked and the mitochondrial matrix is quite dense (Fig. 26). Spherical nucleus is present at the lower-middle part of the cytoplasm. The nucleomembrane with nucleopore, nucleolus, etc. are very prominent. The euchrmatin materials are greater than the heterochromatin material (Figs. 26 and 27). At the peripheral part of the nucleus shows rough endoplasm reticulum (rER), free ribosomes, polyribosomes, secretory vesicles, etc. (Figs. 37 and 38). Golgi complex with distinct secretory axis is clear (Fig. 27). Several phases of secretory vesicles are identified within the cytoplasm of crypt cell (Fig. 27). The cytoplasm is more electron dense than other type of sensory receptor cells. Apparently the secretory vesicles are large at the lower part of the perikaryon *i.e.*, area of axon hillock. The long axon of the crypt cell runs towards the basal lamina. Some of the microvillous supporting cells with several mucous droplets are marked in association with crypt cell (Fig. 24).



Fig. 23 – The electron micrograph shows crypt cell (CC) within the olfactory neuroepithelium of *P. lanceolatus*. The apical tip of the cell possesses microvilli (*) and sunken cilia (\rightarrow). Supporting cells with numerous mucous droplets (>) at their apical cytoplasmic part are also noted.



Fig. 24 - The perikaryon in crypt cell indicates numerous neurofilaments (\rightarrow) with several vesicles (>).

Fig. 25 - The cytoplasmic part within the crypt cell indicates several types of vesicles of variable diameter (\rightarrow) , mitochondria (M), polyribosomes (>), *etc*.

Fig. 26 - The perinuclear cytoplasm of the crypt cell shows rough endoplasmic reticulum (RER) and Golgi apparatus (G). Secretory vesicles (arrows) are showing phases of maturation.

Fig. 27 - The spherical chromatinized nucleus (N) with prominent nucleolus (NO) is centrally located within the cytoplasm of the perikaryon of crypt cell. The nucleomembrane is associated with ribosomes (\rightarrow). The sporadic clusters of polyribosomes (>) are also noted.





Fig. 29 - The ultrastructure shows axonal accumulation in the olfactory neuroepithelium (\rightarrow) . The axonal bundle penetrates the basal lamina (BL). Beneath the basal lamina, several collagen fibers (>) are observed.

Fig. 30 - The photomicrograph is a terminal synaptic knob which possesses several synaptic vesicles (\rightarrow) of different diameters.

2. SUPPORTING CELL

Supporting cells are columnar in nature and are well distributed within the olfactory neuroepithelium of *P. lanceolatus*. Generally these cells are having broad apical surface. Supporting cells are extended from the free surface of the nasal cavity to the basal lamina. There are two types of morphologically distinct supporting cells are present within the olfactory neuroepithelium of *P. lanceolatus i.e.*, ciliated supporting cells and microvillous supporting cells.

i) CILIATED SUPPORTING CELL

This type of supporting cells is frequently marked within the olfactory neuroepithelium of P. lanceolatus (Fig. 31). The apical surface of ciliated supporting cells is broad and flat (Fig. 31). The flat surface of this type of supporting cell bears several cilia (Fig. 31). The number of cilia ranges from 10 to 12. The cilia are supported by microtubules (Fig. 31). Cilia are associated with basal body and are present just beneath the plasmamembrane (Fig. 31). The basal body shows striated rootlets (Fig. 31). Although neurofilaments are absent in the cytoplasm of ciliated supporting cell, but the microtubules are frequently marked. Numerous mitochondria are aggregated at the upper portion just beneath the basal body of the cilia in ciliated supporting cells (Fig. 31). Mitochondria show different shape with dense matrix and clear cristae (Fig. 31). A series of electron dense glycogen particles are marked at the peripheral cytoplasm of mitochondria (Fig. 31). The spherical nucleus is present at the upper part of the cell (Fig. 31). Chromatin materials are packed within the nucleus. Euchromatin and heterochromatin materials are moderately present within the nucleus (Fig. 31). In few ciliated supporting cells, the nucleus is present at the lower middle part of the cell. At the peripheral region of the nucleus shows glycogen droplets (Fig. 31). The diameter of these particles ranges from 20nm. – 30nm. Glycogen droplets are also distributed at the apical cytoplasmic region (Fig. 31). Rough endoplasmic reticulum (rER) polyribosomes and free ribosomes are scattered at the peripheral region of the ciliated supporting cell (Fig. 31). Desmosomes are also well noticeable in between the ciliated supporting cells and adjacent sensory receptor cell respectively (Fig. 31).



Fig. 31 - The electron photomicrograph indicates the columnar ciliated supporting cell with distinct cellular organelles *viz.*, cilia (C), basal body with striated rootlets (\rightarrow), mitochondria (M), rough endoplasmic reticulum (RER), ribosomes (R), glycogen droplet (>), *etc.*

ii) MICROVILLOUS SUPPORTING CELL

The microvillous supporting cells are also columnar in nature and extended from the apical surface to the basal lamina of the olfactory neuroepithelium of P. lanceolatus. The flat free surface of microvillous supporting cell bears several microvilli (Fig. 32). It is a finger - like projected structure and lined by plasma membrane (Figs. 32 and 32a). The plasma membrane shows distinct glycocalyx (Fig. 32a). The cytoplasm is less granulated and more electron lucent. The apical portion of the cytoplasm shows densely packed smooth endoplasmic reticulums (sER) (Fig. 32). Some granulated particles are also noticed at the peripheral part of the smooth endoplasmic reticulum (sER) (Fig. 32). Membrane bound vesicles are also present at upper portion of the cytoplasm (Fig. 32). These vesicles have different diameters and vary from 10nm. to 40nm. Vesicles are showing gradual increasing diameter towards the plasmamembrane (Fig. 32). Mitochondria are scattered within the cytoplasm of microvillous supporting cell (Fig. 32). Rounded and large nucleus is located at the middle part of the microvillous supporting cells. Outer and inner nuclear envelop with perinuclear cisternae (width 100 nm. to 200 nm.) and nucleolus is clearly observed. A series of ribosomes are also marked at the nuclear membrane. Euchromatin and heterochromatin materials are well demarcated. Heterochromatin materials are evenly distributed within the nucleus. The peripheral region of the nucleus shows several rough endoplasmic reticulum (rER) with free ribosomes. Polyribosomes and Golgi complex are also present at this region (Fig. 33). Glycogens are also noticed in the cytoplasm of microvillous supporting cell. The cytoplasm of the microvillus supporting cell shows several microtubules.

3. BASAL CELL

Basal cells are usually round, oval or pear-shaped structure (Fig. 34). These cells are loosely arranged at the base of the olfactory neuroepithelium and form 2 - 3 continuous layers. The polygonal basal cells are lie close to basal lamina (Fig. 34). This cell shows large nucleus. The cytoplasm possesses mitochondria, rough endoplasmic reticulum (rER), free ribosomes, cytoplasmic granules, *etc.* Mitotic stages are found in this layer of polygonal basal cells. There are two distinct types of cells are marked just above the layer of dividing basal cells within the olfactory neuroepithelium *viz.*, electron dense basal cell and electron lucent basal cell. The cytoplasm of the electron dense basal cells is more granulated than the electron lucent basal cell. The nucleus of electron lucent



- Fig. 32 The electron micrograph shows the apical part of the microvillous supporting cell [microvilli (→), smmoth endoplasmic reticulum (SER), secretory vesicles (>), etc.]
 Fig. 22a The microvilli of microvillous supporting cell shows distinct shoesely (>).
- Fig. 32a -The microvilli of microvillous supporting cell shows distinct glycocalyx (\rightarrow).



Fig. 33 – The perinuclear cytoplasm of the functional microvillous supporting cell shows an association of rough endoplasmic reticulum (RER), Golgi apparatus (G) with secretory vesicles (\rightarrow) , free ribosomes (R), polyribosomes (>), *etc.* Apart from that, a distinct chromatinized nucleus (N) with prominent nucleopore (NP) is also identified within the microvillous supporting cell.



Fig. 34 - The small polygonal basal cells are present at the basal region just above the basal lamina of the olfactory neuroepithelium of *P.lanceolatus*. The presence of prominent nucleus (N), clusters of mitochondria (M), enormous free ribosome (\rightarrow) and cytoplasmic granules (>) are the notable characters of the basal cell.



Fig. 35 - The electron micrograph demonstrate the gradual morphological changes in electron lucent and electron dense (arrows) basal cell within the olfactory neuroepithelium of *P. lanceolatus.*

basal cell is rich in euchromatin materials but in electron dense basal cell heterochromatin materials are scattered on euchromatin material. Free ribosomes are present at the peripheral region of the nucleomembrane. The morphology of the electron lucent and electron dense basal cell shows gradual changes within the olfactory neuroepithelium of *P. lanceolatus* (Fig. 35).

B. FILA OLFACTORIA

The fila olfactoria of *P. lanceolatus* is situated just beneath the basal lamina of the olfactory neuroepithelium (Fig. 36). Thus region is characterized with aggregations of axons of different sensory receptor cells, Schwann cell, collagen fibers, fibroblast cells, blood capillaries with erythrocyte cells, etc. (Figs. 36, 37, 38 and 39). A large number of axons belong to different categories of sensory receptor cells are aggregated within the fila olfactoria (Fig. 38). The transverse sections of the axons are roughly circular and unmyelinated (Fig. 38a). Prominent presence of Schwann cell is marked at the peripheral region of unmyelinated axonal bundles (Fig. 38). At the terminal part of each axon shows knob like structure with large accumulation of synaptic vesicles (20nm. to 40nm). The fibroblast cells are most frequent in this region (Fig. 36). It is polarized, fusiform cell and tapering at both ends (Fig. 37). Elliptical nucleus is present at the central portion of the cell (Fig. 37). Chromatins are well demarcated in this cell. Mitochondria are mainly found at the perinuclear region of the cytoplasm (Fig. 37). Free ribosomes, poly ribosomes, rough endoplasmic reticulum (rER), Golgi complex are well noted (Figs. 36 and 37). Tropocollagen fibers (having average diameter of 1.5nm.), are well marked with fibroblast cell (Fig. 37). Blood capillaries are thin walled structure which is most abundant within the fila olfactory of P. lanceolatus (Fig. 39). The diameter of the blood capillary is 8µm -10µm (Fig. 39). The capillary wall is lined by thin endothelium. The endothelial cells are resting on basal lamina of the blood vessels which is supported by loose network of reticular fibers (Fig. 39). The erythrocyte contains chromatinized elliptical nucleus.



Fig. 36 - The electron micrograph shows fila olfactoria of *P. lanceolatus* [axonal bundles (AB), fibroblast cells (FC), collagen fibers (\rightarrow) , etc.]. The hemidesmosomes (>) are well marked on the basal lamina.

Fig. 37 - The electronmicrograph shows the collagen units (\rightarrow) which is one of the constituent of fila olfactoria.



Fig. 38 - The ultrastructure shows the axonal bundles (AB) surrounded by Schwann cell (*) which are the notable features of fila olfactoria of P. *lanceolatus*.

Fig. 38a - The electron micrograph indicates prominent microtubule (>) and neurofilament (\rightarrow) within the axon of *P. lanceolatus*.



Fig. 39 - The vascular elements *i.e.*, the blood capillary, erythrocytes (*), leucocytes (>), endothelial cell are identified under transmission electron microscope (TEM) within the fila olfactoria in *P. lanceolatus*.

> Electron enzymological study on acetylcholine esterase (AchE) activity

The olfactory sensory receptor neuron (OSRN) is bipolar in nature. The acetylcholine esterase (AChE) positive ciliated olfactory sensory receptor cells are distinctly identified under light microscope (LM) and transmission electron microscope (TEM) (Figs. 40, 41 and 42). This cell shows various morphometry of vesicles within the different sub-cellular compartments. These vesicles are classified into small vesicles, small dense core vesicles, pleomorphic vesicles, coated vesicles and synaptic vesicles, etc. The small vesicles (10nm - 20nm) are mostly distributed at the proximal region of the perikaryon, cytoplasm of the dendron along with microtubules (20nm - 25nm) and neurofilaments (7nm -10nm) axonemal region of each kinocilium respectively (Figs. 41 and 42). These vesicles are subsequently crowded near the centriole of the basal body of each kinocilium. The frequency of small dense core vesicles (30nm - 40nm), coated vesicles (60nm - 70nm) and synaptic vesicles (70nm - 90nm) are higher within the axoplasm of olfactory sensory receptor cell (Fig. 42). These vesicles are characteristically docked and fused with the synaptic cleft (20nm - 25nm) of the synaptic knob of OSRN. The egg-shaped perikaryon of this type of cell shows positive reaction against acetylthiocholine iodide as substrate. The dendroplasm does not show any positive reaction in ciliated olfactory sensory receptor cell of neuroepithelium. The electron micrographs are also showing intense reaction within the perinuclear cytoplasm of ciliated olfactory sensory receptor neuron. The vesicles having diameter (30nm to 40nm) are acetylcholine esterase (AChE) positive (Fig. 42). These vesicles are observed within perinuclear cytoplasm, close to Golgi apparatus (Fig. 42). The aggregation of these vesicles is largely noted at the terminal part of axon in ciliated olfactory sensory receptor neuron (Figs. 42). Recent experimental studies indicate that the use of acetylcholine esterase (AChE) inhibitors is helpful for treatment of neurodegenerative disease. Thus, the detail knowledge on acetylcholine esterase (AChE) activity may be convenient for diagnosis as well as drug design for neurodegenerative disorders like Alzheimer's diseases, Parkinson disease, etc.





Fig. 41: The electron micrograph shows a part of perinuclear cytoplasm. Dense core vesicles (diameter 30nm. – 40nm.) are showing positive reaction (arrows) using acetylthiocholine esterase as substrate.

Fig. 42: Accumulation of dense core vesicles (diameter 30nm. – 40nm.) is marked under TEM at the terminal axoplasm of ciliated olfactory sensory receptor neuron in *P. lanceolatus*.

X-ray microanalysis of heavy metal components

Bioaccumulation of heavy metals and cytological consequences in olfactory sensory receptor neuron (OSRN) of Pseudapocryptes lanceolatus [collected near Tribeni (22.99°N 88.40°E) of West Bengal, India] has been studied under transmission electron microscope (TEM) (Table 1 and Table 2). The ecological condition of Ganga River near Tribeni is highly influenced with large amount of sewage and effluents including inorganic and organic pollutants. In this study, we have emphasized on the site specific cytological changes in ciliated OSRN within the olfactory neuroepithelium in P. lanceolatus with special reference to electron enzymology [for acetylcholinesterase (AChE) activity] and X-ray microanalysis [for bioaccumulation of heavy metals]. Ultrastructural details regarding cilia, olfactory knob, dendroplasm, perikaryon and axoplasm of ciliated OSRN have been clearly demarcated (Figs. 43 44 and 45). Frequent occurrences of degenerating ciliated OSRN with distinct features *i.e.*, lysis of plasma membrane at olfactory knob, disintegration of cytoskeletal structures in perinuclear cytoplasm and axoplasm, fragmented chromatin fibers with granules (diameter: 20nm to 30nm) in nucleoplasm, etc. are detailed in P. lanceolatus. Large accumulation of copper (Cu: 94.50%) is measured under TEM-EDX in cytoplasm of olfactory knob in degenerating ciliated OSRN of *P. lanceolatus* (Table 1). Excess bioaccumulation of iron (Fe: 83.81%) in nucleoplasm in degenerating ciliated OSRN (Table 2); probably associated with neuronal death. Crowding of acetylcholinesterase (AchE) positive vesicles (diameter: 30nm to 40nm) at terminal part of axoplasm are assume to be reflected by large accumulation of heavy metals in degenerating ciliated OSRN of *P. lanceolatus* (Fig. 46). Therefore, this baseline electron microscopical study on fish ciliated OSRN may be a prerequisite for monitoring the environmental health as well as metallobiology of several neurodegenerative disorders in fishes caused by bioaccumulation of heavy metals.



Fig. 43 - The olfactory neuroepithelium of *P. lanceolatus* is shows necrotic olfactory knob of ciliated olfactory sensory neuron (stars). Degenerating cilia (c), microtubules (arrows), mitochondria (m), various phases of lysosomes (arrowheads), *etc.* are also marked within the apical cytoplasm of ciliated olfactory sensory receptor cell in *P. lanceolatus*.



Fig. 44 - The perikaryon of ciliated olfactory neuron in *P. lanceolatus* shows necrotic features of nucleus (star) under transmission electron microscope (TEM). Condensation of chromatin granules (gr) and fragmented chromatin fibers (arrows) are also noted within the nucleoplasm. Numerous granules like structures (arrowheads) are also frequently distributed at the peirnuclear cytoplasm of ciliated olfactory sensory receptor cell in *P. lanceolatus*.



Fig. 45 - The electron micrograph shows necrosis of fila olfactoria (star) in the lamina propria of olfactory neuroepithelial system in *P. lanceolatus* (Group II). The cytoplasm of axons is less granulated with disintegrated ^{cytoskeletal} structures (arrows).





Table 1 - The X-ray microanalysis report indicates a part of apical cytoplasmic region (star) near ciliary apparatus at olfactory knob and bioaccumulation of heavy metal components in ciliated olfactory receptor cell in olfactory neuroepithelium of *P. lanceolatus*.





12. ACHIEVEMENTS FROM THE PROJECT

Research Abstracts

National

1. Sarkar, S. K. (2013) Fine structural study on the olfactory neuroepithelium of *Pseudapocryptes lanceolatus* (Bloch and schneider) with special reference to neural regeneration. Seminar organized by Zoological Society of Kolkata, Department of Zoology, University of Calcutta, February 12, 2013. Kolkata, West Bengal.

2. Sarkar, S. K. and De, S. K. (2013) Macroanatomy and electron microscopical study on the olfactory structure of *Pseudapocryptes lanceolatus* (Bloch and Schneider). Proceedings of International Level Seminar on 'Problems and prospects of coastal aquaculture and application of biotechnological tools for rural development'. March 1-3, 2013. Organized by Department of Aquaculture Management Technology, Vidyasagar University, Midnapore, West Bengal, pp. 35.

3. Sarkar, S.K. and De, S.K. (2013) Neuroepithelial cells in olfactory apparatus of *Pseudapocryptes lanceolatus*: A mudskipper of Bengal. 24th All India Congress of Zoology & National Seminar on 'Biodiversity and its Management for Food, Livelihood & Environmental Security' & National Helminthological Congress. November 23-25, 2013. Organized by Department of Zoology, University of Kalyani, West Bengal in collaboration with Zoological Society of India, Bodh Gaya.

4. De, S. K., Acharya, A. Sarkar, S.K., Biswas, S. and Datta, N. C. (2013) Olfactory apparatus of Fish. 24th All India Congress of Zoology & National Seminar on 'Biodiversity and its Management for Food, Livelihood & Environmental Security' & National Helminthological Congress. November 23-25, 2013. Organized by Department of Zoology, University of Kalyani, West Bengal in collaboration with Zoological Society of India, Bodh Gaya.

5. Sarkar, S.K. and De, S.K. (2013) Role of rodlet cell in olfactory neuroepithelium of a fish (*Pseudapocryptes lanceolatus*). UGC- DRS Sponsored National Seminar on 'Bioprospecting of Natural Products'. December 5-6, 2013, Organized by Department of Zoology, The University of Burdwan, West Bengal.

6. De, S.K. and Sarkar, S.K. (2014) Olfactory neuroepithelial cells use as a tool for aquatic health assessment. National conference on "Aquatic Ecosystems and their Management: Recent trends and future perspectives". March 21 – 23, 2014. Organized by Center for Environmental Sciences, Central University of Bihar, BIT Campus, Patna, Bihar.

7. De, S. K. and Sarkar, S.K. (2014) Crosstalk between olfactory sensory and nonsensory neuroepithelial cells of a mudskipper: *Pseudapocryptes lanceolatus* (Bloch and Schneider, 1801). National Seminar on Threats to Biodiversity and Ecosystems: Impacts of Developmental project and Climate Change & 25 th All India Congress of Zoology. November 17 – 19, 2014. Organized by Department of Zoology and Environmental Science, Gurukula Kangri University, Haridwar.

8. Sarkar, S.K. and De, S.K. (2014) Comparative morphoanatomy of olfactory apparatus in two different mud-dwelling teleosts. National Seminar on Threats to Biodiversity and Ecosystems: Impacts of Developmental project and Climate Change & 25 th All India Congress of Zoology. November 17 – 19, 2014. Organized by Department of Zoology and Environmental Science, Gurukula Kangri University, Haridwar.

9. De, S.K. and Sarkar, S.K. (2015) Role of macrophage in olfactory neuroepithelium of fish. National Conference on Applied Zoology in Sustainable Development: An update. January 31 – February 2, 2015. Organized by Department of Zoology, University of North Bengal (NBU), Siliguri.

10. Sarkar, S. K. and De, S.K. (2015) Anatomy and ultrastructural studies on chemosensory cells in an intertidal mudskipper (*Pseudapocryptes lanceolatus*) of South East Asia. National Conference on Applied Zoology in Sustainable Development: An update. January 31 – February 2, 2015. Organized by Department of Zoology, University of North Bengal (NBU), Siliguri.

11. Sarkar, S. K. and De, S. K. (2015) Fine structural modifications in olfactory neuroepithelium of *Pseudapocryptes lanceolatus* (Bloch and Schneider, 1801): An air breathing mudskipper of South East Asia. UGC Sponsored National Seminar on 'Zoological Research: Past, Present and Future'. March 26, 2015. Organized by Department of Zoology, Vidyasagar University, Midnapore (West) – 721102, West Bengal.

International

1. Sarkar, S. K. and De, S. K. (2013) The Olfactory Neuroepithelial components in *Pseudapocryptes lanceolatus* (Bloch and Schneider) with special reference to Neural Degeneration. International Conference on Electron Microscopy and XXXIV Annual Meeting of the Electron Microscope Society of India (EMSI). July 3-5, 2013. Organized by Saha Institute of Nuclear Physics, Kolkata and East Zone Chapter of EMSI, Kolkata, West Bengal, pp. 91.

2. Sarkar, S.K. and De, S.K. (2013) The Olfactory Neuroepithelial components in *Pseudapocryptes lanceolatus* (Bloch and Schneider) with special reference to Neural Degeneration. International Conference on Electron Microscopy, EMSI 2013. July 3-5, 2013, Kolkata, **West Bengal, India**.

3. De, S. K. and Sarkar, S. K. (2014) Vesicular Diversity and Crowding Within the Olfactory Sensory Receptor Neuron. Microscopy and Microanalysis 2014. August 3 – 7, 2014. Hardfort CT, **USA**.

13. SUMMARY OF THE FINDINGS

This research program was focused on the advanced electron microscopical demonstration of olfactory neuroepithelium of *Pseudapocryptes lanceolatus* (Bloch and Schneider), a teleostean gobiid fish of South East Asia to explore macro - and microanatomical features of the neuroepithelial components in respect to various aspects of olfaction. The unilamellar olfactory apparatus of *P. lanceolatus* is located at the ethmoid region of the head. The olfactory lamella of *P. lanceolatus* along with its nasal cavity is well associated with anterior and posterior nostrils. The nasal cavity is surrounded by pseudostratified olfactory neuroepithelium. The cellular components are categorized as sensory receptor cells, supporting cells, basal cells, etc. The sensory receptor cells are bipolar neuron in nature and conveniently divided into dendron, perikaryon and axon. The tip of the dendron protrudes towards the nasal cavity to form olfactory knob. The morphoanatomy of olfactory knob denote structural variations among the sensory receptor cells. The perikaryons of different sensory receptor cells are located at the variable depth of olfactory neuroepithelium and characterized under transmission electron microscope (TEM). The axons of different sensory receptor cell travels towards the basal lamina. The variable

morphometry of vesicles within the different subcellular compartments sensory receptor cells has been marked and concluded vesicular trafficking and crowding in bi-directional ends of the said neuron. These vesicles play important roles in neural signal transduction of olfactory signals. The ultrastructural features of acetylcholine esterase (AchE) activity in sensory receptor cells are also characterized under transmission electron microscope (TEM). Increasing frequencies of acetylcholine esterase (AchE) containing vesicles at axoplasm may denote neural dysfunction of the olfactory neuron in response to aging or heavy metal toxicity. Effect of several heavy metals (e.g., iron, copper, nickel, lead, etc.) at subcellular level is analyzed through x-ray microanalyzer under transmission electron microscope (TEM). The supporting cells are columnar in nature and categorized as ciliated supporting cell and microvillous supporting cell. This fine structural detail on the microvillous supporting cell may be an indicative for neural protection of olfactory neuroepithelium. Small polygonal basal cells show various stages of cellular differentiation within the neuroepithelium and may acts as a progenitor cell of sensory receptor cell. During differentiation, the nucleus shows specific nature of chromatin condensation. The qualitative changes in condensation of euchromatin and heterochromatin structures are marked by using florescence microscope. Beneath the basal lamina of olfactory neuroepithelium, a zone of fila olfactoria is present and characterized with the presence of several axonal bundles, Schwann cells, fibroblast cells, collagen fibers, blood capillaries, etc. It is assume that the fila olfactoria is the site for axonal accumulation which leads to the formation of olfactory nerve tracts for transduction of olfactory signals.

14. CONTRIBUTION TO THE SOCIETY

With the help of this financial assistance from UGC, New Delhi; I have educate the Post Graduate students in Zoology (first generation learner) and Ph.D. students of Jangal Mahal area to perform research work in advanced level on neuroscience of fish (*Pseudapocryptes lanceolatus*) and higher vertebrates respectively.

15. WHETHER ANY PH.D. ENROLLED/PRODUCED OUT OF THE PROJECT

Ph.D. Awarded : One

16. NO. OF PUBLICATIONS OUT OF THE PROJECT (PLEASE ATTACH RE-PRINTS)

Research Articles (Published):

- Sarkar, S. K., Acharya, A., Jana, S. and De, S. K. (2014) Macroanatomical variation of the olfactory apparatus in some Indian teleosts with special reference to their ecological habitat. *Folia Morphologica (Warsz)*, **73** (2): 122 - 128. [ISSN: 0015-5659] [Impact factor- 0.524] [H index - 14].
- De, S. K. and Sarkar, S. K. (2014) Vesicular Diversity and Crowding Within the Olfactory Sensory Receptor Neuron. *Microsc. Microanal.* 20 (Suppl 3): 1272 – 1273. [Impact Factor – 2.495]
- **3.** Sarkar, S.K. and De, S.K. (2014) Functional anatomy of cellular junctions in olfactory neuroepithelium of *Pseudapocryptes lanceolatus* (Bloch and Schneider). *Indian Journal of Biological Sciences*, **20**: 36-39.
- Sarkar, S.K., Jana, S. and De, S.K. (2014) Age-specific anatomy and cytological studies on unilamellar olfactory structure of a teleosts (*Pseudapocryptes lanceolatus*). *European Journal of Experimental Biology*, 4 (6): 105-111. [ISSN: 2248-9215]
- Sarkar, S. K., Nag, T.C. and De, S. K. (2015) Ultrastructural studies on the nuclear elements in differentiating and degenerative ciliated olfactory neuron of *Pseudapocryptes lanceolatus* (Gobiidae: Oxudercinae). *Egyptian Journal of Basic and Applied Sciences* (Elsevier), 2 (2015): 295 302. [http://dx.doi.org/10.1016/j.ejbas.2015.07.004]
- Sarkar, S. K. and De, S. K. (2015) Acetylcholine Esterase (AChE) Activity in Ciliated Olfactory Neuron of a Teleostean: Gobiid [*Pseudapocryptes lanceolatus* (Bloch and Schneider, 1801)]. *International Journal of Science and Nature*,6 (3): 444 – 446. [ISSN: 0973-3140]

Research Article (Communicated):

1. De, S. K. and Sarkar, S. K. (2015) Electron Microscope Based X-ray Microanalysis on Bioaccumulation of Heavy Metals and Neurodegenerative Dysfunctions in Mudskipper [*Pseudapocryptes lanceolatus*], *Journal of Microscopy and Ultrastructure* [Elsevier].

Annexure – V

UNIVERSITY GRANTS COMMISSION BAHADUR SHAH ZAFAR MARG NEW DELHI – 110 002

STATEMENT OF EXPENDITURE IN RESPECT OF MAJOR/MINOR RESEARCH PROJECT

1. Name of Principal Investigator	: Prof. Subrata Kumar De		
2. Deptt. of University/College	: Department of Zoology		
	Vidyasagar University		
	Midnapore (West) – 721 102		
	West Bengal		
3. UGC approval No. and Date	: F. No. 41 – 161/2012 (SR)		
	Dated 13.07.2012		
4. Title of the Research Project	: Fine structural demonstration		
	of olfactory neuroepithelium of		
	Pseudapocryptes lanceolatus (Bloch		
	and Schneider) with special		
	reference to electron enzymology and x-ray microanalysis.		
5. Effective date of starting the project	: 01.07.2012		

6. a. Period of Expenditure : From 01.07.2012 to 31.12.2015

b. Details of Expenditure

S.No.	Item	Amount	Expenditure	
		Approved	Incurred	
		Rs.	Rs.	
i.	Books & Journals	Rs. 45,000.00	Rs. 48,637.00	
ii.	Equipment	Rs. 2,62,000.00	Rs. 2,69,396.00	
iii.	Contingency	Rs. 40,500.00	Rs. 35,993.00	
iv.	Field Work/Travel	Rs. 40,500.00	Rs. 32,259.00	
۷.	Hiring Services	Rs. 40,500.00	Rs. 40,900.00	
vi.	Chemicals & Glassware	Rs. 90,000.00	Rs. 89,234.00	
vii.	Overhead	Rs. 71,800.00	Rs. 71,800.00	
viii.	Any other items (Please specify)			

- c . Staff
- Date of Appointment

13.08.2012

:

S.No.	Expenditure	From	to	Amount	Expenditure
	Incurred			Approved	Incurred(Rs.)
				(Rs.)	
01	Project Fellow	13.08.2012	30.06.2015	Rs.	Rs.
			[Extended	5,23,509.00	5,23,509.00
			upto	[Honorarium:	
			31.12.2015	Rs.4,55,225.00	
			without	+ HRA: Rs.	
			financial	68,284.00]	
			assistance]		

1. It is certified that the appointment(s) have been made in accordance with the terms and conditions laid down by the Commission.

2. It as a result of checks or audit objective, some irregularly is noticed, later date, action will be taken to refund, adjust or regularize the objected amounts.

3. Payment @ revised rates shall be made with arrears on the availability of additional funds.

4. It is certified that the grant of **Rs. 11,13,809.00** (Rupees Eleven Lac Thirteen Thousand Eight Hundred and Nine only) received from the University Grants Commission under the scheme of support for Major Research Project entitled 'Fine structural demonstration of olfactory neuroepithelium of *Pseudapocryptes lanceolatus* (Bloch and Schneider) with special reference to electron enzymology and X-ray microanalysis' vide UGC letter No. F. 41-161/2012 (SR) dated 13.07.2012 has been fully utilized for the purpose for which it was sanctioned and in accordance with the terms and conditions laid down by the University Grants Commission.

S SIGNATURE OF PRINCIPAL INVESTIGATOR ATA KUN DR. SUBK

REGISTRAR/ PRINCIPAL Registrar VIDYASAGAR UNIVERSITY Midnapore-721102