SAINT MARY'S COLLEGE

Grade 'A' NAAC Re-Accredited UGC Recognised Affiliated to NEHU



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No.SMC/MH/SCS/2013-2014/01.

Dated : 30th May, 2014.

 To: Dr. Sandhya R. Shenoy, Department of Biotechnology, Ministry of Science & Technology, Government of Meghalaya, New Delhi – 110003.

Subject : Structured information from St. Mary's College, Shillong, on DBT Star College Programme for the year 2012-13.

Madam,

On behalf of the participating departments namely, Botany, Chemistry, Physics & Zoology of St. Mary's College, Shillong, we the undersigned, take this opportunity to express our sincere appreciation and gratitude to your esteemed department for extending your valuable support to the College. Your support has immensely enriched the academic opportunities and enhanced the learning experiences of the Students and Faculty.

With reference to the subject cited above, we hereby, present the details of the information on the implementation of the DBT Star Programme Department Wise according to the questionnaire sought by you for your kind perusal. The hard copy of the same has also been mailed by registered post to your office.

Thanking you. Yours sincerely,

Mary Harriet.

Principal, St. Mary's College, Shillong

Ashabong (A.P. SHABONG)

Programme Coordinator DBT- Star College Scheme, St. Mary's College, Shillong.

Name of the college : St. Mary's College, Shillong

Type of college: Govt. aided, Undergraduate Women's College

Year of support : 2012 - 13

No. of departments benefitted by the scheme: 4 (Botany, Chemistry, Physics & Zoology)

List of courses run by different departments: Undergraduate

College ranking: Accreditted 'A' grade in 2005 and Reaccreditted 'A' grade in 2011 by NAAC, Bangalore. The third cycle of reaccreditation of the college by NAAC is due in 2016.

Permanent core faculty of participating departments:

Department of Botany

Name & Qualifications	Designation	Specialization	Experience (in years)
Ms I.B. Kharsyntiew M.Sc.	Associate Professor, HOD	Microbial Ecology	34
Dr. (Ms) A.R.Laloo M.Sc. Ph.D.	Associate Professor	Ecology	29
Dr. (Ms) M.B. Tiwari M.Sc. Ph.D	Associate Professor	Microbiology	29
Dr. (Ms) S. Rynjah M.Sc. Ph.D.	Associate Professor	Ecology	27
Ms T. Syiemlieh M.Sc.	Assistant Professor	Microbial Ecology	13

Department of Chemistry

Name & Qualifications	Designation	Specialization	Experience (in years)
Dr (Ms) B. Bhattacharjee M.	Associate	Physical Chemistry	31
Sc Ph. D	Professor, HOD	r nysicar chemistry	51
Dr (Ms) I. Nongkynrih M.Sc	Associate	Organic Chemistry	20
Ph. D	Professor	Organic Chemistry	23
Ms. I. Dey M.Sc	Associate	Physical Chemistry	21
	Professor	r nysicai Chennistry	21

Dr (Ms) P. Dey M.Sc Ph. D	Associate Professor	Organic Chemistry	16
Mr. M. Sawkmie M.Sc. NET	Assistant Professor	Organic Chemistry	7

Department of Physics

Name & Qualifications	Designation	Specialization	Experience (in years)
Dr. (Mrs) E.M.L. Buam, M.Sc. Ph.D	Associate Professor, HOD	Experimental & Theoretical Nuclear Physics	29
Mrs. R.Das, M.Sc	Associate Professor	Theoretical Nuclear Physics	29
Dr.(Mrs) B.Dey, M.Sc. Ph.D.	Associate Professor	Theoretical Nuclear Physics	17
Mr. C.Synrem, M.Sc. NET	Assistant Professor	Theoretical Nuclear Physics	7
Ms. D.War, M.Sc.NET	Assistant Professor	Theoretical Nuclear Physics	3

Department of Zoology

Name & Qualifications	Designation	Specialization	Experience (in years)
Ms. A. P. Shabong M.Sc	Associate Professor, HOD	Cytogenetics	31
Dr (Ms) N.Mishra M.Sc, Ph.D	Associate Professor	Biochemistry & Parasitology	29
Ms M.R. Jyrwa M.Sc M.Phil.	Associate Professor	Limnology	19
Ms Y. Nongrum M.Sc NET	Assistant Professor	Cell Biology & Immunology	4
Mr. L. Marbaniang M.Sc NET	Assistant Professor	Cell Biology & Immunology	4

Extramural support: DST FIST

Impact of the Program: Significant improvements in the field of imparting quality education and training to the students have been achieved upon receiving DBT support. Some improvements may be listed as follows:

- The grant has enabled the participating departments to procure new equipments which have effectively improved the 'hands on' experimental work of the students thereby promoting critical thinking in them.
- The books and journals purchased under this scheme have enriched the departmental libraries which provided quick and easy access of students to reading materials.
- The improvement of IT facilities in participating departments have motivated students and teachers to optimize their learning capabilities.
- The grant have enabled the departments to conduct exposure programmes of students to various Research Laboratories. The visits have greatly helped the faculty and students in establishing linkage with the scientists and relevant laboratories.
- Guest Lectures/Workshops/ Field trips etc conducted by departments proved extremely effective in promoting innovative thinking in students as well as faculty.

Problems faced in implementation of the programme:

- Submission of details required by DBT coincides during time period when faculties are deeply committed in examination procedures.
- The annual academic session followed by the college as per the recommendations of the university to which the college is affiliated extends from the second week of February to the second week of December. Winter vacations are utilized by students for preparation of examinations which begins in February and ends by the first week of May every year.
- Vast curriculum, tight class schedule, level of students admitted, lack of summer break within academic session and time constraints pose difficulty for the college to organize/conduct frequent exposure programmes for students and faculty.

Suggestions for improvement:

- In view of rising cost of education, that DBT may offer stipend/scholarship for economically challenged students to enable them to pursue higher education in life science and biotechnology.
- Remuneration to coordinators as incentives to fulfill the aims and objectives of DBT star college programme in colleges.
- That DBT works out a revised format in relation to information on implementation of the programme from colleges which may be submitted only once a year so that faculty can devote valuable time in the completion of syllabus as implemented by the university to which the college is affiliated.

STAR COLLEGE SCHEME – QUESTIONNAIRE Department of Botany

Faculty:

Names of Faculty of the Department:

1.	Mrs. Iuvenis B. Kharsyntiew	Associate Professor and Head
2.	Dr. (Mrs) Allana R. Laloo	Associate Professor
3.	Dr. (Mrs) Manjumani B. Tiwari	Associate Professor
4.	Dr. (Miss) Sabina Rynjah	Associate Professor
5.	Mrs. Tiewlyngksiar Syiemlieh	Assistant Professor

Faculty activities:

- a. "Faculty training and motivation and Adoption of Schools and Colleges by CSIR Labs" organized by CSIR NEIST (North East Institute of Science & Technology), Jorhat, Assam at St. Edmund's College, Shillong on 21st and 22nd March, 2013.Attended by : Ms. I. B. Kharsyntiew, Dr(Ms) A. R. Laloo, Dr(Ms) M. B. Tiwari, Dr(Ms) S. Rynjah
- b. National Seminar on "Burning Environmental Issues : Risk to Biodiversity and Human Health, with special reference to North East India" organized by St. Mary's College, Shillong in collaboration with State Council of Science, Technology and Environment, Shillong on 15th and 16th May, 2013.
 Participated by: Ms. I.B. Kharsyntiew, Dr. (Ms) A.R. Laloo, Dr. (Ms) M.B. Tiwari, Dr. (Ms) S. Rynjah
- c. Youth Development Seminar held in St. Mary's College, on 6th June, 2013.
 Attended by: Dr. (Ms) M. B. Tiwari, Dr. (Ms) S. Rynjah
- d. "Workshop on Instrumental Techniques and their applications" conducted by Chemistry Department, St. Mary's College, on 15th July 2013.
 Attended by: Dr. (Ms) M. B. Tiwari, Dr. (Ms) S. Rynjah, Ms. T. Syiemlieh
- e. 6th ICCB (International Congress for Conservation Biology) held at Baltimore, Maryland, USA from 21st to 25th July, 2013. Poster Presentation by Dr. (Mrs) M.B. Tiwari on "Technological Innovations in Shifting Agricultural Practices by three tribal farming communities of Meghalaya".
- f. Workshop on "Revision of School Curriculum In the light of CCE (Elementary level) organized by the Director, Educational Research and Training, Govt. of Meghalaya. From 16th to 21st September, 2013 Participated by: Dr. (Ms) S. Rynjah

Extra-Mural Projects from Different Funding Agencies - nil

Recognition to Faculty – nil

	No. of Applicants vs.	Students	M/F Cut-off	Details of Students admitted					Dropout				
Year	Sanctioned seats	admitted (B. Sc. 'I' only)		NI/F	IVI/F	IVI/F	IVI/F	WI/F %	G	ST	SC	OBC	РН
2011	38 vs. 30	32	F	45	4	26	-	2	-	18.75			
2012	36 vs. 30	28	F	45	-	27	-	1	-	32.1			
2013	45 VS. 30	28	F	50	4	24	-	-	-	10.7			

6. STUDENT STATISTICS

Year	Class	Appeared: Passed	Pass %	Position secured in University Exam	PG Admissions to University
	B. Sc. I	14:11	78.5	_	_
2011	B. Sc. II	7:6	85.7	-	-
	B. Sc. III	4:4	100	2 nd	-
	B. Sc. I	26:20	76.9	-	-
2012	B. Sc. II	9:9	100	-	-
	B. Sc. III	6:6	100	7 th	-
	B. Sc. I	18:13	72.2	-	-
2013	B. Sc. II	19:16	84.2	-	-
	B. Sc. III	9:9	100	-	-

N. B.:

1. Results of 2011, 2012, 2013 are of students admitted in the years 2010, 2011, 2012.

2. After B. Sc. Students get admission in other universities for professional courses.

7. TRAININGS/ EXPOSURE VISITS/ OUTREACH

Student Activities carried out during the year (2012 – 2013)

i. Projects:

- a) B. Sc. 1st Year (Hons. Students): Collection of specimens and information about ethnobotanically important plants used by the tribals of Assam, Manipur and Meghalaya.
- b) B. Sc 2nd Year (Hons students): Visits to several farms and gardens in and around Shillong to collect specimens of diseased plants to have an idea of the common Diseases affecting crops in this area.
- c) B. Sc 2nd Year (Pass students): Study of the plant diversity in and around Shillong by collection of plants and preparation of herbarium sheets.
- d) B. Sc 3rd Year (Hons. Students): Study of medicinal plants found in East, West and South Khasi Hills Districts and Ri Bhoi Districts of Meghalaya.

ii. Field trip:

A Field trip was organized by the Department on 1st October 2013 to Mawphlang Sacred Grove. 25(Twenty five) B.Sc I (Hons) students and 54(Fiftyfour) B.Sc II(Pass) students were taken on this trip.

iii. Summer training/Winter School/Autumn School:

- a) Trolian Mawlong(B.Sc. II Hons) attended the Winter School organized by the Science Departments of Lady Keane College, College from 29th November to 13th December, 2012.
- b) Hriiziini Monica and Rimeia C. Lyngdoh(B.Sc. III Hons) attended the "Autumn School in Plant Sciences" organized by the Department of Botany, North Eastern Hill University, Shillong from 18th to 31st October, 2013.

iv. Other activities:

- a) B.Sc. 1st Year (Hons) students attended the "Slow Food Festival" at St. Edmund's College, Shillong on 13th October 2012. Here they witnessed the different types of Plants used by the local tribals for various purposes.
- b) Bharina Kharumnuid, Baniada Thabah, Clarisha Nongbri, Meiairihulang Fancon, Metisha Langstieh, Superlity Ryntathiang and Wandaphisha Dkhar of B.Sc. II (Hons) took part in the Poster Campaign on "Eat, Think and Save" organized by the Nature Club of the College to observe "World Environment Day" on 5th June 2013.
- c) Rimeia C. Lyngdoh (B.Sc III Hons) took part in the Debate on "Cloning a human design to save endangered species" at Shillong College, Shillong on 27th July 2013.

 d) Hriiziini Monica and Mary Diana Tariang (B.Sc. III Hons) presented papers on 'PHYTOREMEDIATION' and 'ALLELOPATHY' respectively in the Inter – College Seminar held at St, Anthony's College, Shillong on the 8th August, 2013.

v. Guest Faculty:

A Popular Talk on "PLANT – **MICROBES INTERACTION**" was organized by the Department for the B.Sc I(Hons), B.Sc II(Hons) and B.Sc III(Both Pass and Hons) students on the 24th October 2013.

The Guest Speaker was **Dr. P. L. Nongbri.** He did his M. Sc. From JNU, New Delhi, got his Ph.D degree from Friedrich Schiller University, Jena, Germany. At present, he is working on a Project with the M.Sc. Biotechnology Department of St. Anthony's College, Shillong.

vi. List of New Techniques, Practicals and Demonstrations:

Since the college is affiliated to the North Eastern Hill University, new practicals cannot be introduced outside the syllabus given by NEHU. But the students of the Department are taught different techniques such as preparation of herbarium

Sheets, permanent slides etc. They are also shown how to use different instruments such as Colorimeter, Centrifuge, Autoclave, Distillation Apparatus, Laminar Flow, BOD Incubator etc.

8. Facilities Created Under the Scheme

I. Procurement of New Equipment, Books, etc.

STATEMENT OF EXPENDITURE:

A. Non – recurring (16th July 2012 till 31st March 2013)

i. Laboratory Equipments:			Rs. 82894.00		
ii. IT Equipment	ii. IT EquipmentS:				
	TOTAL:	Rs.	252025.00		
Non – recurring(1 st A	April 2013 till date)				
i. Laboratory Equi	Rs. 212469.00				
ii. Camera + UV f	Rs. 40141.00				
	Total:	Rs. 2	252609.00		
	GRAND TOTAL:	Rs.	504634.00		
	Amount Received:	Rs. 5	5,00,000/-		
	Total Amount utilized:	Rs. 5	5,04,634/-		
	Remaining Balance: (–)	Rs. 4	1,634/-		

B. Recu	B. Recurring (16 th July 2012 till 31 st March 2013) - NIL								
Recurring (1 st April 2013 till date)									
i.	Laboratory Glasswares:	Rs. 79265.11							
ii.	Chemicals:	Rs. 29348.00							
iii.	Chromatography paper, Filter papers, etc:	Rs. 20767.00							
iv.	Mounting specimens:	Rs. 13733.00							
v.	Books:	Rs. 38543.00							
vi.	Journals:	Rs. 5050.00							
vii.	Field trip:	Rs. 7500.00							
viii.	Contribution to Coordinator's trip to Mumbai for the Coordinators Meeting:	Rs. 5000.00							
ix.	Popular talk:	Rs. 2000.00							
	GRAND TOTAL:	Rs. 201206.00							
	Amount Received:	Rs. 2,00,000/-							
	Total Amount Utilized:	Rs. 2,01,206/-							
	Remaining Balance: (–)	Rs. 1,206/-							

DBT GRANT UTILISATION: DETAILED STATEMENT OF EXPENDITURE.

A. NON – RECURRING (16th JULY 2012 till 31st March 2013)

Sl. No.	Equipments	Model	Quan tity	Cost	TOTAL Cost & VAT	Date of Order	Date of purchase/install
1.	Vertical autoclave Dimension 250 x 450 mm	NSW	1	73034.00	82894.00	21.12.2012	8.3.2013
2.	Desktop with monitor	HP P2 1403	1	30005.71			
3.	Laptop with carry case	HP – G62204TX	1	35328.10	95356.00	7.2.2013	20.3.2013
4.	Laser Jet Printer	HP-M1536 DNF	1	23380.95			
5.	UPS – Intex	600VA	1	2190.48			
6.	LCD Projector(Epson)	EB – X02	1	37000.00	73775.00	21.12.2012	21.3.2013
7.	LCD Camera(Dewinter)	EB – X02	1	28000.00			

TOTAL: 252025.00

Sl. No.	Equipments	Model	Qty.	Cost	TOTAL Cost & VAT	Date of Order	Date of Purchase/ Install
1.	Student monocular Microscopes(Olympus)	HSA	10	85000.00			
2.	40x Objective	HAS/HB	10	16800.00	151386.00	21.12.2012	19.4.2013
3.	Dissecting microscopes with special lens & 75mm diameter	DM-4	20	31500.00			
4.	Double distillation unit with auto cut-off design	JSGW	1	25317.00	28735.00	21.12.2012	17.6.2013
5.	Canon Camera(DSLR)	EOS1100D	1	38990.00	40140.00	21 12 2012	17.6.2013
6.	Canon UV – filter	EOS1100D	1	1150.00	10110.000	21.12.2012	17.0.2015
7.	Photoelectric Colorimeter	AIMIL	1	9500.00			
8.	Compact Laboratory Centrifuge with rotor head.	REMI	1	19000.00	32348.00	26.10.2013	28.10.2013

NON – RECURRING (1st APRIL 2013 till date)

NON RECURRING:

AMOUNT RECEIVED	=	Rs. 5,00,000/-
AMOUNT UTILIZED	=	Rs. 2,52,025/- (16/07/2012 to 31/03/2013)
	=	Rs. 2,52,609/- (01/04/2013 till date)
TOTAL	=	Rs. 5,04,634/-
BALANCE	= ()	Rs. 4,634/-

B. RECURRING (1st APRIL 2013 till date)

i. LABORATORY GLASSWARES

Sl.No.	Items	Brand	Capacity	Qnty	Cost	TOTAL cost with VAT	Date of Order	Date of Purchase
1.	Beaker	Riviera	250ml	12	636.00	17463.11	21.12.2012	17.6.2013
	Beaker	Riviera	500ml	16	1504.00			
	Beaker	Riviera	1000ml	6	1140.00			
2.	Measuring cylinders	Riviera	10ml	6	1224.00			
	Measuring cylinders	Riviera	50ml	10	2670.00			
	Measuring cylinders	Riviera	100ml	4	1224.00			
3.	Conical flasks	Riviera	250ml	6	474.00			

Sl.No.	Items	Brand	Capacity	Qnty	Cost	TOTAL cost with VAT	Date of Order	Date of Purchase
	Conical flasks	Riviera	500ml	6	594.00			
4.	Pipettes	JSGW	1ml	12	1032.00			
	Pipettes	JSGW	2ml	12	1164.00			
	Pipettes	JSGW	5ml	12	1296.00			
	Pipettes	JSGW	10ml	12	1428.00			
5.	Test tubes	JSGW	10ml	100	1000.00			
6.	Haemocytometer	Neum baur	-	5	14000.00	15890.00	21.12.2012	17.6.2013
7.	Micropipette	Borosil	100- 1000 Ml	2	6924.00	15201.00	21 12 2012	10 7 2012
8.	Electronic pipette Filling device	Borosil	-	1	6636.00	15591.00	21.12.2012	10.7.2013
9.	BOD bottles	JSGW	125ml	30	9150.00	25147.00	21.12.2012	17.9.2013
10.	Burette	JSGW	25ml	10	1650.00			
11.	Burette	JSGW	50ml	10	2250.00			
12.	Conical flask (narrow mouth)	Riviera	100ml	10	550.00			
13.	Funnels, long stem	Riviera	100mm	4	336.00			
14.	Apparatus for investigating transpiration	JSGW	-	12	8220.00			
15.	Conical flask	Riviera	100ml	14	770.00	1446.00	4 10 2012	0 10 2012
16.	Funnels	JSGW	100mm	6	504.00	1440.00	4.10.2015	9.10.2015
17.	Slides	Abdes	-	30pkts	1470.00			
18.	Cover slips	Blue Star	-	30pkts	2012.00	3928.00	9.10.2013	25.10.2013

TOTAL: 79265.11

ii. CHEMICALS

Sl. No.	Chemical name	Company	Packing	Qnty	Cost	Total Cost with VAT	Date of Order	Date of Purchase
1.	DPX Mountant	Himedia	500ml	2	972.00	14788.00	21.12.2012	17.6.2013
2.	Aniline Blue (Cotton blue)	دد	25gm	4	664.00			
3.	Coomasie Brilliant Blue	۵۵	25gm	1	4619.00			
4.	Methyl violet	دد	25gm	2	300.00			
5.	Carmine powder	دد	25gm	1	2535.00			

Sl. No.	Chemical name	Company	Packing	Qnty	Cost	Total Cost with VAT	Date of Order	Date of Purchase
6.	Diphenylamine	"	250gm	1	590.00			
7.	Petroleum ether	دد	500ml	1	2300.00			
8.	Potassium iodide	دد	100gm	1	1900.00			
9.	Iodine	دد	50gm	1	204.00			
10.	Safranin powder	"	100gm	1	1270.00	11499.00	21.12.2012	1.7.2013
11.	Copper sulphate (Pentahydrate)	۵۵	500gm	1	430.00			
12.	Manganese sulphate	۵۵	500gm	1	600.00			
13.	Agar agar powder	۵۵	100gm	1	1915.00			
14.	Haematoxylin	"	25gm	1	4560.00			
15.	Orcinol	.د	10gm	1	1076.00			
16.	Ninhydrin	"	10gm	2	1100.00	-		
17.	Sulphuric acid	"	500ml	2	550.00	3061.00	9.10.2013	25.10.2013
18.	Petroleum ether	۲۲	500ml	1	2065.00			

TOTAL : 29348.00

iii. Chromatography paper, Filter paper, indicator paper

Sl. No.	Item	Brand	Packing	Qnty	Cost	TOTAL cost with VAT	Date of Order	Date of Purchase
1.	Chromatography	Whatman	100	3 pkts	13080.00	14846.00	21.12.2012	16.7.2013
	Paper		Sheets					
2.	Filter paper	Whatman	100	5 pkts	4250.00	5921.00	9.10.2013	25.10.2013
			Sheets	_				
3.	Filter paper	Ordinary	100	5 pkts	225.00			
		-	Sheets	_				
4.	Indicator paper	Rankem	-	5 pkts	1000.00			

TOTAL : 20767.00

iv. Mounting specimens.

TOTAL:		Rs. 13733.00
Total cost with VAT	=	Rs. 13733.00
550 bottles (different species) Cost	=	Rs. 12100

v. Books

Sl. No.	ISBN	Title	Author	Publisher	Qty.	Amount	Dealer
1.	9788177540949	Plant Physiology: Fundamentals & Applications	Kumar, A	AGRO	1	1800.00	Eastern Book House
2.	9788177544589	Laboratory Manual of plant physiology, biochemistry & ecology	Akhtar, I	AGRO	1	595.00	۲۵
3.	9788177540567	Plant Breeding : Theory & Techniques	Gupta, S.K.	AGRO	1	1200.00	۲۵
4.	9789350300398	Genetics & Plant Breeding	Agarwal, V.	Oxford – ABD	1	1625.00	۲۵
5.	9788177541722	AGRO's Colour Atlas of Medicinal Plants.	Purohit, S.S.	AGRO	1	1595.00	۲۵
6.	9780070486676	Principles of Genetics	Tamarin	TMH	1	740.00	"
7.	9788180303043	Modern College Botany(3 vols)	Mohanka'R.	Campus Book	1	5500.00	۲۵
8.	9788180301544	Taxonomy of Angiosperms(2 Vols)	Vardhana, R	Campus Book	1	3500.00	۲۵
9.	9788180302183	Text Book of Botany(4 vols)	Singh, M.P.	Campus Book	1	8000.00	۲۵
10.	9788177541175	Cell Biology: Fundamentals & Applications	Gupta Jangir	AGRO	1	1800.00	۰۵
11.	81772332556	Perspectives in Biotechnology	Reddy, S.M.	Scientific	1	1200.00	۰۵
12.	9788170194705	Latest Approaches in Pollen Biology & reproduction	Raju, A.J.S.	Daya – Today	1	795.00	۰۵
13.	9788131102084	Biogeography	Mehtani, S.	Common- Wealth	1	1950.00	۲۵
14.	9788177543957	Biochemistry: Fundamentals & Applications	Purohit,S.S.	AGRO	1	1500.00	۰۵
15.	9788189233648	Sacred Forest: Their ecology & Diversity	Pandey, H.N.	Daya	1	700.00	۰۵

Sl. No.	ISBN	Title	Author	Publisher	Qty.	Amount	Dealer
16.	9789380199986	Text Book of Plant	Saxena, P.	Wisdom	1	1195.00	Eastern
		Anatomy		(Domi)			Book
							House
17.	9788177543537	Modern Plant Pathology	Dube, H.C.	AGRO	1	1200.00	"
18.	9788189473846	Embryology	Kaushik, R.	Oxford –	1	1300.00	"
				ABD			
19.	9788189473457	Biotechnology:	Ganguli, A.	Oxford –	1	950.00	"
		Fundamentals &		ABD			
		Applications					
20.	9789381084700	Advanced Plant	Dube, S.M.	Swastik	1	850.00	"
		Geography					
21.	9788182930247	Principles of Palaeobotany	Bora, L.	Mittal	1	950.00	"
22.	9788179103630	Plant Tissue Culture &	Kumar, R.	AAVIS	1	2200.00	دد
		Applied Plant					
		Biotechnology					
23.	9788179021541	Medicinal plants of North	Islam, M.	AAVIS	1	600.00	۲۲
		East India					
24.	9788171326235	Ethnic Tribes &	Trivedi,	Pointer	1	1600.00	۲۲
		Medicinal plants	P.C.				
25.	9788179101568	Biotechnology & Biology	Trivedi,	AAVIS	1	2000.00	دد
		of plants	P.C.				
						452 45 00	

TOTAL:

45345.00

Discount % 15.00:

NET TOTAL:

6801.75 38543.00

 Date of Order
 : 16.02.2012

 Date of Purchase
 : 24.07.2013

vi. JOURNALS

Sl. No.	Name of the journal	Distributor	Subscription Charges	Demand Draft No.	Bank	TOTAL	Date of Payment
1.	Journal of Indian Botanical Society	Indian Botanical Society Bareilly	Rs. 1000/-	750003	SBI	Rs. 1000/-	7.10.2013
2.	Journal of Bio- Sciences	Indian Academy of Science, Bangalore	Rs. 400/-	750004	SBI	Rs. 400/-	7.10.2013

Sl. No.	Name of the journal	Distributor	Subscription Charges	Demand Draft No.	Bank	TOTAL	Date of Payment
3.	Indian Journal of Traditional Knowledge	Director, NISCAIR, New Delhi	Rs. 1200/-	750005	SBI	Rs. 1200/-	7.10.2013
4.	Bulletin of the Botanical Survey of India	Director, BSI, Kolkata	Rs. 750/-	750006	SBI	Rs. 750/-	7.10.2013
5.	Indian Phyto- Pathology	Indian Phytopatho- Logical Soc. IARI, New Delhi	Rs. 500/-	750007	SBI	Rs. 500/-	7.10.2013
6.	Indian Journal of Biotechnology	Director, NISCAIR, New Delhi	Rs. 1200/-	750055	SBI	Rs. 1200/-	7.10.2013

TOTAL: Rs. 5050/-

vii. FIELD TRIP.

Place of Visit: Mawphlang Sacred Grove & Horticulture Farm, Govt. of Meghalaya, Upper Shillong.

Date of Visit: 1.10.2013

Purpose of Visit: To study the nature of the native flora of Khasi Hills which has been preserved in the Sacred Grove at Mawphlang. To study the different methods adopted in plant Propagation carried out in the Horticulture Farm.

	TOTAL	=	Rs. 7500/-
	Guide Fee (Sacred Grove)	=	Rs. 250/-
	Entry Fee (Sacred Grove)	=	Rs. 450/-
Expenditure:	Conveyance (2 buses)	=	Rs. 6800/-

viii. POPULAR TALK.

Topic: PLANTS – MICROBES INTERACTION Resource Person: Dr. P. L. NONGBRI Date: 24. 10. 2013 Expenditure: Honorarium Rs. 2000/-TOTAL = Rs. 2000/-

ix. CONTRIBUTION FOR COORDINATOR'S TRIP TO MUMBAI FOR THE SECOND COORDINATORS MEETING ON 5th to 7th AUGUST, 2013.

Conveyance from Shillong to Guwa	hati and	back @ Rs. 2500)/- x 2=	Rs. 5000/-
		TOTAL	=	Rs. 5000/-
RECURRING:				
AMOUNT RECEIVED	=	Rs. 2,00,000/-		
AMOUNT UTILIZED	=	Rs. 0/-	(16/7/12 t	o 31/3/13)
	=	Rs. 2,01,206/-	(1/4/13 till	date0)
BALANCE	= ()	Rs. 1,206/-		

S. O. P. DEPARTMENT OF BOTANY

I. PLANT DIVERSITY.

1. ALGAE: Requirements: Slides, cover slips, microscopes, needles, forceps, tissue paper, algal specimens, safranin.

Procedure: Few specimens are taken on a clean slide. 2 - 3 drops of safranin are placed over the specimen. Wait for 5 minutes. Wash off the stain with water. 2 - 3 drops of glycerine are then put on the specimen. The specimen is spread on the slide using needles and forceps. The specimen is then covered with a cover slip. The slide is then observed under a microscope.

2. **FUNGI**: Requirements: Fungal specimens, cotton blue, lactophenol, new blades, slides, cover slips, needles, brush etc.

Procedure: Cut very thin sections of the specimen and place on a petridish containing water. Then select the thinnest section and put it on a clean slide. Put one drop of cotton blue stain and one drop lactophenol and then put a coverslip over the specimen. Observe under the microscope.

3. **BRYOPHYTES**: Requirements: Bryophyte specimens, safranin, slides, cover slips, microscopes etc.

Procedure: First the external features of the specimens are studied and important characters noted. Then cut sections through the thallus, archegoniophore, stem, leaf of the specimen. Stain with safranin. Wash off excess stain with water. Place on a slide with a cover slip over the specimen. Observe under the microscope. Take the sporophytes and place on a slide. With the help of another slide, press the sporophyte. Stain with safranin and then observe under the microscope.

4. **PTERIDOPHYTES**: Requirements: Pteridophyte specimens, slides, cover slips, microscopes etc.

Procedure: First, external features of the pteridophyte specimens are studied and important characters are noted. Then the anatomy of the rhizomes, aerial shoots, leaf, spore producing organs and gametophytes are studied. For the anatomical studies, sections of the different parts are cut, then stained with a safranin and fast green combination, mounted in glycerine and then observed under the microscope.

II. PLANT ANATOMY:

Requirements: Stems, safranin and fast green stains, forceps, needles, blades, tissue paper, alcohol grades (30%, 50%, 70%, 90% and 100%)

Procedure:

- 1. Cut a thin transverse section from the supplied stem.
- 2. Stain it in safranin for 15 minutes.
- 3. Transfer it into 30% alcohol. Keep for 10 minutes.
- 4. Then transfer the section to 50% alcohol. Keep for 5 minutes.
- 5. Transfer the section to 70% alcohol. Keep for 5 minutes.
- 6. Transfer to 90% alcohol. Keep for 5 minutes.
- 7. Transfer to 100% alcohol. Keep for 5 minutes.
- 8. The section is then kept in fast green stain for 5 minutes.
- 9. Wash it again in 100% alcohol.
- 10. Transfer to a petridish containing xylol.
- 11. Take a clean slide and pour few drops of DPX.
- 12. Take out the section from the xylol and mount it on the slide containing DPX.
- 13. Cover with cover slip and observe under the microscope.
- 14. Leave the slide to dry.

III.PLANT PHYSIOLOGY.

1. STUDY OF TRANSPIRATION RATES IN DORSIVENTRAL AND ISOBILATERAL LEAVES BY BLACKMANN'S APPARATUS.

Theory/Principle: Loss of water in the form of vapour from the aerial parts of the plant is known as transpiration. Most of the transpiration takes place through the stomatal openings of the leaves. The rate of transpiration therefore, besides other factors also depends upon the size, position and distribution of stomata on the leaves.

Requirements: Blackmann's apparatus, two potted plants(with dorsiventral and isobilateral leaves respectively), calcium chloride, olive oil, Vaseline, weighing machine.

Procedure:

- (i) The two bell jars of the Blackmann's Apparatus are fixed on the two sides of (a) a dorsiventral leaf (b) an isobilateral leaf by means of a clamp stand.
- (ii) The contact between the leaf(s) and the bell jars is made airtight by vaseline.
- (iii) In two small tubes calcium chloride is taken. The weight of each test tube is taken (W_1) .
- (iv) One tube each is then kept in the upper and lower bell jars.
- (v) The U tubes fitted at the ends of the two bell jars are filled with olive oil to disconnect the inner air from the outer atmosphere.
- (vi) The whole set-up is then left in the sun for about two hours.

(vii)

(viii) After this, the two small calcium chloride tubes are taken out and weighed again.

RESULTS:

Dorsiventral leaf: Initial weight of small tube (W1) = ------

Final weight of small tube (W2) = -----

Isobilateral leaf: Initial weight of small tube (W1)	=
Final weight of small tube (W2)	=
W2 - W1	=

2. EXTRACTION AND SEPARATION OF PLANT PIGMENTS BY PAPER CHROMATOGRAPHY.

REQUIREMENTS: Tall glass jars, fresh leaves (Rumex), mortar and pestle, Acetone 80%, Capillary tubes, measuring cylinders, hair dryer, beakers, Chromatography Paper (Whatman No. 1), Scissors, Scales, pencil, threads, Vaseline, cellotapes etc.

Procedure:

- (i) A tall glass jar is taken. The jar and the lid are wiped clean with petroleum ether and acetone.
- (ii) A solvent mixture is prepared taking petroleum ether and acetone in the ratio of 100:12.
- (iii) The solvent is then poured into the jar which is then covered with the lid so that it becomes saturated.

- (iv) Chromatogaphy paper is cut into a square sheet in a size which could fit easily into the jar.
- (v) A fine line is drawn with a pencil about 2.6 cm parallel to the bottom edge of the sheet.
- (vi) A small circle is drawn at the centre of the line.
- (vii) Fresh green Rumex leaves are taken, cut into small pieces and crushed in a mortar by adding a little amount of acetone at intervals.
- (viii) The pulp is squeezed and the extract was collected. The extract is filtered and the filtrate collected.(pigment extract).
- (ix) With the help of a capillary tube, the pigment extract is loaded on the small circle on the chromatography paper. The pigment is dried with a hair dryer.
- (x) The process of loading the pigment extract is repeated for 4-5 times.
- (xi) The sheet is then placed vertically inside the jar by hanging it on a thread which is already tied around the lid. The spot edge is allowed to just dip inside the solvent.
- (xii) The jar is re-covered and then made airtight by applying Vaseline all around the edges.

It was then left for 1 - 2 hours for development.

- (xi) When the solvent has risen to about ³/₄ of the chromatography paper, the paper is removed from the jar and dried with a hair dryer.
- (xii) The solvent front is noted and the pigments identified and marked with a pencil.

OBSERVATION: Different pigments were seen separated at different levels on the chromatography paper.

CALCULATIONS: Pigments were identified by calculating the Rf value of each.

 $Rf = A_1/A_2$ where A_1 = distance travelled by pigment,

 A_2 = distance travelled by the solvent.

3. DETERMINATION OF WATER POTENTIAL OF CELL SAP BY PLASMOLYTIC METHOD.

Principle: The potential with which pure water will diffuse into a solution is called osmotic potential (ψ_S) of that solution. Addition of solutes to a solution increases its osmotic potential. Therefore, higher the concentration of the solution, greater is its osmotic potential.

Requirements: Molar sucrose solution, red onion bulbs, forceps, slides and cover slips, graph paper, tissue paper, microscopes etc.

Procedure:

- (i) The upper epidermis of dark red onion fleshy leaf is peeled off and then cut into small pieces.
- (ii) A few drops of sucrose solution of different concentrations (0.1 to 1.0M) are poured into 10 different slides.
- (iii) In each slide, a piece of onion peel is placed on the sucrose solution and then covered with a cover slip.
- (iv) In another slide, a few drops of distilled water is placed and another piece of onion peel is added to it and covered with a cover slip.
- (v) The slides are then left for about 30 minutes.
- (vi) Each peel is then observed under the microscope to find out the progress of plasmolysis.
- (vii) The total number of plasmolysed cells and unplasmolysed cells are counted separately for each slide and the percentage of plasmolysis is calculated.
- (viii) The concentration of the sucrose solution in which 50% of the cells show plasmolysis is found out from the plotted graph.

The osmotic potential (ψ_S) of any cell sap can be found by calculating the osmotic pressure (ψ_P) .

The value of the former is negative (-) while that of the latter is positive (+).

Tabulation:

SI. No.	Conc. Of Sucrose soln)	No. of plasmolysed cells(x)	No. of unplasmolysed cells(y)	Total No. of cells in a microscopic field. (x + y = z)	% of plasmolysis
1.	0.1M				
2.	0.2M				
3.	0.3M		-		
			-		

Total number of cells in a microscopic field = z% of plasmolysis = <u>No. of plasmolysed cells</u> X 100 Z

Osmotic potential (ψ_s) = $-M \times 0.082 \times (273^{\circ} + \text{Room temp})$ atms.

4. DETERMINATION OF PHOTOSYNTHETIC ACTIVITY BY MODIFIED WINKLER'S METHOD.

Requirements: Submerged hydrophyte(Hydrilla), BOD bottles, Pipettes, Burettes, Electronic balance, Sodium thiusulphate(0.025N), Alkaline iodide, Manganous sulphate, Starch indicators, Conical flasks.

Procedure:

- (i) A known weight of photosynthetic tissue (hydrophytes) in two tightly is kept in two closed containers containing water. One container is kept in the dark and the other is kept in the dark.
- (ii) The water samples from the containers are collected in two 250ml BOD bottles. Special care is taken to avoid air bubbling into the water. The bottles are filled completely and stoppered.
- (iii) To each BOD bottle (light and dark) add 2 ml of MnSO₄ at the surface of the liqid. Replace the stopper immediately.
- (iv) Next 2ml of alkaline iodide is added to each bottle at the surface. Replace the stopper immediately taking care to avoid trapping air bubbles. Both the bottles are the inverted several times.
- (v) The bottles are then left undisturbed for 5 minutes to allow complete reaction to take place. Brown precipitate is formed.
- (vi) Add 2ml of conc. H_2SO_4 by allowing the acid to run down the neck of the bottles above the surface of the liquid.
- (vii) Re-stopper the bottles, rinse the tops of the bottles to remove any acid and shake well until all the precipitate has dissolved.
- (viii) 50ml of water sample is taken from each bottle in two 100ml conical flasks. 2ml of starch indicator is added to each bottle. The water sample turns blue in colour.
- (ix) The water sample is then titrated against 0.025N sodium thiosulphate, till the blue colour disappears. The amount of sodium thiosulphate used is noted.
- (x) One BOD bottle is filled with distilled water. Steps (iii) to (ix) are repeated to this bottle. This is the BLANK.

OBSERVATIONS:

BLANK		DARK			LIGHT			
R1	R2	Mean	R1	R2	Mean	R1	R2	Mean

CALCULATIONS:

Net photosynthesis = Net Oxygen evolved Net O_2 evolved = Amount of O_2 evolved + amount of O_2 consumed by respiration Amount of O_2 present = Initial amount of O_2 + O_2 evolved by photosynthesis(O_2 lost due to to respiration)

To calculate the amount of dissolved oxygen in the, the following formula is used :

Dissolved Oxygen(DO) = ? x 1000 x N x Volume of titrant/ Volume of sample titrated

? is a constant since 1ml of 0.025N of $Na_2S_2O_3$ is equivalent to 0.2mg of Oxygen.

N = strength of the titrant(sodium thiosulphate) in normality.

Now

Initial amount of O_2 'X' = ? x 1000 x 0.025N x Vol. of titrant used/ Vol of sample taken

"
در
= Y - X
$= \mathbf{X} - \mathbf{Z}$
= (Y - X) + (X - Z)

IV. CELL BIOLOGY AND GENETICS.

1. PREPARATION OF SLIDES OF ROOT TIPS AND STUDY OF MITOSIS.

Procedure: The onion bulbs are placed on conical flasks full of water, with the root base touching the water. These are kept for one week to allow growth of new roots. The root tips are then cut (5mm from the tip) and placed in a vial containing a mixture of 1 : 3, acetic acid : methanol for one hour. This process is called Fixation.

The root tips are removed and hydrolysed by warming to 60°C in 1N HCl for 15 minutes. The root tips are washed thoroughly in water. A hydrolysed root tip is mounted on a slide with a drop of acetocarmine and covered with a cover slip. The slide is then gently warmed over a flame for a few seconds. The root tip is gently squashed and observed under the microscope for the cells showing the different stages of mitosis.

2. PREPARATION OF SLIDES OF FLOWER BUDS AND STUDY OF MEIOSIS.

Procedure: The onion flower buds were plucked in the morning between 8 am to 10am and then kept in a fixative (Acetic acid and methanol, 1:3) for 2-3 hours. Then, the unopened flower buds were taken out and washed thoroughly with water. A drop of acetocarmine was placed on a slide. The anther from the washed floral bud was taken and placed in the drop. The slide was gently warmed over a flame for a few seconds. The anther was covered with a coverslip and gently squashed. The different stages of meiosis were then observed under the microscope.

3. PREPARATION OF STANDARD CURVE OF DNA BY DIPHENYLAMINE METHOD

Principle: The 2 – deoxyribose of DNA in the presence of acid is converted to β – hydroxylevulnic aldehyde which reacts with diphenylamine (DPA) to form a blue-coloured complex with absorption maximum at 595 – 600 nm.

In DNA, since only deoxyribose of purine nucleotides is released, the value obtained represents half of the total deovyribose in the sample. The reactions leading to the formation of the coloured complex are as follows:



Requirements: DNA sample, saline solution (0.9%), DPA, glacial acetic acid, H_2SO_4 (conc), colorimeter, test tubes, beakers, water bath, pipettes etc.

Preparation of Reagents:

(a) Standard DNA solution($400\mu g/ml$) = (0.4g/100ml)

O.4gms of DNA is dissolved in 100ml of 0.9% saline solution.

(b) Diphenylamine Reagent:

2gms of DPA is dissolved in 200ml of glacial acetic acid. Add 5ml of conc. H₂SO₄.

Procedure:

- (i) Take different amounts of standard DNA solution (0.2, 0.4, 0.6, 0.8, 1.0ml) in different test tubes and make the volume upto 1ml by adding distilled water.
- (ii) In another test tube, take 1ml of distilled water only. This is the BLANK sample.
- (iii) Add 4ml of DPA reagent to each test tube, including the blank. Mix well. Place the test tubes in a boiling hot water bath for 10 minutes or till the solution turns blue.
- (iv) Cool the test tubes to room temperature and take O.D.(Optical Density) at 595-600nm.
- (v) Draw the standard graph by plotting the volume of standard DNA solution against the OD.
- (vi) In a separate test tube, an unknown amount of DNA will be supplied. Proceed in the same manner as mentioned above in steps 3 & 4.
- (vii)Estimate the amount of DNA present in the unknown sample from the standard graph.

Sample	Vol. of Std DNA(ml)	Vol. of dist. H ₂ O(ml)	Vol. of DPA reagent(ml)	O.D. at 595nm	O.D _{sample} – O.D _{blank}	Mean
		2 - ()				

Calculations and Results :

Standard DNA taken = $400\mu g/ml$

From the standard graph, it is seen that OD of unknown = ------ which

corresponds to -----ml of DNA solution.

1ml of standard DNA solution contains 400µg of DNA

Therefore, ------ml of unknown DNA solution contains ------µg DNA

4. STAINING AND STUDY OF NUCLEUS AND NUCLEOLUS.

Requirements: Onion peel (inner epidermis), Harris haematoxylin, eosin/safranin, different concentrations of alcohol (50%, 70%, 80%, 95%, 100%), xylene, slides, coverslips, petridish etc.

Procedure :

- (i) Immerse the sections of onion peel in the filtered Harris haematoxylin for 1 minue.
- (ii) Rinse with tap water.
- (iii) Continue rinsing till water becomes clear.

- (iv) Immerse sections in eosin/safranin stain for 1 2 mins.
- (v) Rinse with tap water till water becomes clear.
- (vi) Dehydrate in ascending alcohol solutions (50%, 70%, 80%, 95% x 2, !00% x 2)
- (vii) Clear with xylene (2X)

(viii) Mount on a slide, cover with coverslip and observe under the microscope.

V. ENVIRONMENTAL AND CONSERVATION BIOLOGY.

Study of the spatial and temporal variations in climatic factors: light, temperature, relative humidity.

Instruments used for the measurement of: Light - Luxmeter

Temperature - Maximum and Minimum thermometer

Relative Humidity - Wet and Dry Thermometer or Hygrometer

Procedure:

For the study of spatial variations in light, temperature and relative humidity, readings were measured in different areas e.g. open and shaded to see the variations of these climatic factors in contrasting spaces or areas. Readings were taken in replicates.

For the study of temporal variations in light, temperature and relative humidity, readings were measured at different timings e.g. in the morning and evening to see the variations at different timings or to show the temporal variations in climatic factors.

2. DETERMINATION OF SOIL ORGANIC MATTER CONTENT BY WALKLEY & BLACK'S RAPID TITRATION METHOD.

Requirements: Distillation flasks, burettes, measuring cylinders, $K_2Cr_2O_7$, Diphenyl indicator, [Fe(NH₄)₂SO₄], H₂SO₄, NaF, Orthophosphoric acid, distilled H₂O etc.

Procedure:

- (i) Take 1gm of soil sample and add 10 ml of $K_2Cr_2O_7$ solution.
- (ii) Add 20ml of conc. H_2SO_4 and keep for 30 minutes.
- (iii) Add 200ml distilled water
- (iv) Then add 10ml of conc. Orthophosphoric acid
- (v) Then 0.2gm NaF is added. The solution turns green
- (vi) Add 1 ml of diphenyl indicator. The colour of the solution turns muddy blue/blackish blue/ brownish blue.
- (vii) This solution is then titrated against ferrous ammonium sulphate solution [Fe(NH₄)SO₄].
- (viii) The end-point of titration is bluish green or green.

Calculations:

% of Carbon = $10 - (T \times 10/ \text{Blank}) \times 0.3$ Soil wt(1 gm) Where T = Titration value(volume of titrant) 0.3 = a constantBlank = control sample(without sample) $10 = \text{volume of } K_2 Cr_2 O_7 \text{ solution.}$

TO DETERMINE DISSOLVED OXYGEN (D.O.) OF POND AND STREAM/TAP WATER SAMPLES

PRINCIPLE: Dissolved Oxygen is of paramount importance to all living organisms and is considered to be the lone factor which to a greater extent can reveal the nature of the whole aquatic system. D.O. of the water is due to direct diffusion of oxygen from air and photosynthetic evolution of O_2 by aquatic autotrophs. The D.O. is measured by titrimetric method of modified Winkler's method.

PRINCIPLE OF MODIFIED WINKLER'S METHOD:

Oxygen combines with manganous hydroxide $[Mn(OH)_2]$ to form higher hydroxides, which on acidification with conc. H₂SO₄, liberate iodine equivalent to the amount of oxygen fixed. The iodine is titrated by standard sodium thiosulphate(Na₂S₂O₃) using starch as an indicator.

$$\begin{split} MnSO_4 &+ 2KOH &= Mn(OH)_2 &+ K_2SO_4 \\ Mn(OH)_2 &+ O &= MnO(OH)_2 & [Manganous oxyhydrate][brown ppt] \\ MnO(OH)_2 &+ 2H_2SO_4 &+ 2KI &= MnSO_4 &+ K_2SO_4 &+ 3H_2O &+ I_2 \end{split}$$

REQUIREMENTS:

Manganous sulphate(MnSO₄) solution, Alkaline-iodide solution, conc. H_2SO_4 , starch solution, Sodium thiosulphate (Na₂S₂O₃) solution (0.025N), BOD bottles, Pipettes, burettes, Burette stand, conical flasks, measuring cylinders etc.

PREPARATION OF REAGENTS:

1. Manganous sulphate solution: Dissolve 182gm of MnSO₄.H₂O in distilled water and dilute it to 500ml.

2. Alkaline – iodide solution: Dissolve separately 350gm of KOH and 75gm of KI in distilled water. Mix them and make the volume upto 500ml.

3. Sodium thiosulphate titrant(0.025N) : Dissolve 6.6205gm of sodium thiosulphate in freshly boiled and cooled distilled water and dilute to 1 litre. Add one pellet of NaOH as preservative.

4. Starch indicator: Dissolve 1gm starch (soluble) in 200ml distilled water.

PROCEDURE:

- 1. Collect two water samples each, in BOD bottles, from pond and stream/tap water without bubbling.
- 2. To each BOD bottle of each water source, add 2ml of MnSO₄ solution, followed by 2ml of Alkaline iodide solution right at the bottom of each bottle using separate pipettes.
- 3. The bottles are then shaken upside down at least six times and then left aside to allow the brown precipitate (ppt) to settle down.
- 4. The ppt. is dissolved by adding 2ml of conc. H₂SO₄. Shake the bottles again.
- 5. A sample of 50ml is taken from each bottle into separate conical flasks. This is titrated against $Na_2S_2O_3$ solution till the colour changes to pale straw.
- 6. Add 2 drops of starch solution to each sample. The colour changes to blue. The solution is further titrated till the solution becomes colourless.
- 7. The total amount of titrant used is noted and the Dissolved Oxygen content is calculated.

OBSERVATIONS:

Sample	Replicate No	Amt. of sample taken(ml)	Initial burette reading(ml) (I)	Final burette reading(ml) (F)	Amt. of titrant used(ml) [F - I]	MEAN
POND	1.					
	2.					
TAP/STREAM	1.					
	2.					

CALCULATIONS:

Since 1ml of 0.025N $Na_2S_2O_3$ solution is equivalent to 0.2mg of Oxygen

Therefore : D.O. in mg/lit = $\underline{8 \times 1000 \times N}$ v

V

Where V = volume of sample(ml)

v = volume of titrant used(ml)

N = normality of titrant(0.025N)

$$8 = 0.2 \text{mg } O_2 / 0.025 \text{N} \text{ Na}_2 \text{S}_2 \text{O}_3$$

RESULTS: D.O. Pond water	=mg/lit
D.O. Stream/Tap water	= mg/lit

TO DETERMINE B.O.D. OF POND AND STREAM/TAP WATER SAMPLES

PRINCIPLE: B.O.D. (Biological Oxygen Demand) is the amount of dissolved oxygen (D.O) required in milligrams per litre for stabilizing water under aerobic conditions in a stated time. In other words, it represents that fraction of dissolved organic matter, which is degraded and easily assimilated by bacteria. It is roughly proportional to the amount of degradable organic material present in the water sample. It is a good index of the organic pollution and therefore helps in deciding the suitability of water for consumption.

The B.O.D. of a water sample is the decrease in oxygen concentration after incubation in the dark at a certain temperature for a period of time.

Usually, B.O.D. is measured by incubating the sample at 25°C for 7 days. To determine B.O.D. it is required to find out the D.O. of water samples before incubation and after incubation. This is done by titrimetric method of modified Winkler's method.

Oxygen combines with manganous hydroxide $[Mn(OH)_2]$ to form higher hydroxides, which on acidification with conc. H_2SO_4 , liberate iodine equivalent to the amount of oxygen fixed. The iodine is titrated by standard Sodium thiosulphate(Na₂S₂O₃) using starch as an indicator.

$$\begin{split} MnSO_4 &+ 2KOH &= Mn(OH)_2 &+ K_2SO_4 \\ Mn(OH)_2 &+ O &= MnO(OH)_2 \ [Manganous oxyhydrate](brown ppt) \\ MnO(OH)_2 &+ 2H_2SO_4 &+ 2KI &= MnSO_4 &+ K_2SO_4 &+ 3H_2O &+ I_2 \end{split}$$

REQUIREMENTS: Manganous sulphate solution($MnSO_4$), Alkaline – iodide solution, H_2SO_4 , Starch solution, Sodium thiosulphate solution (0.025N), BOD bottles(125ml), burettes, burette stand, conical flasks, measuring cylinders, pipettes etc.,

PREPARATION OF REAGENTS:

- 1. Manganous sulphate solution: Dissolve 182gm of MnSO₄.H₂O in distilled water and dilute it to 500ml.
- 2. Alkaline iodide solution: Dissolve separately 350gm KOH and 75gm KI in distilled water. Mix them and make the volume upto 500ml.
- 3. Sodium thiosulphate titrant(0.025N): Dissolve 6.205gm of $Na_2S_2O_3$ in freshly boiled and cooled distilled water and dilute to 1 litre. Add one pellet of NaOH as preservative.
- 4. Starch indicator: Dissolve 1gm starch (soluble) in 200ml distilled water.

PROCEDURE:

1. Collect two water samples in two BOD bottles each, from pond and stream/tap respectively without bubbling.

- 2. To one BOD bottle of each water source, add 2ml of MnSO₄ solution and 2ml of alkaline-iodide solution one after another. The solutions are to added right at the bottom of each BOD bottle using separate pipettes. The bottles are then stoppered.
- 3. The bottles are then shaken upside down at least 6 times and then left to stand to allow the brown precipitate to settle.
- 4. The ppt is then dissolved by adding 2ml of conc, H_2SO_4 and the bottles are then shaken.
- 5. A sample of 50ml is taken from each bottle in a conical flask. This sample is titrated against sodium thiosulphate solution till the colour of the solution turns to pale straw.
- 6. 2 drops of starch solution is added to each conical flask. The colour changes to blue. The solution is further titrated till the solution becomes colourless.
- 7. The total amount of titrant used is noted and the dissolved O_2 content is calculated.
- 8. The remaining two BOD bottles from each water source are covered with black paper and incubated for 7 days at 25°C.
- 9. The same steps (2) (7) are then repeated for these bottles.

OBSERVATIONS:

non meedmied								
Samples	Repl. No	Amount of sample taken(ml)	Initial burette reading(ml) (I)	Final burette reading(ml) (F)	Amount of titrant used(ml) (F - I)	MEAN		
					/ /			

NON – INCUBATED

INCUBATED

Sample	Repl. No.	Amount of sample taken(ml)	Initial burette reading(ml) (I)	Final burette reading(ml) (F)	Amount of titrant used (ml) (F - I)	MEAN

CALCULATIONS:

Since 1ml of 0.025N Na₂S₂O₃ solution is equivalent to 0.2mg of O₂

D.O. in mg/lit = $\frac{8 \times 1000 \times N}{V}$ v

Where V = volume of sample(ml) v = volume of titrant used(ml) N = Normality of titrant(0.025N) 8 = $0.2 \text{mg O}_2/0.025 \text{N} \text{Na}_2 \text{S}_2 \text{O}_3$

VI. MICROBIOLOGY

1. PREPARATION OF SLANTS AND PLATES OF PDA MEDIUM.

Requirements : Potato(peeled) - 100gm, Dextrose(glucose) - 10gm, Agar - 10gm, distilled water - 500ml, flasks, cooton plugs, petridish, test tubes, measuring cylinders, absolute alcohol, beaker, spirit lamp etc.

Procedure : To make 500ml of Potato Dextrose Agar(PDA), 100gm of freshly peeled and cut potato is taken in a beaker and 250ml distilled is added to it. It is boiled for about 10 minutes. In a 500ml conical flask take 10gm of Dextrose and 10gm of Agar. Then add the supernatant of the potato solution and make up the volume up to 500ml by adding distilled water.

The flask is then cotton-plugged and autoclaved (at 121°C and 15lb pressure) for 15 minutes. Put off the autoclave, allow it to cool and then take out the flask containing the media. When it comes down to room temperature, open the cotton plug of the flask inside the laminar flow cabinet and pour the media into sterililized test tubes ans petridishes to make plates and slants.

2. STUDY OF EFFECTS OF GLUCOSE, FRUCTOSE AND SUCROSE ON THE GROWTH OF MICROBES.

Theory or Principle: In any biological system, growth can be defined as an orderly increase of all the chemical constituents of an organism. An increase in total mass is not necessarily a reflection of growth. It may result in cell nultiplication which can lead to an increase in the number of individuals.

Requirements: PDA media, beaker, absolute alcohol, cotton, petri-dishes, soil, inoculating needles etc.

Procedure: 0.5gm of soil was taken in an inoculating needle and placed in 3 petridishes. 1ml of sterilized water was sread over the soil in each dish. Then PDA media containing Sucrose, Glucose and Fructose respectively was poured into the petridishes. The dishes were then incubated at 30°C for 3 days. After that the effects of the three sugars on growth was studied.

3. DETERMIATION OF DENSITY OF MICROBIAL POPULATION USING HAEMOCYTOMETER.

Procedure:

- (i) The density of cells, spores etc., of microorganisms can be found out by counting the number in a unit volume. The simplest technique is to use a special counting chamber, of a type that is used in a haemocytometer for counting blood cells.
- (ii) Count the number of cells, spores in central large square. 25 medium sized squares(0.2mm x 0.2mm) in the large square are to be used. Begin counting from top row of medium –sized squares and continue to the bottom row. For cells, spores touching the lines, count only if they are on a line forming the top or right side of the square. Do not count those touching the bottom or left side of a square. This reduces the chances of counting the same cell/spore twice.

For e.g. If 220 cells are present in four large squares. 220/4 = 55 cells per large square. $55 \times 10^4 = 550,000$ cells per cm³.

4. STUDY OF GRAM – POSITIVE AND GRAM – NEGATIVE BACTERIA IN LEGUMINOUS ROOT NODULES AND CURD.

Procedure:

- (i) Take a small amount of curd/a root nodule and place it in the centre of a clean slide.
- (ii) With the help of another slide, smear the curd/root nodule to spread it uniformly.
- (iii) Fix it on the slide with the help of a burner/spirit lamp.
- (iv) Put few drops of crystal violet stain(Grams' stain) and leave for 1 minute.

- (v) Rinse the slide in running tap water.
- (vi) Add few drops of iodine solution for about 1 minute.
- (vii) Again rinse in tap water. Remove the excess water with tissue paper.
- (viii) Observe the slide under the microscope. It is observed that all the cells are deeply stained and appear purple in colour.
- (ix) To the slide, then add few drops of absolute alcohol and leave for 20 seconds. Rinse in tap water to remove the excess stain.
- (x) Then add few drops of safranin and leave for 1 minute.
- (xi) Rinse the slide in running water to remove the excess stain. Dry with tissue paper.
- (xii) Observe under the microscope.

Observations: Gram – positive bacterial cells remain purple in colour.

Gram – negative bacterial cells are pink in colour.

VII. PALYNOLOGY

1. PREPARATION OF SLIDES USING ACETOLYSIS METHOD AND STUDY OF POLLEN MORPHOLOGY.

Theory/Principle: The wall of mature pollen grains is stratified. It comprises two principal layers. The outer layer is called exine and inner layer is called intine. The terms intine and exine were proposed by Fritsche in 1837. The intine is the inner, more or less uniform layer. It is pectocellulose in nature and usually destroyed during acetolysis. The exine, which is the outer layer is chiefly composed of sporopollenin. It is derived from carotenoids by oxidative polymerization. Sporopollenin is resistant to physical and biological decomposition. The exine is acetolysis resistant layer. Because of this property of the exine, pollen grains are well preserved for long periods in fossil deposits.

Requirements: Absolute alcohol, acetic acid, acetolysis mixture, water, water bath, glycerine, glass rod, slide, coverslip, microscope, mortar and pestle, petridish, centrifuge tubes and centrifuge. Different flowers.

Acetolysis mixture: 1ml of conc. H₂SO₄ is added drop by drop to 9ml acetin anhydride.

Procedure: Crush the pollen of different flowers with absolute alcohol. Pour out the alcohol and add 5ml glacial acetic acid and centrifuge for 1 minute. Then, decant the supernatant and add 6ml acetolysis mixture. Keep the tubes in water bath and heat from 70°C to boiling point for about 15 minutes. Stir with a glass rod and centrifuge for 1 minute. Then decant the acetolysis mixture. Two tubes of the centrifuge should then be filled 1/3 rd with water and two tubes should be filled with pollen solution. Add 10ml

acetic acid and stir again and then centrifuge for 2 minutes at 1000rpm. Decant the supernatant.

Add water and shake well. Then add 2 ml glycerine to the sediment and mount the pollen in glycerine jelly on a slide and put a coverslip. Acetolysed pollens look dark brown in colour,

VIII. ECONOMIC BOTANY.

1. DETECTION OF STARCH, PROTEIN, FATS AND CELLULOSE IN PLANT MATERIALS.

TEST FOR STARCH:

A potato tuber is scraped and a concentrated solution is prepared in distilled water.

- (a) To the sample, water is added and boiled. A few drops of iodine solution was added. A blue black colour appears which indicates the presence of starch.
- (b) The solution is then boiled with HCl(starch will be hydrolysed). Now NaOH is added and 2ml of Benedict's solution is added.

A red precipitate is formed which indicates the presence of starch.

TEST FOR PROTEIN:

Soyabean seeds were soaked overnight and ground. A concentrated solution in distilled water was prepared.

- (i) Xanthoproteic test: To the filtered solution, conc. HNO₃ was added. A white precipitate is formed. Then it is heated and conc. Ammonium hydroxide is added. Yellow colour appears which changes to orange. This indicates the presence of Protein
- (iii)Biuret test: The concentrated solution is boiled and filtered. To the filtrate add 2 drops of 20% NaOH and few drops of $CuSO_4$ solution.

Violet colour appears which indicates the presence of protein.

TEST FOR FATS:

- (i) Groundnuts are crushed in a piece of white paper. If a translucent spot appears, it confirms the presence of fats.
- (ii) Groundnuts are soeked in water and sections of plant materials are cut. The sections are placed in Sudan III stain for 10 minutes and observed under the microscope.

Oil droplets are seen in the section and they will be stained red. This indicates the presence of fats

TEST FOR CELLULOSE:

- (i) Cotton fibres are taken on a slide in a drop of water. A few drops of Iodine solution is added and warm slightly over a flame. The material turns yellow which indicates the presence of cellulose. Now, a few drops of H_2SO_4 and a few drops of water are added to the material. The material turns blue which confirms the presence of cellulose.
- (ii) A thin transverse section of a dicot stem is cut. It is stained with iodine solution for 5 minutes. The section is mounted on a slide with a drop of water and then observed under the microscope.

The walls of the cells stain yellow which indicates the presence of cellulose in the cell walls.

IX. BIOCHEMISTRY

1. PREPARATION OF STANDARD CURVE FOR SOLUBLE PROTEIN BY BRADFORD METHOD.

Requirements: Bradford's Reagent, (Coumassie Brilliant Blue and ethanol 95%), Working solution, Whatman No. 1 Filter paper, Protein solution (1gm of arhar dal in 1000ml distilled water), Conical flasks, test tubes, Colorimeter, Distilled water.

Procedure:

- (i) Different test tubes were taken and in each test tube, different concentrations of protein solution (0.2, 0.4, 0.6, 0.8, 1.0 ml) was added.
- (ii) The volume was made up to 1ml by adding distilled water.
- (iii) 5ml of working solution (blue colour) was added in each test tube. The solutions are mixed gently by inverting the test tube while covering its mouth with a thumb.
- (iv) The test tube is then incubated for 10 minutes at room temperature.
- (v) OD at 595nm is taken for each test tube in a colorimeter.
- (vi) One BLANK test tube is also taken in which 1ml of distilled water and 5ml of working solution is added(no proteins are added).
- (vii) A standard graph is drawn by plotting the different concentrations of protein against the OD on a graph.
- (viii) The amount of solule protein in the unknown sample can be found out from the graph paper.

OBSERVATIONS

No. of Obs.	Vol. of std protein(ml)	Vol. of dist.water(ml)	Vol. of working soln(ml)	OD at 595nm	OD _{sample} - OD _{blank}	Mean OD

Calculations:

From the standard graph, it is observed that -----OD = -----ml of protein

Since, 1ml of standard protein solution contains 200µg of protein

Results:

The Optical Density for the unknown sample was found to be ------ and concentration of the soluble protein was found out to be ------µg/ml.

2. PREPARATION OF STANDARD CURVE FOR RNA BY ORCINOL METHOD

Requirements: Test tubes, Pipettes, Beaker, Hot water bath, measuring cylinder, colorimeter, RNA sample, saline, NaOH, Orcinol, Ethanol, Ferric chloride, Conc. HCl etc.

Procedure:

- (i) Different volumes (0.2, 0.4, 0.6, 0.8, 1.0ml) of standard RNA solution in replicates is taken in different test tubes.
- (ii) The volume in each test tube is then made up to 1 ml by adding distilled water.
- (iii) Blank tubes are prepared by taking only 1ml distilled water.
- (iv) To each test tube 3ml of orcinol reagent is added and mixed thoroughly.
- (v) The tubes are then kept in a boiling water bath for about 2 minutes or until green colour develops.
- (vi) Unknown samples of RNA are taken and processed as steps (iv) and (v) above.
- (vii) A standard graph is plotted taking the volume of RNA against OD. The concentration of unknown RNA can be obtained.

Observations:

No. of Obs.	Vol. of std RNA(ml)	Vol. of dist.H ₂ O(ml)	Vol. of Orcinol(ml)	OD at 665nm	OD _{sample} - OD _{blank}	Mean OD

Calculations:

From the standard graph, it is observed that ----- OD = -----ml of RNA Since, 1ml of standard RNA solution contains 200µg of RNA

Therefore, ------ml of unknown RNA solution contains ------µg of RNA

Results:

The amount of unknown RNA solution contains about ------µg/ml of RNA.

PREPARATION OF STANDARD CURVE FOR ESTIMATION OF STARCH BY \mathbf{I}_2 AND KI METHOD

PRINCIPLE: Starch reacts with $I_2 - KI$ reagent to give a deep – blue coloured complex known as Starch – Iodine Complex, having maximum absorbance at 580 nm. However, Iodine(I_2) does not react directly with starch since it is not very soluble in water. Therefore, I_2 is made soluble first by dissolving it in water in the presence of potassium iodide(KI). This makes a linear tri-iodide (I_3^-) ion complex which is soluble in water. It is the tri-iodide ions that react with amylose of starch to give the intense deep-blue colour of starch-iodine complex.

 $KI \rightarrow K+ I_2 + I^- \rightarrow I_3^-$ (tri-iodide complex)

 I_3 + starch \rightarrow Starch - iodine complex(deep - blue).

REQUIREMENTS : Starch stock solution(50 μ g/ml), I₂ – KI reagent, Glasswares and pipettes, colorimeter.
PREPARATION OF REAGENTS :

- STARCH WORKING SOLUTION(50µg/ml) : 100 mg of starch is dissolved in 50 ml of distilled water. The mixture is then boiled for 1 minute till a clear solution was obtained. The volume is then made up to 100 ml with distilled water. From this starch solution, 10 ml is taken and 190 ml of distilled water is added to dilute the starch solution 20 times(20X dilution)
- 2. $I_2 KI REAGENT : 0.6gm$ of KI and 60mg of Iodine are dissolved in 10ml of distilled water. 1ml of this solution is taken and the volume is then made up to 100ml with distilled water.

PROCEDURE :

- 1. Concentration gradients of starch solutions $(5 50 \mu g/ml)$ of 2ml each are prepared in test tubes by diluting the Starch Working Solution $(50\mu g/ml)$ with distilled water in appropriate proportions.
- 2. A 'Blank' is also prepared with 2ml of distilled water only.
- 3. 0.5ml of I_2 KI reagent is added to all the gradients of starch solution, including the blank.
- 4. 2.0ml of provided unknown starch solution are taken in two separate test tubes and 0.5ml of $I_2 KI$ reagent is added.
- 5. Optical Density(O.D.) of all the test tubes is taken at wavelength of 580nm. A Starch Standard Curve is generated by plotting the OD against different concentration gradients of starch solution.
- 6. The concentration of the unknown starch solution is estimated by plotting its OD from the Starch Standard Curve.

Starch	Starch	Dist.	I ₂ —KI	Absorbance(OD) at 580nm				
concn(µg/ml	working soln(ml)	H ₂ O(ml)	reagent(ml)	R 1	R2	Mean		

PREPARATION OF STARCH STANDARD CURVE :

ESTIMATION OF UNKNOWN STARCH SOLUTION :

Unknown	I ₂ –KI	Absorbance(OD) at 580nm					
starch soln.	reagent(ml)	R1	R2	Mean			

Plot a graph with starch concentration(μ g/ml) on the X – axis and O.D. at 580nm on the Y– axis and generate a linear Starch Standard Curve. The concentration of the Unknown Starch Solution is found out from the Starch Standard Curve.

RESULTS:

The generated Starch Standard Curve is a linear slope passing through the Origin.

Plotting the absorbance(OD) of the unknown starch solution from the standard curve, it was found that the unknown starch solution has a concentration of $-----\mu g/ml$.

DISCUSSION:

Starch generally contains 20 - 25% amylase and 75 - 80% amylopectin and the tri-iodide ions react only with amylase of starch to give the starch – iodine complex which is blue in colour. The strength of the blue colour depends on the amount of amylase present. However, the reaction between amylase and the tri-iodide ions is not a chemical reaction. It is a physical reaction where the tri-iodide ions intercalates within the coils of amylase molecules.

EFFECT OF TEMPERATURE ON AMYLASE ACTIVITY

PRINCIPLE : Enzymes are proteins that catalyze chemical reactions. In enzyme reactions, the molecules called **Substrates** are converted into different molecules, called **Products.** Almost all chemical reactions in a biological cell need enzymes in order to occur at rates sufficient for life.

However, enzymes are highly specific for their substrates e.g. starch which is a substrate can enzymatically be broken down into its products(maltose and other reducing sugars) only in the presence of a specific enzyme which is **amylase**.

amylase Starch \rightarrow Maltase + other reducing sugars.

Enzyme activity can be affected by many factors viz., substrate concentration, enzyme concentration, temperature, pH, inhibitors etc.

There are many methods to study enzyme activity and it can be done by either measuring the amount of substrate consumed or the amount of products formed during the reaction. This experiment is about studying the effect of temperature on enzyme activity. Here, the activity of enzyme(**amylase**) is studied at varying reaction temperatures, keeping the substrate concentration and other factors constant. The activity of amylase is measured by determining the amount of starch consumed in the reaction. The amount of starch consumed can be considered equivalent to amylase activity.

REQUIREMENTS:

Amylase extract, Starch working solution(1mg/ml), $I_2 - KI$ reagent, Acetic acid(1N), Glasswares and pipettes, Colorimeter.

PREPARATION OF REAGENTS:

- 1. **Amylase Extract** : 10g of potato is first crushed into a fine paste using a mortar and pestle. 50ml of distilled water is added to the paste and then filtered. The filtrate is centrifuged at 3000rpm for 10 minutes. The supernatant obtained is transferred to a measuring cylinder and the volume is made up to 100ml with distilled water.
- 2. **Starch Working Solution** : To 100ml of distilled water, 100mg of starch is added. The solution is heated till a clear solution is obtained.
- **3.** $I_2 KI$ Reagent : 0.6gm of KI and 60mg of iodine are dissolved in 10ml of distilled water. 1ml of this solution is taken and made up to 100ml with distilled water.

PROCEDURE:

- 1. A 'Blank' is prepared by adding 2ml distilled water, 0.5ml 1N acetic acid, 0.2ml amylase extract and 0.5ml $I_2 KI$ reagent.
- 2. Two replicas of 'CONTROL' test tubes are prepared by adding 1ml starch working solution(1mg/ml), 1ml distilled water, 0.5ml 1N acetic acid, 0.2ml amylase extract and 0.5ml I_2 KI reagent.
- 3. The FIRST absorbance reading (OD_1) of the "control" test tubes is taken at 580n wavelength.
- 4. Prepare test tubes(in two replicas) for each temperature setting (0°C, room temp., 40°(60°C, and 80°C).
- 5. To test tubes of all temperature settings, add 1ml of starch working solution followed by 1ml of distilled water.
- 6. Five new test tubes are taken and 5ml of amylase extract is taken in each one. One test tube is then kept in each temperature setting.

- 7. Both the starch and amylase containing test tubes are allowed to reach their corresponding temperature settings(about 5 minutes).
- 8. Next, 0.2ml amylase extract is added to the starch containing test tubes (under each temperature setting) only.
- 9. Reaction of the enzyme with the substrate is allowed to take place for 10 mins, at their corresponding temperature settings.(Incubation Time).
- 10. After 10 minutes, immediately add 0.5ml 1N acetic acid to all the test tubes to stop the reaction.
- 11. Allow the 40°C, 60°C and 80°C test tubes to reach room temperature.
- 12. 0.5ml of I_2 KI reagent is then added to all the test tubes of all temperature settings.
- 13. Take OD of all temperature settings at 580nm.(OD₂).
- 14. Subtract the OD₂ readings of all temperature settings from the OD₁ reading of "control".
- 15. The amylase activity at different temperature settings is observed by plotting a graph with temperature ($^{\circ}$ C) on the X axis and amylase activity (OD₁ OD₂) on the Y axis .

Test	Starch working	Distilled water	1N acetic	Amylase extract	I ₂ – KI reagent	OI	OD ₁ (580n	
lubes	soln(ml)	(ml)	(ml)	(ml)	(ml)	R_1	R_2	Mean
Blank	0.0	2.0	0.5	0.2	0.5			
Control	1.0	1.0	0.5	0.2	0.5			

TABLE I

TABLE II

Test	Starch	Distilled	Amylas	INCU BATI	1N acetic	COO LING	$I_2 - KI$	OD 2	(58	0nm)
tubes (°C)	working soln(ml)	water (ml)	extract (ml)	ON (10mi ns)	acetic acid(m l)	(40°,6 0°,80 °C)	0°,6 reagent 2,80 (ml) C)	\mathbf{R}_1	R ₂	Mean

TABLE	III
-------	-----

Temp(°C)	OD ₁	OD_2	Amylase activity($OD_1 - OD_2$)
0			
Room temp.			
40			
60			
80			

RESULTS: From the graph plotted, it was observed that at 0°C, amylase activity was very low. With increasing temperature, amylase activity was also observed to increase. However, amylase activity increased up to a temperature of ------°C beyond which, it began to decline and there was almost no amylase activity above 80°C.

DISCUSSION: At very low temperatures, the frequency of collision between enzyme molecules and their substrate molecules is very low causing the rate of the reaction(**enzyme activity**) to be very low. But with increasing temperature, the frequency of such collisions increases, thereby increasing the rate of reaction up to an optimum temperature(------°C) where the conformation of the enzyme molecules are still maintained. Beyond the optimum temperature, the frequency of collisions further increases but the enzyme molecules get denatured and their conformation to catalyze reactions is lost. This brings down the enzyme activity.

EFFECT OF SUBSTRATE CONCENTRATION ON AMYLASE ACTIVITY

PRINCIPLE : Enzymes are proteins that catalyze chemical reactions In enzyme reactions, the molecules called **Substrates** are converted into different molecules, called **Products**. Almost all chemical reactions in a biological cell need enzymes in order to occur at rates sufficient for life. However, enzymes are highly specific for their substrates e.g. starch which is a substrate can enzymatically be broken down into its products(maltose and other reducing sugars) only in the presence of a specific enzyme which is amylase.

Starch \rightarrow Maltose + other reducing sugars.

Enzyme activity can be affected by many factors viz., substrate concentration, temperature, pH, inhibitors etc.)

There are many methods to study enzyme activity and it can be done by either measuring the amount of substrate consumed or the amount of products formed during the reaction.

This experiment is about studying the effect of substrate concentration on enzyme activity. Here, the activity of enzyme(amylase) was studied at varying concentrations of the substrate(Starch), keeping other factors constant. The activity of amylase was measured by determining the amount of starch consumed in the reaction. The amount of starch consumed can be considered equivalent to amylase activity.

REQUIREMENTS :

Amylase extract, Starch working solution(1mg/ml), $I_2 - KI$ reagent, Acetic acid(1N), Glasswares and pipettes, Colorimeter.

PREPARATION OF REAGENTS :

- **1. Amylase Extract** : 10gms of potato is first crushed into a fine paste using a mortar and pestle. 50ml of distilled water is added to the paste and then filtered. The filtrate is centrifuged at 3000rpm for 10 minutes. The supernatant obtained is transferred to a measuring cylinder and the volume is made up to 100ml with distilled water.
- 2. **Starch Working Solution** : To 100ml of distilled water, 100mg of starch is added. The solution is heated till a clear solution is obtained.
- 3. $I_2 KI$ Reagent : 0.6gm of KI and 60mg of iodine are dissolved in 10ml of distilled water. 1ml of this solution is taken and made up to 100ml with distilled water.

PROCEDURE :

- 1 Concentration gradients of starch solutions(0.2, 0.4, 0.6, 0.8 and 1.0mg/ml) of 1ml each were prepared in test tubes(in two replicas) by diluting the starch working solution(1mg/ml) with distilled water in appropriate proportions.
- 2. A 'BLANK' was also prepared with 1ml of distilled water only.
- 3. Another 1ml of distilled water is added to all the test tubes including the 'blank'.
- 4. 0.5ml of I_2 KI reagent is added to all the gradients of starch solution including the blank.
- 5. The first absorbance readings(OD_1) are taken at 580nm wavelength.
- 6. 0.2ml of amylase extract is added to all test tubes including the blank and incubated at 40°C for 10 minutes.
- 7. After incubation time is over, 0.5ml of 1N acetic acid is added to all test tubes including the blank to immediately stop the enzyme reaction.

- 8. The second absorbance readings(OD₂) are taken at 580nm wavelength.
- 9. The amylase activity at varying starch concentrations is observed by plotting a graph with starch concentration(mg/ml) on the X axis, and amylase activity $(OD_1 OD_2)$ on the Y axis .

Starch conc.	Starch working	Dist. H2O	$I_2 - KI$ reagent	OD ₁ (580 nm)		$\begin{array}{c c c c c c c c c c c c c c c c c c c $			Amylase extract	Incubation for 10	\rightarrow
(mg/ml)	soln(ml)	(ml)	(ml)	R_1	R_2	Mean	(ml)	40°C			

TABLE I

Acetic		OD_2 (580 nm)		
acid	R ₁	R ₂	Mean	
(1N)(ml))				

TABLE II

Starch conc.(mg/ml)	OD ₁	OD ₂	Amylase activity($OD_1 - OD_2$)
0.2			
0.4			
0.6			
0.8			
1.0			
Blank			

RESULTS :

From the graph plotted, it was observed that at lower starch concentrations, amylase activity was low. With increasing starch concentrations, amylase activity was also observed to increase. However, at starch concentration of -----mg/ml and beyond, there was no further increase in amylase activity and it remained constant at its maximum.

DISCUSSION :

X. BIOTECHNOLOGY

AIM: PREPARATION OF TISSUE CULTURE MEDIUM AND INOCULATION OF EXPLANTS

PROCEDURE:

- 1. All glassware were washed by detergent especially designed for the purpose to remove all traces of acids.
- 2. All the ingredients (according to Murashige and Skoog) are weighted and dissolved in 1000 ml distilled water. The pH is adjusted at 5 to 6.
- 3. 15g of agar is added to the above preparation if solid medium is used,
- 4. The above culture medium and the glassware are autoclaved at 1000 C for 20 minutes and then cool it at 600 C before pouring.
- 5. Instruments such sa forceps, scalpel, needles etc. are sterilized by dipping in 95% ethanol followed by flaming and cooling.
- 6. Plants, materials (flower buds) is surface sterilized either by sodium or calcium hypochloride solution or by 95% ethanol followed by flaming and cooling.
- 7. After surface sterilized, the plant materials are rinsed with distilled water.
- 8. All operations including transfer of materials were carried out under aseptic condition i.e. under the hood of a laminar air flow cabinet.
- 9. After sterilizing by UV light, the flower buds are opened and the anthers are removed and inoculated on a nutrient medium in a petri plate.
- 10. The petriplates are then kept in a culture room.

11. IMPACT OF THE PROGRAM

Impact of DBT Support :

1. Improvement in cytological, anatomical, microbiological studies due to availability of new and better microscopes.

- 2. Availability of latest computers(Windows 8), Laptop and LCD have improved the students' understanding of various topics due to visual impact.
- 3. Addition of new Books and Journals to the Department Library has helped teachers and students to gather extra information about the subject.
- 4. Additional number of equipments has helped in more hands on involvement of students in practicals.
- 5. Availability of funds for guest lectures, field trips etc have helped to expose the students to better understanding of the subject.

Department of Chemistry:

	No. of applica	Stude			Detai	ils of admi	studen tted	ts			Position	Pg
Ye ar	nts vis- à-vis sanction ed seats	nts admitt ed	Cut off percenta ge	G	S T	S C	OB C	P H	Drop out rate	Pass Percent age	secured in Univers ity	Admissi ons to universit y
201 1	1 st Year-31	25	45%	3	22	-	-	-				
	2 nd Year-19	7	NA		7	-	-	-)			
	3 rd Year-10	1	NA	-	1	-	-	-	0%	100%		
201 2	1 st Year-35	27	50%		27	-	-	-	18.5 %			
	2 nd Year-17	19	NA	2	17	-	-	-	0%)		
	3 rd Year-16	8	NA	1	7	-	-	-	0%	100%		
201 3	1 st Year-35	37	55%	2	34	-	-	-	18.9 %)	Result Awaited (RA)	Result Awaited (RA)	Result Awaited
	2 nd Year-15	24	NA	3	21	-	-	-	0%	RA		
	3 rd Year-13	18	NA	-	8	-	-	-	0%	RA		

• The high dropout rate in the student number is due to the fact that the students leave the B.Sc. course to pursue other vocational courses.

7. Training/Exposure/Outreach

• Student Activities:

Activities	Purpose	Class	Date
 1.Industrial visit Visit to North eastern Space application Centre (NESAC) Umiam, Meghalaya Visit to industrial Estate Barapani, Meghalaya 	To Study the application of Remote sensing To study the process involved in the manufacture of cement	 B.Sc. I Year B.Sc. II Year B.Sc. I Year B.Sc. II Year B.Sc. III Year B.Sc. III Year 	14 th September 2012 20th October 2013
 2.Research Labs ii)Visit to Sophisticated Analytical Instrumentation Facility ,NEHU, Shillong 3.Training 	To understand the working mechanism and applications of some of the instruments used in biological sciences	B.Sc. I Year B.Sc. IIYear B.Sc. II Year	11 th November, 2011
Programme Attended			
4.Seminar /Workshops attended	 One day seminar on "Youth Development" organized by St. Mary's College, Shillong. A two day National seminar on " Burning Enviornmental Issues : Risk 	BSc. III year Honours students BSc 2 nd year and Final year	6 th June 2013

to Biodiversity and Human Health	students	15^{th} and 16^{th}
with special reference to North East		May 2013.
India" organized by St. Mary's		-
College, Shillong.		

Guest Lectures /Invited lectures attended by students:

Name of the guest speaker	Designation	Date	Host Institute	Торіс
Dr. D Dey	Scientific officer	15.7.13	NEHU	Instrumental Techniques and their Applications.

Training Course for the laboratory staff:

Name of the laboratory staff	Name of the workshop attended	Date	Host Institute
Ms. Serilla	Maintenance of Electronic	24 th -28 th September,	North-Eastern Hill University ,Shillong in
	Laboratory Instruments	2012	collaboration with Western Regional Instrumentation Centre, Mumbai

• Faculty Activities:

Activities	Name & Date of the	Name of	Organizing Dept./
	Workshop/Seminar	faculty	Host Institute
i.Seminar/Workshop/ Summer school attended	 i)Summer School Dt- 22nd August to 11th September,2012 ii)A two day Workshop on "Faculty Training and Motivation and Adoption of Schools and Colleges by CSIR Labs" Dt27th &28th November 2012 iii)One day Seminar on "Youth Development" held as a part of Platinum Jubilee celebrations of St. Mary's College 	Mr. M L Sawkmie All the faculty of the department	Organized by Academic Staff College, NEHU, Shillong Organized by CSIR- NEIST, Jorhat, Assam. Venue- St. Anthony's College,Shillong

Dt6 th June2013 iv)One day workshop on "Instrumental Techniques and their Applications" Dt 18 th July 2013		Organized by St.Marys College ,Shillong Organized by
v) Attended theSymposium on InorganicChemistry at Interface,	the department	department of Chemistry, St. Mary's College, Shillong.
vi) Attended a symposium on "Historical Aspects Of Chemistry and its Future Perspectives"		Organised by Department of Chemistry, IIT Kharagpur on 13 th - 14 th October 2012
	Dr. B Bhattacharjee	organized by St. Edmund's College on 15 th October 2012

8.Facilities created under the Scheme:

(i) Equipment

Department of Chemistry:

Sl. No.	Equipment Name	Quantity	Cost	Date Of Order	Purchase/Install
1.	HP P-2 1403 Desktop with monitor	2	60011.42	12.02.2013	Installed
2.	HP G6-2204TX Laptop with Carry Case	5	176190.50	12.02.2013	Installed
3.	HP Laser Jet M1536 DNF	1	23380.95	12.02.2013	Installed
4.	UPS 600 VA	03	6571.44	12.02.2013	Installed
5.	Rocker oil free vacuum pump, model	01	22800	21.12.2012	purchased

	rockyvac 400				
6.	Digital melting point apparatus	02	66,000	21.12.2012	purchased
7.	NSW water bath, model SBS4, size 605x300x175mm	1	32400	21.12.2012	purchased
8.	Gooch crucible without lid 30 ml	10	450	21.12.2012	purchased
9.	Gooch crucible without lid 40ml	10	580	21.12.2012	purchased
10.	Gooch crucible without lid 50 ml	10	640	21.12.2012	purchased
11*.	LCD Projector PG LS2000	1	32000	27.09.2013	purchased

(ii) Books and Journals

Sl. No	Title	Author	Quantity	Rate	Amount
1	An Introduction to Green Chemistry	V Kumar	2	230	460
2	Principles of NanoScience and Nano Technology	M A Shah & T Ahmed	3	195	585
3	Green Solvents for Organic Synthesis	V K Ahluwalia & R S Verma	1	525	525
4	Green Chemistry	V K Alluwalia	3	550	1650
5	Chemistry For Green Environment	M M Srivastava & R Sanghi	3	550	1590
6	Advance Organic Chemistry	R B Rao	2	1100	2200
7	Organic Chemistry 3 rd	Bruice	1	675	675

	Edition				
8	Inorganic Chemistry 3 rd Edition	Sharpe	1	650	650
9	Green solvent	Ahluwalia Verma	1	495	495
10	Organic Chemistry Vol. 1 6 th Edition	Finar	1	565	565
11	Organic Chemistry Vol 2	Finar	1	750	750
12	Organic Chemistry Vol 2	Finar	1	750	750
13	Systematic Practical chemistry	Kamboj	1	400	400
14	Group Theory in Chemistry	M s Gopinathan	1	285	285
15	Physical Chemistry Vol 1	Dr. S Pahari	1	350	350
16	Physical Chemistry Vol 2	Dr. S Pahari	1	285	285
17	An Advanced Course in Practical chemistry	Ghashal	1	385	385
18	Group Theory in Chemistry	Gopinathan	1	285	285
19	Systematic Practical chemistry	Kamboj	1	400	400
20	Organic Chemistry	P Y Bruice	1	725	725
21	Concise Inorganic Chemistry	J D Lee	2	650	1300
22	Inorganic Chemistry: A Modern Treatise	D Banerjea	2	2195	4390
23	Electronic spectra of transition complexes	R K ray	1	835	835
24	Inorganic Chemistry Principles of Structure and reactivity	J E Hueey & O K Medhi	1	650	650

25	Photochemistry	A L Gupta	2	140	280
26	Organic Chemistry vol 1	I L finar	1	750	750
27	Stereochemistry of Organic Compounds	D Nasopuri	2	295	590
28	An Advanced Cource in Practical chemistry	Ghoshal, Nad	1	385	385
29	Text book of physical chemistry	Kapoor	5	1685	1685
30	Reaction mechanism and problems in organic chemistry	P Chatopadhya	1	425	425
31	Physical chemistry	levine	1	485	485
32	Quantitative chemical analysis		1	699	699
33	Organic chemistry vol 2	S Mukherjee	1	350	350
34	Organic chemistry	Sodhi	1	495	495
35	Organic analytical chemistry	Mohan	1	395	395
36	Organic chemistry	sorel	1	995	995
37	Physical chemistry	donal	1	1	795
38	Physical chemistry	levine	1	795	795
39	Basic inorganic chemistry	F a cotton	1	350	350
40	Basic organometallic chemistry	gupta	1	675	675
41	Advance inorganic chemistry	F A Cotton	1	579	579
42	University chemistry	P Siska	1	699	699
43	Organic Chemistry	Crary	1	699	699
44	Vogel's Qualitative inorganic analysis		1	295	295

45	Advance Practical	Vishnoi	1	250	250	
	Organic Chemistry	VISIIIOI	1	230	<i>4</i> 00	
46	Organic Chemistry	I L Finar	1	799	799	
	organic chemistry		-	177	())	
47	Organic Chemistry	Graham	1	769	769	
48	Fundamentals of					
	Reaction mechanism in organic chemistry	Narain	1	275	275	
49	Text Book Of Chemistry	Venugopal	1	199	199	
50	Objective Chemistry	Arun	1	395	395	
51	Reaction Mechanism in organic chemistry	Mukherjee	1	400	400	
52	Text book of Pharmaceutical Chemistry	Mathew	1	395	395	
53	Practical in Physical Chemistry	Sindhu	1	195	195	
54	Stereochemistry of organic compounds	Nasipuri	1	295	295	
55	Organic Name Reaction	Gautam	1	395	395	
56	Chemistry of Natural products	Bhat	1	1700	1700	
57	Organic Chemistry through solved problems	Goutam	1	395	395	
58	Natural Products	Sujata	1	525	525	
59	Green Chemistry	Rashmi	1	550	550	
60	Organic Chemistry for undergraduate	Pillai	1	595	595	
61	Atkins Physical Chemistry	Oxford	1	725	725	
62	Introduction to modern inorganic chemistry	Мс Кау	1	2696	2696	

63	Principles of Biochemistry	Lehninger	1	2066	2066
64	Food Chemistry	Chopra	1	440	440

Laboratory Equipments/Chemicals Under Recuring

Sl. No.	Description of goods	rate	per	quantity	Amount
1	Periodic table chart	180	each	2	360
2	Periodic table elements	180	Each	2	360
3	Stop watch	2437	Each	5	12,185
4	Spirit lamp	50	Each	4	200
5	Burette 50ml	484	Each	12	5,808
6	Oil bath	580	Each	12	6960
Vat @	213.5				3492
Total					29365/-
7	R B Flask	170	Each	10	1,700
8	Condenser	470	Each	10	4,700
9	Guard Tube	80	Each	10	800
10	Beaker	61	Each	10	610
11	Pipette	148	Each	10	1,480
12	Volumetric Flask	277	Each	10	2770
13	Vo. Flask	185	Each	10	1,850
14	Pipette	120	Each	10	1,200
	Vat @13.5			· · ·	1877
Total					15,778/-

Sl. No.	Description of goods	rate	per	quantity	Amount			
1	Manganous Chloride	410	500g	2*500	820			
2	Potassium Sulphate	280	500g	3*500	840			
3	Ferrous Sulphate	146	500g	6*500	876			
4	Amm. Ferrous Sulphate	140	500g	3*500	420			
5	Ortho phosphoric acid	450	500g	24*500	2,700			
6	Sulphuric Acid	230	500ml	12	5,520			
7	Ortho phosphoric acid	450	500ml	6*500	2,700			
8	Manganous chloride	410	500g	1*500	410			
	Vat @ 5%							
	Tot	al			15,000/-			
9	Silver Nitrate	4539	25g	1*25	4539			
10	Iodine	1998	100g	1*100	1998			
11	Nickel Sulphate	1706	500g	1*500	1706			
12	Cobalt Nitrate	2620	500g	1*500	2620			
13	Nickel Chloride	1877	100g	1*500	1877			
14	Dimethy Glyoxime	457	100g	1*100	457			
15	Amyl Alcohol	396	500ml	1*500	396			
Vat@	Vat@ 5%							
Total	Total							

SOP

Experiments Conducted For First , Second and Third Year Students <u>Physical Experiments:</u>

AIM:- Determination of the molecular weight of naphthalene by rast's method.

THEORY:- The addition of the solute decreases the freezing point of the solvent. The extent of decrease in the freezing point depends on the concentration of the solute. If a non-

volatile solute is dissolved in a solvent of known molarity, the solution is sufficiently diluted, the depression of freezing point, ΔTf is given by

$$\Delta Tf = Kf \times Cm = \frac{Kf \times 1000 \times W2}{M \times W1}$$
$$M = \frac{Kf \times 1000 \times W2}{\Delta Tf \times W1}$$

Where, Kf is the molar depression of the solution, that is the depression per gram mole of the solute dissolved in 1000 gram of the solvent,

W2 = gram of the substance of molecular weight M.

 $\Delta Tf =$ depression of freezing point.

The molar depression of camphor is usually 40. The used of solvent for cryoscopic work is made use of naphthalene for determination of molecular weight of the solute which forms homogenous mixture and does not form a compound with camphor.

APPARATUS REQUIRED:- melting point apparatus, Thermometer with gradation 0° , 1° -200°C, flat-bottomed test tube, chemical balance, capillary tube and watch glass.

PROCEDURE:- 5 gram or 500 milligram of camphor is taken in a clean and dry test tube. To this 0.5 gram or 50 milligram of the solute (Napthalene) for which the molecular weight is to be determined is added. The mouth of the test tube is closed with cotton wool. The test tube is slowly heated in an oil bath. Camphor melt first and then naphthalene it is then shaken thoroughly. It is dried and taken in a watch glass. Few crystals of it is introduced into a capillary tube and the melting point is noted down. In another capillary tube. crystals of pure camphor is introduced and the melting point of pure camphor and melting point of the mixture give Δ Tf.

<u>AIM</u>: To study the adsorption of oxalic acid on activated charcoal and to prove the validity of Freundlich adsorption isotherm.

<u>THEORY</u>: The molecule (or atom or ions)on the surface of liquid or solid are relatively fixed and the moving molecules in the surrounding gas or liquid phase interact with relatively fixed molecule of and of the result they are hold by the surface by rather weak forces. Adsorption may be defined as a accumulation or superficial adhesion of molecules of an interphase. Generally, the accumulation is only one molecule thick and is referred to as monomolecular adsorption. The solid which takes up gas vapour or solute from a solution is known as adsorbent while the gas or the solid which is held to the surface of the solid is called adsorbate.

The amount of the substances adsorbed is proportional to the concentration of this solution and is given by the relation (Freundlich adsorption isotherm)

x = kc'(1)

Where,

x = amount of solute adsorbed

m = quantity of adsorbent

k = a constant depending upon the nature of both adsorbent and adsorbate

c = equilibrium concentration of adsorbate in solution

n = a constant depending upon the nature of the adsorbate.

The value of 1\x is generally more or less than unity. Taking logarithm of equation 1, we get

 $\text{Log } x \mid m = \log K + 1 \mid xc$

If the value of log xm is plotted as co- ordinate against log c as abscissa, we get a straight line with a slope 1x and intercept on the co-ordinate log K.

APPARATUS REQUIRED:-

Burette, pipette, reagent, bottle, charcoal (activated)

PROCEDURE

- 1. 250ml of 0.5 (N) oxalic acid solutions is prepared by weighing out exactly 7.878 gms of oxalic acid and dissolving it in water to make 250ml. Again 250ml of 0.5 (N) NaOH solutions is prepared. NaOH solution prepared is then titrated against standard oxalic acid prepared about to find out the exact normality of NaOH.
- 2. Four stoppered bottle are taken, cleaned and dried and the following solution are prepared. Each of the bottles are labelled as 1,2,3,4.
- 3. Now in each of these bottles, 1gm of activated charcoal is added, then by means of a measuring cylinder 50, 40, 30, 20ml of 0.5(N) oxalic acid and 0, 10, 20, 30ml of distilled water is transferred respectively to each of these bottles and stoppered. The flasks are shaken well for about 7-8 minutes. It is then filtered and 10ml of this solution is titrated standard NaOH solution using phenolphthalein indicator.

Bottle no.	N\2 oxalic	Distilled	Amount of	0.5(N) NaOH
	acid(ml)	water(ml)	charcoal(gm)	solution(ml)
1				
2				
3				
4				

OBSERVATION:

In the last or beginning, titrate the stock solution of acetic acid (10ml) also by means standard (N10) NaOH solution.

Bottle	Initial	Equilibrium	Log	Amount of	$X \setminus M$	Log	10 +
no.	concentration(C ₀)	concentration	Ce	acid (C_0 –		X\M	log
	of acid before	of acid after		C _e)adsorbed			Ce
	adsorption(ml) of	adsorption(ml		(ml of			
	$NaOH(C_0)$	of NaOH)		NaOH)(x			
		(C _e)		gm)			
1							
2							
3							
4							

Let 10ml of stock acetic acid solution = x ml of (N\10) NaOH.



CALCULATION

$X=C_o-C_e$

M=1 gm in each case.

CONCLUSION

Thus we can calculate $x \in ach$ bottle and find the value of log $x \in ach$. The logarithm of Ce term is also calculated in each case.

Now a graph is plotted with log $x\mbox{m}$ as ordinates and log Ce as abscissa. We get a straight line. The slope of this line will thus be equal to $1\mbox{n}$. This proves the validity of Freundlich adsorption isotherm.

PRECAUTION

- 1. The flask should be absolutely clean and dry.
- 2. NaOH used should be free from CO₂.
- 3. For titration, small filter paper should be used so that error due to any adsorption of the acid by the filter paper is minimised.

Aim: Determination of heat neutralization of a strong acid by a strong base.

Theory: Chemical reaction are accompanies by thermal change where heat is either evolved or absorbed during the reaction, i.e an exothermic and endothermic. Heat of reaction depends on the nature of the reactant and quality of the reactants on strong acids and bases like HCl and NaOH are regarded as undergoins complete ionization in aqueous solution when equivalent amount of HCl and NaOH are mixed the reaction is H+Cl+Na+OH NaCl +H2O + H

Heat of neutralization is defined as the heat evolved in kilo joule, when 1g equivalent weight of an acis is neutralized by 1g equivalent weight of an alkali. This is the enthalpy change introducing 1g mole of water.

APPARATUS AND MATERIAL REQUIRED

- 1. A calorimeter fabricated in the laboratory with a glass stirrer.
- 2. Thermometer
- 3. Stop-watch
- 4. Two beaker of 250ml capacity.
- 5. 100 ml of 2(N) HCl, 100ml of 2(N) NaOH.
- 6. Distilled water
- 7. One measuring cylinder of capacity 100ml.

Procedure:-

STEP 1 Determination of water equivalent of the calorie meter. A beaker of 250 ml capacity I wrapped with insulating material generally, a brown paper and this beaker is placed in another beaker of 500 ml capacity. This forms the laboratory calorie meter. The inner beaker must have a lid made up of insulating material with two holes, one for inserting the thermometer and other for inserting the glass stirrer. The thermometer must be inserted in such a way that the bulb nearly touch the bottom of the beaker. 100ml of water is taken in a calorie meter and is uniformly stirred and the temperature is noted down every minute for about 10 minutes, when the temperature is steady 100 ml of hot water of the temperature of which is noted exactly abut 50-70' c about 40' higher than that of the cold water in the calorie meter is added quickly and the mixture is stirred thoroughly, the time at which the hot water is added is also noted down stirring is continue and the temperature become uniformed. Water equivalent of the calorie meter is then calculated.

STEP 2 100ml of 2(N) HCl is transferred into an insulated beaker and the temperature of an acid is noted down after an interval of 1 minute for about 10 mins, 100ml of 2(N) NaOH solution is also noted, stirred in another beaker and the temperature of this alkaline solution is also stirred down when the temperature of the acid in calorie meter and the base kept in a separate beaker are ready, the NaOH solution is added to the acid in the calorie meter and stirred well. The final temperature is taken every minute for about 15-20 mins.



OBSERVATION AND CALCULATION

Volume of cold water in the calorie meter = ____ ml initial temperature of water in caloriemeter = T1'c=

Volume of hot water added = ____ml

Temperature of hot water added= T2'c=_____

Temperature of mixed water=T3'c=____

Heat taken by beaker and water=volume of water(T3-T1) cal, when M1 and S are the weight and specific heat of the beaker and stirred specific heat of ater taken as 1 g/ml.

Heat given out by hot water = volume of water (T2-T3) Calculated heat gain = Heat lost M1S+volume of water (T3-T1)= volume of water (T2-T3) (M1S+100) (T3-T1)=100(T2-T3) M1s=100(T2-T3)-100(T3-T1)/T3-T1

Table 2

Time in mins.	Initial temperature of HCl (T4'c)	Initial temperature of NaOH(T5'c)	Temperature of mixture	Final temperature from graph (T6'c)

Graph



Calculation Volume of 2(N) HCl=___ml Volume of 2(N) NaOH=___ml Initial temperature of NaOH= T4'c=___ Initial temperature of HCl= T5'c=___ Final temperature of mixture = (HCl+NaOH) T6'c=___ Rise in temperature (t)=T6-(T4+T5)/2

Heat produced on mixing two solutions

 \triangle =(M \Rightarrow +200)X t Cals

Therefore, Heat of neutralisation = (X 1 / 20) X 4.184 / 2X 100

Conclusion

Since the reaction between HCl and NaOH is exothermic, therefore heat of neutralization has a negative value. Therefore, water equivalent of calories meter = _____ calories.

Heat of Neutralization of HCl and NaOH=____KJ

EXPERMENT 2

Determination of the heat of dilution of H2SO4 and hence to determine the strength of an unknown sample of H2SO4.

THEORY: The main aim of the experiment is to determine the change in heat contents. H occupying the dilution of H2SO4. The fact that heat of solution varies with its concentration implies that there must be a change in enthalpy when a solution containing a molecule of solutes is diluted from one into another. For the sake of simplicity the experiment is carried out adiabatically. The water equivalent of the calories meter is determined, then the heat-evolved. When a known amount of acid of particular strength is added to a definite amount of water and the heat of dilution is determined.

APPARATUS REQUIRE: Calorimeter (Fabricated in the laboratory) 2 beaker, Pipette and H2SO4

Procedure

Step1: determination of water equivalent of caloriemeter.

A beaker of 250ml capacity is wrapped with a insulating material like brown paper and is placed in another beaker of 500ml capacity.

This form a laboratory caloriemeter. The inner beaker must have a lid made of isulating material with two holes i.e on for inserting the thermometer and the other for the glass rod

(stirrer). The Thermometer must be kept in such a way that the bulb nearly touches the bottom of the beakers.

100ml of H2O is taken in the calorimeter and the glass stirrer is placed in the caloriemeter water in the caloriemeter is uniformly stirred and the temperature is noted down every minutes for about 10 mins when the temperature of which is noted down exactly about 50-70 i.e about 40'c higher than that of the cold water in the caloriemeter is added is also noted down. Stirring is continued and the temperature is noted as intervals of one minute for every 15 minutes until the fall of temperature become uniform water equivalent of caloriemeter is then calculated.

STEP 2

Determination of the heat of dilution of H2So4 100 ml of distilled water is transferred into a beaker and the water is regularly stirred and the temperature of water is noted exactly 10ml of 2(N) H2So4 is added. The water in the beaker and the mixture is stirred regularly and the temperature is noted down at regular intervals of one minute. The process is continued for abpout 20 mins until the fall of temperature for unit time is uniform. The change in temperature is obtained after applying the radiation connection. The inner beaker of calorie meter is washed and dried. The above process is repeated with other samples of acid whose strength are known and with the unknown supplied acid.

Time in mins	Temp. of cold water (T1'c)	Temp. of hot water (T2'c)	Final temp of m	ixture (T3'c)
			Temp of mixture	From Graph

Table 1



Volume of cold water in the calorie meter=100ml initial temperature of water in caloriemeter=T1'c=____

Volume of hot water added=____ml

Temperature of mixed water = T3'c=____

Heat taken by beaker and water=volume of water (T3-T1) cal, when M1 and S are the weight and specific heat of the beaker and stirred specaftc heat of water taken as 1g/ml. heat given out by hot water= volume of water (t2-t3) calories. Heat gained= Heat lost M1s+volume of water (T3-T1)= volume of water (T2-T3)

M1S = 100(T2-T3)-100(T3-T1)/T3-T

Table

Strength of H2So4	Temp. of Cold water (T4)'c	Tiime in Mins.	Temp. of Mixture	Final temp. of mims. (T6)'c	<u></u> ∆T=T6-T4
2(N)					
3(N)					
4(N)					
5(N)					
Unknown					



OBSERVATION AND CALCULATION

(A) 10ml of 2(N) H2so4 is added to 100ml of water taken in the caloriemeter Volume of water taken=___ml

Volume of acid taken=____ml Strength of H2so4 = 2(N) Temperature of cold water =t4 =____ Temperature of Acid before mixing = t5 =____ Temperature of acid water mixture = t6 =____

 \triangle H for 2(N) H2so4=(M1S+100+10) [t6-(t4+t5)/2] cals.

(B) 10ml of 3(N) H2so4 is added to 100ml of water taken in the calorimeter Volume of water taken=___ml Volume of acid taken=___ml Strength of H2so4 = 3(N) Temperature of cold water =t4 =___ Temperature of Acid before mixing = t5 =___ Temperature of acid water mixture = t6 =____

 \triangle H for 3(N) H2so4=(M1S+100+10) [t6-(t4+t5)/2] cals.

(C) 10ml of 4(N) H2so4 is added to 100ml of water taken in the caloriemeter Volume of water taken=___ml Volume of acid taken=___ml Strength of H2so4 = 4(N) Temperature of cold water =t4 =___ Temperature of Acid before mixing = t5 =___ Temperature of acid water mixture = t6 =

 \triangle H for 4(N) H2so4=(M1S+100+10) [t6-(t4+t5)/2] cals.

(D) 10ml of 4(N) H2so4 is added to 100ml of water taken in the caloriemeter Volume of water taken=___ml Volume of acid taken=___ml Strength of H2so4 = 4(N) Temperature of cold water =t4 =___ Temperature of Acid before mixing = t5 =___ Temperature of acid water mixture = t6 =

\triangle H for 4(N) H2so4=(M1S+100+10) [t6-(t4+t5)/2] cals.

(E) 10ml of unknown (N) H2so4 is added to 100ml of water taken in the caloriemeter Volume of water taken=___ml
Volume of acid taken=___ml
Strength of H2so4 = unknown (N)
Temperature of cold water =t4 =____
Temperature of Acid before mixing = t5 =____
Temperature of acid water mixture = t6 =____

 \triangle H for unknown (N) H2so4=(M1S+100+10) [t6-(t4+t5)/2] cals.

Results

The Heat of dilution of 2(N) H2So4 = _____cals. The Heat of dilution of 3(N) H2So4 = ____cals. The Heat of dilution of 4(N) H2So4 = ____cals. The Heat of dilution of 5(N) H2So4 = ____cals. The Heat of dilution of unknown (N) H2So4 = ____cals.

Conclusion

A graph of \triangle H against the concentration of the acid was platted and theunknown strength of the acid was calculated from the graph and was found to be 6(N).

<u>AIM</u>: To determine solubility of the given salts at two temperature and hence to determine the heat of solution.

THEORY: When a small quantity of solid substance (solute) is shaken in a fixed quantity of liquid (solvent) the solid dissolve. If one goes on adding more and more solute the resulting solution becomes saturated and further dissolution of the solute is arrested.

Solubility of a substance at a definite temperature is defined as the amount of solute dissolved per 1000gm of solvent. So as to make a saturated solution at that temperature, it depends on temperature, nature of solute and solvent. If the process is endothermic the solubility decrease with temperature.

If W_{A} is the amount of the solute dissolved on W_{B} gm of solvent the solubility of solute is given by

 $S = W_A \setminus W_B *1000$

Solubility = Mass of residue Volume of solution evaporate * 1000gm/L of solution

Where S = Solubility of the solute, the solubility is related to the heat of the solution as

 $d \ln S dt = Q Rt^2$

 $Log S_2 \backslash S_1 = Q \backslash 4.576 * T_2 - T_1 \backslash T_2 T_1$

Where Q= the heat solution, S_2 and T_2 refers to the higher temperature and S_1 and T_1 refers to lower temperature.

APPARATUS AND MATERIALS REQUIRED

- 1. Two beaker of 100ml capacity
- 2. Two porcelain dishes 15.20ml capacity
- 3. Glass rod
- 4. Thermometer
- 5. Constant temperature water bath
- 6. Balance weight
- 7. The given sample

PROCEDURE:

100 ml of solvent (water or alcohol) is taken in a clean beaker and the sample (Bacl₂\ Benzoic acid) is added with constant stirring till a saturated solution is obtained. The temperature of the solution is noted 25ml of the solution of the sample (Bacl₂\ Benzoic acid) is pipetted out and transferred into a previously weighted Porcelain dish and is weight is taken again the solution is slowly allowed to evaporate on a water bath till the solvent is fully evaporated and a solid residue is kept in the porcelain dish. The outer surface of the porcelain dish is wiped off. The dish containing the solid is weighed again. In another beaker, 100ml of water is taken and the beaker is placed in a constant temperature both at 50⁰ C saturated solution of the sample is prepared. At that temperature 25ml of supernated solution is quickly transferred into a weighed porcelain dish and then evaporated to dryness. The weight of the porcelain dish and the solid is taken.

SOLUBILITY TABLE OF BARIUM CHLORIDE Bacl2

Temperature of Experiment = 0° c	At room temperature T_1 =	At high temperature $T_2 =$
1. Weight of porcelain dish in		
gram (W ₁)		
2. Weight of porcelain dish solid		
after evaporating gram (W ₂)		
3. Weight of solid in gram (W_2 –		
W_1)		
4. Solubility, $S = (W_2 - W_1) \setminus 5$		
1000gm\L		

OBSERVATION AND CALCULATION

Weight of porcelain dish = W_1 gram Weight of porcelain dish + Solid after evaporation = W_2 gm Weight of solute = $(W_2 - W_1)$ gm Solubility of the sample is gm\L of solution is calculated as, Solubility = Mass of residue\Volume of solution evaporated * 1000 To determine the heat of solution 'Q' Log $S_2 \setminus S_1 = Q \setminus 4.576 * T_2 - T_1 \setminus T_2 T_1$ $Q = 4.576 * \log S_2 \setminus S_1 * T_1 T_2 \setminus T_2 - T_1$ Heat of solution 'Q' of Bacl₂ is given by Log $S_2 \setminus S_1 = Q \setminus 4.576 * T_2 - T_1 \setminus T_1 T_2$ And $Q = 4.576 \log S_2 \setminus S_1 * T_1 T_2 \setminus T_2 - T_1$ **CONCLUSION** Solubility of BAcl₂ at room temperature $T_1 =$ Solubility of BAcl₂ at high temperature $T_2 =$

Heat of solution of $Bacl_2 =$

Aim : To determine the partition co-efficient of benzoic acid.

REQUIREMENT:

Four small glass stoppered bottles, 5ml pipette, burette, titration flask, 0.1 (N) NaoH solⁿ, benzene, benzoic acid, phenolphthalein.

THEORY:

At a given temperature, the partition co-efficient is given by $K = {c_1/c_2}$, where c_2 is the concentration of benzoic acid in benzene, c_1 is the concentration of benzoic acid in water, let 'n' be the no. of molecule of the solute associated in one of the above solvent, the law is expressed mathematically as, $\frac{c_1}{\sqrt{c_2}} = K$.

Where n = no. of molecule which associates is solvent 2, benzoic acid is found to exist in simple molecule in water but in benzene, its two molecule are found to associate, therefore this can be verified y determining the ratio between c_1 and $\sqrt{c_2}$

PROCEDURE:

- 1) 0.1(N) NaoH is prepared by dissolving 1gm in 250ml of H₂O (Volumetric flask)
- 1% of benzoic acid solution is prepared by dissolving 2.5gms in 250ml of water (Volumetric flask)
- 3) Four stoppered bottles are taken and labelled to each 50ml of benzene is added. Then to them –

- a) 50ml of benzoic acid
- b) 40ml of benzoic acid+10ml of water
- c) 30ml of benzoic acid+20ml of water
- d) 20ml of benzoic acid+30ml of water

These bottles are shaken vigorously and allowed to stand for sometime till the two layers separated.

- 4) 5ml of upper layer i.e, benzoic acid layer is pipette out from each of the bottle. To each titration flask is added 1-2 drops of Phenolphthalein indicator. This coloured solution titrated against 0.1 (N) NaoH solution, till the pink colour disappears. This volume is noted down.
- 5) The upper end of the pipette is tightly closed with finger and the nozzle is pushed into the water layer, in one bottle, 5ml is pipette out and this is titrated against 0.1 (N) NaoH solution using phenolphthalein indicator.

Similarly, the other bottles are also titrated K, the partition co-efficient of benzoic acid is given by –

$$K = \frac{K_1 + K_{11} + K_{11} + K_{11}}{4}$$

OBSERVATION AND CALCULATION:

Preparation of 0.1(N) NaoH in 250ml of water 1(N) of NaoH in 1000ml requires 40gms

0.1 (N) of NaoH in 1000ml of requires 40x0.1gm = 4gms

0.1 (N) of NaoH in 250ml requires $\frac{40x0.1}{4} = 1$ gms.

-	ът	-	
ĽΔ	RL	H.	1.
IN	DL		1.

Bottle no.	vol ^m of benzoic $\bar{\alpha}$ in $H_2O(ml)$	vol ^m of benzene(ml)	vol^m of $H_2O(ml)$
1	50	50	0
2	40	50	10
3	30	50	20
4	20	50	30

TABLE 2: Aqueous layer

Bottle no.	vol ^m of pipette (ml)	vol ^m of 0.01 (N) NaoH	$c^1 = \frac{B.R \ x \ 0.01}{5}$
1			
2			
3			
4			

TABLE 3 : Organic layer

Bottle no.	vol ^m of pipette (ml)	vol ^m of 0.1 (N) NaoH	$c^1 = \frac{B.R \times 0.01}{5}$
1			
2			
3			
4			

TABLE 4:

Bottle no.	C ₁	C ₂	$\sqrt{c_2}$	$K = c_1 / \sqrt{c_2}$
1				
2				
3				
4				

$$K = \frac{K\iota + K\iota\iota + K\iota\iota + K\iota\nu}{4}$$

RESULT:

____·

The partition co-efficient of benzoic acid between water and benzene is found to be

GRAPH:

A graph is plotted against volume of aqueous layer c_1 an square root of organic layer, $\sqrt{c_2}$, which gives a straight line. This proves the complexity of molecules of benzoic acid in benzene in two form that is the molecular state of benzoic acid in benzene is dimer.

$$K = slope = \frac{y}{\chi}$$

Inorganic Practicals:

Course number CHEM -224

INORGANIC PRACTICAL

QUANTITATIVE ANALYSIS

COMPLEXOMETRIC TITRATION OF Mg+2 USING E.D.T.A.

[1] standardisation of E.D.T.A.

[A] fill the burrete with e.d.t.a

[b]make up the volume of the zinc solution

[c]Pippette out 25 ml of the solution and add 3 test tube of water and half test tube of buffer solution

[d] add 2 drops of eriochrome black t indicator

[e] solution is warmed slightly

[f] It is then titrated with edta till the colour turns blue from red.

[2]ESTIMATION OF MAGNESIUM SOLUTION OF UNKNOWN STRENGTH.

[1]25 ml of the solution of unknown strength is taken in conical flask

To this soltion 3 test tube of water half test tube of buffer is added

[2] Few drops of indicator is added

- [3] The solution is titrated with standardised EDTA solution
- [4]The end point is blue in colour.
[B] PROCEDURE OF ESTIMATION OF COPPER AND ZINC

SEPARATION OF COPPER AND ZINC

[A]25 ml of the solution is pippeted out.To it dilute ammonium hydroxide is added till a turbidity is produced

[b] the ppt is dissolved in dilute Hcl and sodium sulphite is added

[c]To it freshly prepared NH4 SCN is added

[d]copper is precipated out as copper thio cyanate It is filtered and separated from zinc.

ESTIMATION OF ZINC GRAVIMETRICALLY.

[1] To the above filterate 2 drops of methyl orange [indicator]is added.

[2] Excess of acid is neutralised by adding 2 drops of NH4 OH and buffer ammonium acetate and ammonium chloride is added

[3] The solution is heated to boiling 10 ml of ammonium hydrogen phosphate is addede

[4]The solution is allowed to stand for sometime

[5] It is then filtered through sintered glass crucible

[5] The ppt is washed with water and dried at 1100 c in the hot air oven.

[C] ESTIMATION OF CALCIUM AND BARIUM IN A MIXTURE.

[I]SEPARATION OF BARIUM FROM CALCIUM.

[I]Barium is precipated out as bariumchromate from the solution.

[A]25ml of the soluton is pippeted out from the volumetric flask after making up the volume.

[b]The solution is neutralised with NH4 OH and glacial acetic acid and ammonium acetate is added

[c] The solution is heated to the hot solution K2CrO4 IS Added and barium is precipated out as barium chromate. The ppt is filtered through filtr paper.

[d]The ppt is dissolved in hot dilute HCl and titrated iodometricall

[2] ESTIMATION OF CALCIUM FROM THE FILTERATE

[I]Cal cium is precipated from the filterate as calcium oxalate.

[a] To the filterate NH4 OH solution is added till alkaline

[b] It is then heated to the hot solution ammonium oxalate is added. The white ppt of calcium oxalate is filtered through what man 40

[c] The ppt is dissolved in hot dilute sulphuris acid and titrated prmangano metricaly.

<u>Aim</u>: To estimate the amount of Cu^{2+} and Zn^{2+} ion present in a given mixture volumetrically.

1.Standardization of Hypo using 0.05(N) K2Cr2O7:-

Pipette out 25ml of standard $K_2Cr_2O_7$ solution in a conical flask and to it add ¹/₂ test tube of conc. HCl and 1gram of *NaHCo*₃. Dilute with H₂O, add 2gram of 10% potassium and keep the solution in the dark for 5minute. Filtrate the liberate iodine with hypo solution until the colour becomes yellow. Add few drop of starch solution to it and continue the titration until blue colour just disappear leaving the solution light green in colour.

For Cu^{2+} and Zn^{2+}

Material used

(i) $CuSO_4$ solution+ $ZnSO_4$ soln 0.1(N), NH_4OH (1:1) glacial acetic and 5ml; KI-2gram, Hypo solution (0.1) N for Cu^{+2} .

(ii) H₂SO₄ Di-Ammonium Hydrogen Phosphate 20% NH3 1.5g for Zn^{+2} .

For estimation of Copper: 25ml of the solution is pipette out in a conical flask. The solution is neutralized with *NH4OH* solution till smell of ammonia is produced and turbidity appear. 5.6ml of glacial acetic acid is added along with 2gram of KI. The flask is kept in the dark for 5minutes. The solution is then diluted in 150ml and the liberated iodine is titrated with standard hypo using starch indicator.

2.Separation of Cu^{+2} and Zn^{+2}

25ml of the solution is pipette out. To it dil. NH_4OH is added till a turbidity is produced. The ppt. is dissolved in few drops of HCl and to it 2gm of $Na_2S_2O_3$ is added and the solute is diluted to 100ml and to it 20ml of freshly prepared NH_4SCN solution (10%) is added by stirring. A white ppt. of *CuSCN* is formed. The solution is allowed to stand for some time and the ppt. is filtered through filter paper. The ppt. is washed with 10% NH_4SCN and few drops of Sulphurous acid. Washing is done 2-3 times taking small portion at a time.

<u>3. EStimation of Zn^{+2} </u>: to the filtrate, add 2drops of methyl red indicators, excess of acid is neutralized with 1.5 *NH*₄*OH* till the solution is yellow. To it 1gram of ammonium acetate and 1.5 gram of 100-150 ml of diammonium hydrogen phosphate solution is added with constant stirring.

The solution is allowed to stand for sometimes the ppt. is filtered through a weighed standard sintered glass crucible. The ppt. is washed with 1% diammonium hydrazine phosphate till the filtrate is chloride free. It is then washed with H₂O and dried at 110° C.

Organic Practicals

Organic Practicals

Organic Preperations:

Phthalimide

Intimately mix 9.9 g of pure phthalic anhydride and 2g of urea, and place the mixture in a 500 ml, long;necked, round-bottomed flask. Heat the flask in an oil bath at 130-135° C. When the contents have melted, effervescence commences and gradually increases in vigour; after 10-20 minutes, the mixture suddenly froths up to about three times the original volume and becomes almost solid. Remove the flame from beneath the bath and allow to cool. Add about 80 ml of water to disintegrate the solid in the flask, filter at the pump, wash with a little water and then dry at 100° C.

M.pt-

Yield-

m-Dinitrobenzene

Place 37.5 g(21 ml) of conc. Sulphuric acid and 22.5g(15 ml) of fuming nitric acid in a 250ml round-bottomed flask; add a few fragments of unglazed porcelain. Attach a reflux condenser and place the apparatus in a fume cupboard. Add slowly , in portions of about 3 ml,15g(12.5ml) of nitrobenzene; after each addition, shake the flask to ensure thorough mixing. Heat the mixture , with frequent shaking, on a boiling water bath for 30 minutes. Allow the mixture to cool somewhat and pour it cautiously with vigorous stirring into about 500ml of cold water; the dinitrobenzene soon solidifies. Filter, wash thoroughly with cold water. Recrystalize from rectified spirit to get colourless crystals of m-dinitrobenzene.

M.pt :- , Yield:-

Picric Acid

Place 9.5g of phenol in a dry 1-litre flat-bottomed flask and 12 5 ml of concentrated sulphuric acid, shake the mixture and heat it on a boiling water bath for 30 minutes ,and then cool the flask thoroughly in an ice-water mixture. Place the flask in a fume cupboard, and add 38 ml of conc. nitric acid and immediately mix the liquids by shaking for a few seconds. Allow the mixture to stand; generally within 1 minute a vigorous but harmless reaction takes place and copious red fumes are evolved. When the reaction subsides, heat the flask in a boiling water bath for 1.5-2 hours with occasional shaking , the heavy oil, initially present , will ultimately form a mass of crystals. Add 100ml of cold water, chill thoroughly in ice-water, filter the crystals at the pump, wash well with water to remove all the nitric acid and drain. Recrystallize from dilute alcohol.

, Yield:-

BENZILIC ACID

In a 500-ml round bottomed flask, place a solution of 35 g of potassium hydroxide in 70 ml of water, then add 90 ml of rectified spirit and 35g (0.167 mol) of recrystallised Benzene. A deep bluish-black solution is produced. Fit a reflux condenser to the flask and heat the mixture on a boiling water bath for 10-15 mins. Pout the content of the flask into a porcelain dish allow to cool, preferably overnight. The potassium salt of the benzilic acid crystallizes out. Filter off the crystals at the pump and wash with a little ice-cold alcohol. Dissolve the potassium saly in about 350 ml of water, and add 1 ml of concentrated hydrochloric acid from a burrete slowly and with stirring. The precipitate thus produced is coloured red-brown and is somewhat sticky. Filter this off; the filtrate should be nearly colourless. Continue the stirring of hydrochloric acid with stirring until the solution is acid to Congo red paper. Filter off the benzilic acid with suction, wash it thoroughly with cold water until free from chlorides and allow it to dry. The yied of the crude benzilic acid, which is usuallu light pink or yellow in colour, is 30 g (79%). Purify the product either by recrystallisation from hot benzene (about 6 ml per gram) or from hot water with the use of a little decolourising carbon. The coloured and the sticky material obtained by the first precipitation may be recrystallised from hot water with the addition of a little decolourising carbon, and a further 1-2 g obtained. Pure benzilic acid has a m.p 150° C.

METHYL ORANGE

In a 250-ml conical flask place 10.5 g of sulphanilic acid dehydrate, 2.65 g (0.025 mol) of anhydrous sodium carbonate and 100 ml of water, and warm until clear solution is obtained. Cool the solution under the tap to about 15° C, and add a solution of 3.7 g (0.059 mol) of sodium nitrate in 10 ml of water. Pour the resulting solution slowly and with stirring into a 600-ml beaker containing 10.5 ml of concentrated hydrochloric acid and 60 g of crushed ice (1). Test for the presence of the free nitrous acid with potassium iodide starch paper after 15 mins. Fine crystals of diazobenzene sulphonate will soon separate; do not filter these off as they will dissolve during the next stage of the preparation. Disssolve 6.05 g (6.3 ml, 0.05 mol) of dimethylaniline in 3.0 ml of glacial acetic acid, and add it with vigorous stiring to the suspension of diazotized sulphanilic acid. Allow the mixture to stand for 10 mins; the red or acid form of methyl orange will gradually separate. Then add slowly and with stirring 35 ml of 20 per cent sodium hydroxide solution: the mixture will assume a uniform orange colour due to the separation of the sodium salt of methyl orange in fine particles. Direct filtration of the later is slow, hence, whilst stirring the mixture with a thermometer, heat it almost to the boiling point. Most of the methyl orange will dissolve. Add about 10 g of sodium chloride (to assist the subsequent separation of the methyl orange) and warm at 80-90° C until the salt has dissolved. Allow the mixture to cool undisturbed for 15 mins and cool in ice-water; this gives a fairly easily filterable product. Filter off the methyl orange at the pump, but apply only

gentle suction so as to avoid clogging the pores of the filter paper; rinse the beaker with a little saturated salt solution and drain well. Recrystallise from hot water (about 150 ml are required); filter the hot solution, if necessary, through a hot water funnel or through a preheated Buchner funnel. Reddish –orange crystals of methyl orange separate as the solution cools. Filter these at the pump, drain well, wash with a little ethanol, and finally with a small volume of ether.

M. pt. :-

Yield :-

p-Bromoacetanilide

Dissolve 13.5g of finely powered acetanilide in 45ml of glacial acetic acid in a 250ml conical flask . In another small flask dissolve 17g(5.3 ml) of bromine in 25 ml of glacial acetic acid, and transfer the solution to a burette or a separatory funnel supported over the flask. Add the bromine solution slowly and with constant shaking to ensure thorough mixing: stand the flask in cold water. When all the bromine has been added, the solution will have an orange colour due to the slight excess of bromine; a part of the reaction product may crystallize out. Allow the final reaction mixture to stand at room temperature for 30 minutes with occasional shaking. Pour the reaction product into 400ml of water; rinse the flask with 100ml of water. Stir the mixture well and if it appreciably coloured, add just sufficient sodium metabisulphite solution to remove the orange colour. Filter it, wash it with cold water. Recrystallise from dilute methanol.

M.pt:- , Yield:-

I.Separation of mixtures (Marks:10)

- (a) Separation of binary mixtures based on acid-base concept
- (b) Determination of melting points

Method:-

A small portion of the organic mixture(AB) is taken into two clean test tubes where the solubility of the sample is checked by adding dil.HCl and dil.NaOH solution respectively. Now, since one part of the salt is soluble in dil NaOH(or dil. HCl), the whole sample is taken in a beaker and 2(N) NaOH (or 2(N) HCl) is added slowly with continuous stirring till the soluble part is completely soluble and the insoluble part is left behind as residue. Then it is filtered, and the residue(B) is washed for 2-3 times with 2(N) NaOH (or 2(N) HCl) and water. The residue is dried and the melting point of it is determined. To the filtrate (A), which is collected in a beaker, 2(N) HCl (or 2(N) NaOH) is added slowly as it is acidic (basic), till the precipitate is obtained. It is then filtered and washed 2-3 times with water. It is then dried and melting point is determined.

Colour of component A:-

Colour of component B:-

Nature of component A:-

Nature of component B:-

Melting point of component A:-

Melting point of component B:-

<u>Aim of the experiment-Detection of the characteristics element and the functional</u> groups present in the given organic compound and preparation of its derivative

Preliminary Investigation

Colour :

State :

Odour:

Solubility:

- 1. In water
- 2. In dil. Hcl
- 3. In dil. NaOH
- 4. In alcohol

Experiment	Observation	Inference

Test for aromaticity. A small	Sooty flame	Aromatic
amount of the compound is burnt over the luminous flame on wire gauge.	Non-sooty flame	Aliphatic
Action toward litmus. A small amount of the compound is dissolve in neutral solvent (water or alcohol) and tested with litmus paper	Blue to red Red to blue	Maybe acidic Maybe basic
Test for unsaturation . To a dil .KMnO ₄ solution ,a small amount of the solution of the sample is added.		
	Solution is decolorized	Maybe unsaturated

Detection of Element.

Preparation of Lassaigne's Filtrate: a small piece clean metallic sodium is taken in a clean dry ignition tube and heated over a flame to melt it. Then a small amount of the organic substance under test is added over the molten sodium .The tube is again heated first slowly and then strongly till the red heat. The tube is immersed into 20ml of water taken in porcelain basin. The tube is broken completely by tapping. It is boiled for about 10 minutes and then filtered. The filtrate is called the LASSAIGNE'S filtrate or the sodium extract ;with this sodium extract the following test are performed.

Experiment	Observation		Inference
Test for Halogen . To a little portion of the Lassaigne's filtrate dil.HNO ₃ is added and AgNO ₃ solution is added.	i. ii.	White curdy ppt soluble in NH ₄ OH and insoluble in HNO ₃ . False yellow ppt soluble in NH ₄ OH and	Chlorine present
	iii.	insoluble in HNO ₃ . Yellow ppt insoluble in NH ₄ OH and HNO _{3.}	Bromine present

	Deep blue colouration	Iodine present
Test for Nitrogen . To a portion of the extract a few drops of freshly prepared $FeSO_4$ solution is added and boiled. It is then cooled. Then dilute H_2SO_4 is added. Test for Sulphur. To a portion of the extract,freshly prepared sodium nitropruside solution is added.	Violet colouration	Nitrogen is present
		Sulphur present

Detection of Functional Groups

Experiment	Observation	Inference
Test for Carboxyl	Yellow or orange ppt	-CHO or C=O group present
group:		
a) To a small amount of alcoholic solution of the sample about 3ml of2,4- dinitro phenol hydrazine is added and shaken		
Silver mirror test for aldehyde: 1cc of 10%AgNO ₃ solution is		

tak	en in a test tube .To this		
sol	ution NaOH solution is		
ado	led drop wise. A ppt is		
for	med which dissolved by		
ado	ling NH_4OH . This is		
tol	en's Reagent and warmed		
in a	a water bath.		
			-CHO group present
			-ene group present
		Formation of silver mirror	
Te	st for phenolic-OH group:		
a)	To a small amount of the		
	alcoholic solution a few		
	drops of neutral ferric		
	chloride solution is added		
	and the mixture is shaken		
	well.		
b)	Dve test: -13ml of the of		
	the aniline is taken in the		
	test tube and it is		
	dissolved in excess of		
	dilute Hcl or few drops of		
	an NaNO ₂ solution is		
	added and the test tube is		Phenolic –OH group present
	cooled In this alkaline of	X7' 1 / 1	
	the sample is added i e	violet, blue or green	
	dissolve in NaOH	colouration	
Te	st for carboxylic Acid		
Gr	oup:-		
a)	Solid sodium bicarbonate		
	is sprinkled on the neutral		
	solution (water/alcohol) of		
	the sample.		
b)	`1 drop of phenolpthalain		
	indicator is added to a		
	very dilute solution		
	ofNaOH. To this solution		
	alcoholic solution of th		
	sample is added.		

For Amino Group:-		Phenolic –OH group present
Azo-Dye test:- a small amount of the substance is dissolve in dilute Hcl.To it few drops of any solution of sodium nitrite(NaNO ₂) and a cold solution of alkaline β - napthol is added.	Red dye is obtained	
Test for Nitro Group:		
A little amount of the sample is dissolve in 3cc of 50% alcohol. Then a little solid NH4cl and a pinch of Zn dust is added and heated. It is allowed to stand for 5mins aand then filtered into ammoniacal AgNO ₃ (AgNO ₃ +NaOH+NH ₄ OH) in a test tube.		
Test for Amide:		
About 5gm of the sample is taken and to it 3ml of 10% NaOH solution is added and boiled for a few minutes.		
		-COOH group present
	Effervescence	

	-COOH group present
Pink colour disapear	
1	
	Primary amine present
Red brown ppt formed	
	NO group present
	-NO ₂ group present
Grey or black ppt obtained	

	Amide is present
NH ₃ gas evolved	

Preparation of AMIDE Derivative

Above 5gm of the sample is taken in dry crucible, then a few cc of the thionyl chloride is added and stirred well and allowed to stand for sometime then half test tube of conc. NH3 is added very cautiously. The dried sample is then taken in a beaker and alcohol is added and the solution is then kept in water bath till the volume is reduced and then it is allowed to cool. The crystals then forms and dried and melting point is taken.

Preparation of BENZOYL Derivative

About 1rg of the sample is suspended in 20cc of 50% NaOH solution in a conical flask. Then 3cc of Benzoyl chloride is measured and is added little at a time to the solution in a conical flask . After each addition the conical flask is stopped and shaken thoroughly. The mixture should be kept cool throughout the addition . This process is repeated till the whole 3cc of Benzoyl chloride is added. The product is then filtered wash thoroughly with water and recrystallise from alcohol.

Preparation of HYDROLYSIS Derivative

The organic compound (0.5) is refluxed with an excess of 25% sodium hydroxide solution and heated until ammonia has been driven off. The resulting solution is acidified with conc. Hcl acid and liberated acid separated by filtration, distillation or estuation.

Preparation of PICRATE Derivative

A saturated solution of the sample in alcohol is prepared. Then a saturated solution of picrate acid in alcohol is prepare in another beaker. Then the solution of picrate acid is added to the saturated solution of the sample. The mixture is stirred well and is kept under the water bath till the volume is decreased. The solid obtain is filtered dried and melting point is taken.

II Permanent Core Faculty of the Department of Physics benefitted by the scheme

Name & Qualifications	Faculty Training Programs	Name and date of the workshop	Organizing department/host	Remark
Dr. (Mrs) E.M.L. Buam, M. Sc., M. Phil, Ph. D	nil			The dept has paid the Registration fee
Mrs. R. Das, M. Sc	nil			for the participating teachers to attend
Dr. (Mrs) B. Dey Chowdhury, M.Sc., Ph. D, NET	2	 National Seminar on Particle Physics from 21st to 23rd March 2013 National seminar on the Commemoration of the centenary year of Bohr's Atomic theory on 23rd & 24th August 2013 	Department of Physics, St. Anthony's College Department of Physics, Lady Keane College	these National Seminars and Workshop from the Recurring Fund of the Scheme.
Mr. C.G. Synrem, M.Sc., NET	3	 National Seminar on Particle Physics from 21st to 23rd March 2013 National seminar on the Commemoration of the centenary year of Bohr's Atomic theory on 23rd & 24th August 2013 National workshop on Maintenance of Opto- Analytical Instruments- 2013 (NWMOAL13) 	Department of Physics, St. Anthony's College Department of Physics, Lady Keane College Department of Chemistry, St. Anthony's College	
Ms. D. A. War, M.Sc., NET	2	from the 16 th to 20 th September 2013 1. National Seminar on Particle Physics from 21 st to 22 rd March 2012	Department of Physics, St.	
		2. National workshop on Maintenance of Opto- Analytical Instruments- 2013 (NWMOAI-13) from the 16 th to 20 th September 2013	Department of Chemistry, St. Anthony's College	
Mr. Terrence O. Thangkhiew, M.Sc.	1	1. National workshop on Maintenance of Opto- Analytical Instruments- 2013 (NWMOAI-13) from the 16 th to 20 th September 2013	Department of Chemistry, St. Anthony's College	

7. Training/Exposure Visits/Outreach

Activities	Purpose	Class	Date	Organizing Dept/ Institute
I. Industrial Visits/exposure visits:				
1. Educational Trip to North Eastern Space Application Centre, Barapani	Exposure . programme	BSc I, II, III	5/12/2012	Physics Dept, St. Mary's College
2. Educational trip to Central Seismological Observatory, India Meteorological Department, observatory, Magnetic Observatory & Indian Air Force Museum	Exposure and Awareness Programme	BSc I, II, III	24 - 07 - 2013	Physics Dept, St. Mary's College
II. Guest Lecture:	To initiate in depth			
Head of the Dept of Physics, St. Anthony's College.	understanding of basic concepts of Ouantum Mechanics	BSc I, II, III	20 - 07 - 2013	Physics Dept, St. Mary's College
Talk Title: Uncertainty principle	through interactive lectures			
III. Seminars/Training courses for the students:				
1. Annual Departmental Seminar	To train students to present paper	BSc I,II,III	1/12/2012	Physics Dept, St. Mary's College
2. Workshop on ' Use of Simple Medical Physics Instruments'	To give first hand learning of applied Medical Physics Instruments for emergency purpose and to understand the Physics behind their use	BSc I, II, III	15 - 07 - 2013	Physics Dept, St. Mary's College
3. National Seminar on the Commemoration of the Centenary Year of Bohr's Atomic theory	For Awareness and Exposure	BSc III	23,24 August 2013	Physics Dept, Lady Keane College

Activities	Purpose	Class	Date	Organizing Dept/ Institute
4. Hands on training in Electronics	To create interest in the field of			
	Electronics which, if the students pursue for higher studies, would generate a good source of income to them.	XII, BSc I,II,III	28-Sep-13	Physics Dept, St. Mary's College

DEPARTMENT OF PHYSICS:

I PROCUREMENT OF NEW EQUIPMENT FROM NON-RECURRING FUND:

I NON-RECURRING FUND:

Sl. No.	Equipment Name (Model)	Quantity	Rate (Rupees)	Total Cost (Rupees)	Date of order placed	Date of Purchase/ install
1	Kater's Reversible pendulum (BESTO)	1	2,250	2,250	21.12.2012	13.07.2013
2	Compound Bar Pendulum (BESTO)	1	1,040	1,040	21.12.2012	13.07.2013
3	Jaeger's surface tension apparatus (BESTO)	1	2,100	2,100	21.12.2012	13.07.2013
4	Carey-Foster's Bridge (OMEGA)	1	1,685	1,685	21.12.2012	13.07.2013
5	Lee's & Charlton's Conductivity apparatus(BESTO)	1	4,590	4,590	21.12.2012	13.07.2013
6	Determination of J by Callendar & Barne's method (OMEGA)	1	7,303	7,303	21.12.2012	13.07.2013
7	Dispersive power of a plane transmission diffraction grating (OMEGA)	1	15,730	15,730	21.12.2012	13.07.2013
8	Melde's experiment by using electrically maintained tuning fork (OMEGA)	1	6,966	6,966	21.12.2012	13.07.2013

List of Equipments (under Non- Recurring Head)

Sl. No.	Equipment Name (Model)	Quantity	y Rate (Rupees) Total Cost (Rupees)		Date of order placed	Date of Purchase/ install
9	De-Sauty bridge (BESTO)	1	12,950	12,950	21.12.2012	13.07.2013
10	Series & Parallel Resonance LCR kit (BESTO)	2	3,400	6,800	21.12.2012	13.07.2013
11	LDR Characteristics (BESTO)	1	3,640	3,640	21.12.2012	13.07.2013
12	Integrating, Differentiating & Clamping circuits (OMEGA)	1	3,708	3,708	21.12.2012	13.07.2013
13	Determination of energy gap in semiconductor diode (OMEGA)	1	5,169	5,169	21.12.2012	13.07.2013
14	Two stage RC Coupled transistor amplifier (OMEGA)	1	3,034	3,034	21.12.2012	13.07.2013
15	Transistor characteristics of CE, CB, CC & transfer characteristics (BESTO)	1	5,580	5,580	21.12.2012	13.07.2013
16	Torsion Pendulum(HYTEK)	1	3,150	3,150	21.12.2012	23.05.2013
17	Capillary Tube apparatus(HYTEK)	1	2,500	2,500	21.12.2012	23.05.2013
18	Dip Circle(HYTEK)	1	1,400	1,400	21.12.2012	23.05.2013
19	Deflection & Vibration Magnetometers for determination of BH and M(OMEGA)ES-305	1	4,500	4,500	21.12.2012	23.05.2013
20	P. O. Box(HYTEK)	1	2,100	2,100	21.12.2012	23.05.2013
21	Mirror Galvanometer(HYTEK)	1	6,250	6,250	21.12.2012	23.05.2013
22	Tangent Galvanometer, TG-192 (OMEGA)	2	2,200	4,400	21.12.2012	23.05.2013
23	Pullinger's linear expansion apparatus with steel pipe(HYTEK)	1	3,050	3,050	21.12.2012	23.05.2013
24	Fresnel's Biprism apparatus ES- 214 (OMEGA)	1	16,100	16,100	21.12.2012	23.05.2013
25	Young's modulus of glass by Cornu's method ES-262	1	12,075	12,075	21.12.2012	23.05.2013

Sl. No.	Equipment Name (Model)	Quantity	Rate (Rupees)	Total Cost (Rupees)	Date of order placed	Date of Purchase/ install
	(OMEGA)					
26	Determination of Planck's constant by solar cell, ES- 214(OMEGA)	1	14,950	14,950	21.12.2012	23.05.2013
27	Zener diode characteristics apparatus, ETB-245 (OMEGA)	1	6,325	6,325	21.12.2012	23.05.2013
28	Decade Audio Frequency apparatus, AO-301 (OMEGA)	1	6,350	6,350	21.12.2012	23.05.2013
29	AC Milli-voltmeter, ACV-25 (OMEGA)	1	5,980	5,980	21.12.2012	23.05.2013
30	Semi-conductor diode characteristics, ETB-68 (OMEGA)	1	5,980	5,980	21.12.2012	23.05.2013
31	Bread board, BBS-103, 320 tie points (OMEGA)	3	1,400	4,200	21.12.2012	23.05.2013
32	Mercurial B. P. Apparatus Deluxe(Diamond)	1	2,370	2,370	21.12.2012	23.05.2013
33	Stethoscope, Dual Deluxe(Diamond)	1	700	700	21.12.2012	23.05.2013
34	Digital Precision balance, Model: PGB-300 (WENSAR)	1	8,500	8,500	21.12.2012	23.05.2013
35	Audio Frequency Generator 20 Hz to 200KHz O/P 0-20V RMS (OMEGA)	1	6, 400	6, 400	24.09.2013	27.09.2013
36	Battery Eliminator 3 amp (BESTO)	1	1, 948	1, 948	24.09.2013	27.09.2013
37	Equilateral Prisms	5	60	300	24.09.2013	27.09.2013
38	Sodium Vapour Lamp, 35 Watts (BESTO)	1	3, 125	3, 125	24.09.2013	26.09.2013
39	Sodium Vapour Lamp Transformer, 35 Watts (BESTO)	1	2, 252	2, 252	24.09.2013	26.09.2013
40	Wooden Box for Sodium Vapour Lamp, 35 Watts	1	969	969	24.09.2013	26.09.2013

Sl. No.	Equipment Name (Model)	Quantity Rate (Rupees)		Total Cost (Rupees)	Date of order placed	Date of Purchase/ install
	(BESTO)					
41	Battery Eliminators, BED-71, 3 amp (BESTO)	3	1, 948	5,844	24.09.2013	26.09.2013
42	Battery Eliminators, BED-71, 5 amp (BESTO)	2	3, 135	6, 270	24.09.2013	26.09.2013
43	Tangent Galvanometer (Precision Type) (BESTO)	3	1, 701	5, 103	24.09.2013	26.09.2013
44	Copper Voltameter with Jar (BESTO)	2	618	1, 236	24.09.2013	26.09.2013
45	P.O. Box 3-ratio (Manganin) (BESTO)	1	5,577	5,577	24.09.2013	26.09.2013
46	P.O. Box 4-ratio (Manganin) (BESTO)	1	5, 938	5, 938	24.09.2013	26.09.2013
47	Lead with stackable Banana Plug, Length 50 cm (BESTO)	15	60	900	24.09.2013	26.09.2013
48	Lead with stackable Banana Plug, Length 100 cm (BESTO)	15	60	900	24.09.2013	26.09.2013
49	Lead with Banana Plug and Crocodile clip set of two(BESTO)	24	57	1, 368	24.09.2013	26.09.2013
50	Lead with Banana Plug length 50 cms (BESTO)	18	48	864	24.09.2013	26.09.2013
51	Lead with Banana Plug length 100 cms (BESTO)	18	64	1, 152	24.09.2013	26.09.2013
		TOTAL		2,43,571		
		VAT 13.5	5%	32882.085		
		TOTAL 1		2,76,453		
	HP P-2 1403 Desktop with monitor	3	30, 005.71	90017.13	07.02.2013	21.03.2013
	HP G6-2204 TX Laptop with	1	35,	35238.1	07.02.2013	21.03.2013

Sl. No.	Equipment Name (Model)	Quantity	Rate (Rupees)	Total Cost (Rupees)	Date of order placed	Date of Purchase/ install
	carry case		238.1			
	HP Laserjet M1536 DNF	1	23, 380.95	23380.95	07.02.2013	21.03.2013
	UPS 600 VA	3	2, 190.48	6571.44	07.02.2013	21.03.2013
		TOTAL		155207.62		
		CST @ 59	%	7760.38		
		TOTAL 2		162968.00		
	Canon Projector LV-7295 (E)	1	50, 995	50,995	22.07.2013	23.07.2013
	Projector Screen 6" x 8"	1	9,000	9,000	22.07.2013	23.07.2013
	Laser Pointer	1	1, 750	1,750	24.09.2013	24.09.2013
		TOTAL 3	3	61745		
	GRAND TOTAL 1 + TOTAL 2 + TOTAL 3			501166		

II Recurring Fund

1. Glass wares: Order Date25.9.2013 supply date: 27-09-2013

SL No.	Type of Glass ware	Company	Quantity	Rate	Total cost			
51.110.	Type of Glass wate	company	Quantity	(in Rs.)	(in Rs.)			
1	Polarimeter tubes	BESTO	2	600	1200			
2	Diffraction grating (38 x 50 mm) approx 15000 lines/inch	BESTO	2	990	1980			
3	Thermometer (0-110°C)		12	150	1800			
4	Beaker (500ml)	Borosil	12	101	1212			
5	Lens (focal length-10cm)		12	40	480			
6	Mirror (focal length-10cm)		12	40	480			
7	Prism (equilateral)		6	60	360			
8	Measuring Cylinder (100ml)	Borosil	6	350	2100			
	Amount				9612			
VAT @ 13.5%								
	Total Amount (Total 1)							

II. PROCUREMENT OF BOOKS AND JOURNALS FROM RECURRING FUND

CI		Books received from the	Dublication		A 4	Data of
SI. No	Name of the book	Author	house	1	Amount (in Rs.)	Date of Purchase
110.			nouse		(111 185.)	I ur chase
1	3000 solved Problems in Physics, Schaum's Outlines	Alvin, Halpern	Tata Mcgraw	1	599	25.07.2013
2	A textbook of Optics	Brijlal, Subrahmanyam, M. N. Avadhanulu	S. Chand	1	375	25.07.2013
3	A textbook of Practical Physics	Samir Kumar Ghosh	NCBA	1	235	25.07.2013
4	A treatise of General Properties of matter	H. Chatterjee & R. Sengupta	NCBA	1	650	25.07.2013
5	Advanced Practical Physics	Chattopadyay,Rakshit	NCBA	1	350	25.07.2013
6	Advanced Practical Physics	Samir Kumar Ghosh	NCBA	1	295	25.07.2013
7	Advanced Practical Physics Vol.1	S. P. Singh	Pragati Prakashan	1	200	25.07.2013
8	Advanced Practical Physics Vol.2	S. P. Singh	Pragati Prakashan	1	120	25.07.2013
9	Advanced Practical Physics Vol-1	B. Ghosh, Mazumdar	Sreedhar Pubn	1	260	25.07.2013
10	Advanced Practical Physics Vol-2	B. Ghosh	Sreedhar Pubn	1	200	25.07.2013
11	An introduction to mechanics	Kleppner & Kolenkow	Tata Mcgraw	1	399	25.07.2013
12	Atomic and Nuclear Physics	Brijlal, Subrahmanyam	S. Chand	1	165	25.07.2013
13	Atomic Physics	S. N. Ghoshal	S. Chand	1	250	25.07.2013
14	B. Sc Practical Physics	C. L. Arora	S. Chand	1	310	25.07.2013
15	B. Sc Practical Physics	Harnam Singh,Hemne	S. Chand	1	425	25.07.2013
16	Basic Electronics	D.C. Tayal,V. Tayal	Himalaya	1	410	25.07.2013

Books received from the first order

Sl. No.	Name of the book	Author	Publication house	1	Amount (in Rs.)	Date of Purchase
			Pubn.			
17	Basic Electronics: Solid state	B. L. Theraja	S. Chand	1	440	25.07.2013
18	Basic Nuclear Physics	B. N, Srivastava	Pragati Prakashan	1	235	25.07.2013
19	Classical mechanics	G. Aruldhas	PHI	1	275	25.07.2013
20	Classical mechanics	Goldstein, Poole, Safko	Pearson	1	550	25.07.2013
21	Classical mechanics	Herbert Goldstein	Narosa	1	310	25.07.2013
22	Classical mechanics	J. C. Upadhyaya	Himalaya Publication	1	350	25.07.2013
23	Classical mechanics	Satya Prakash	Kedarnath RamNath	1	260	25.07.2013
24	Computer Programming in Fortran 77	V. Rajaraman	PHI	1	195	25.07.2013
25	Concepts of Modern Physics 6th Edn	Arthur Beiser, Mahajan, S. Rai Choudhury	Tata Mcgraw	1	410	25.07.2013
26	Concepts of Nuclear Physics	B. L. Cohen	Tata Mcgraw	1	235	25.07.2013
27	Concepts of Physics Vol-1	H. C. Verma	Bharati Bhawan	1	268	25.07.2013
28	Concepts of Physics Vol-2	H. C. Verma	Bharati Bhawan	1	268	25.07.2013
29	Electricity and electronics	D. C. Tayal	Himalaya Pubn.	1	390	25.07.2013
30	Electromagnetics	B. B. Laud	New Age	1	200	25.07.2013
31	Electronic spectra of transition metal complexes	Ray	NCBA	1	835	25.07.2013
32	Elements of Properties of Matter	D. S. Mathur	S. Chand	1	295	25.07.2013
33	Elements of Quantum	Kamal Singh,S.P. Singh	S. Chand	1	110	25.07.2013

Sl. No.	Name of the book	Author	Publication house	1	Amount (in Rs.)	Date of Purchase
	mechanics					
34	Elements of solid state physics	J. P. Srivastava	PHI	1	350	25.07.2013
35	Elements of Spectroscopy	Gupta,Kumar,Sharma	Pragati Prakashan	1	295	25.07.2013
36	Foundations of Electromagnetic theory	J. R. Reitz, F. J. Milford & R. W. Christy	Narosa	1	280	25.07.2013
37	Handbook of Electronics	Gupta, Kumar	Pragati Prakashan	1	625	25.07.2013
38	Heat & Thermodynamics 7th Edn.	Zemansky, Dittman	Tata Mcgraw	1	395	25.07.2013
39	Heat, Thermodynamics and Statistical physics	Brijlal, Subrahmanyam, Hemne	S. Chand	1	450	25.07.2013
40	Introduction to Electrodynamics	D. J. Griffiths	PHI	1	275	25.07.2013
41	Introduction to Mathematical Physics	Charlie Harper	PHI	1	157	25.07.2013
42	Introduction to Solid State Physics	Charles Kittel	Wiley	1	449	25.07.2013
43	Laser & Non linear optics	B. B. Laud	New Age	1	165	25.07.2013
44	Mathematical Physics	Binoy Bhattacharya	NCBA	1	735	25.07.2013
45	Mathematical Physics	H. K. Dass	S. Chand	1	495	25.07.2013
46	Mechanics	D. S. Mathur revised edn.	S. Chand	1	395	25.07.2013
47	Modern Atomic & Nuclear Physics	A.B. Gupta & Dipak Ghosh	Books & Allied	1	525	25.07.2013
48	Modern Physics	R. Murughesan	S. Chand	1	525	25.07.2013
49	Molecular Structure	Aruldhas	PHI	1	325	25.07.2013

Sl. No.	Name of the book	Author	Publication house	1	Amount (in Rs.)	Date of Purchase
	& Spectroscopy					
50	Nuclear Physics	Irving Kaplan	Narosa	1	230	25.07.2013
51	Nuclear Physics	S. N. Ghoshal	S. Chand	1	425	25.07.2013
52	Physics 5th Edn. Vol- 1	Resnick,Halliday,Krane	Wiley	1	480	25.07.2013
53	Physics 5th Edn. Vol- 2	Resnick,Halliday,Krane	Wiley	1	529	25.07.2013
54	Physics for Degree Students BSc 1st year	C. L. Arora	S. Chand	1	370	25.07.2013
55	Physics for Degree Students BSc 1st year	C. L. Arora, P.S. Hemne	S. Chand	1	375	25.07.2013
56	Principles of Electronics 7th edn.	V.K. Mehta, R. Mehta	S. Chand	1	395	25.07.2013
57	Quantum mechanics	G. Aruldhas	PHI	1	295	25.07.2013
58	Quantum mechanics, statistical mechanics and Solid state physics	D. Chattopadhyay, P. C. Rakshit	S. Chand	1	195	25.07.2013
59	Quantum Physics	Stephen Gosioroveicz	Wiley	1	419	25.07.2013
60	Schaum's Outline series: Vector Analysis and introduction to Tensor analysis	Murray R Spiegel	Tata Mcgraw	1	415	25.07.2013
61	Solid State Physics	A. J. Dekker	Macmillan	1	325	25.07.2013
62	Solid State Physics 5th Edn	S. O. Pillai	New Age	1	399	25.07.2013
63	Statistical Mechanics	Gupta, Kumar, Sharma	Pragati Prakashan	1	250	25.07.2013
64	The Feynman Lectures on Physics Vol-2	Feynman, Leighton, Sands	Narosa	1	285	25.07.2013

Sl. No.	Name of the book	Author	Publication house	1	Amount (in Rs.)	Date of Purchase
65	The Feynman Lectures on Physics Vol-2	Feynman, Leighton, Sands	Pearson	2	375	25.07.2013
66	The Feynman Lectures on Physics Vol-3	Feynman, Leighton, Sands	Pearson	1	375	25.07.2013
67	The Physics of Vibrations & Waves	H. J. Pain	Wiley	1	449	25.07.2013
68	Waves and Oscillations	Brijlal, Subrahmanyam	Vikash	1	300	25.07.2013
Amount (in Rs.)						
		s.)	2017			
		s.)	22179			

Books received from the second order

Sl.	Name of the book	Author	Publication	1	Amount	Date of
110.			nouse		(111 K5.)	I ul chase
1	A textbook of Optics	Brijlal, Subrahmanyam, M. N. Avadhanulu	S. Chand	3	425	28.09.2013
2	A textbook of Practical Physics	Samir Kumar Ghosh	NCBA	1	705	28.09.2013
3	A treatise of General Properties of matter	H. Chatterjee & R. Sengupta	NCBA	3	595	28.09.2013
4	Advanced Practical Physics	Samir Kumar Ghosh	NCBA	3	1050	28.09.2013
5	Advanced Practical Physics Vol.1	S. P. Singh	Pragati Prakashan	3	420	28.09.2013
6	Advanced Practical Physics Vol.2	S. P. Singh	Pragati Prakashan	1	360	28.09.2013
7	Advanced Practical Physics Vol-1	B. Ghosh, Mazumdar	Sreedhar Publication	1	220	28.09.2013
8	Advanced Practical	B. Ghosh	Sreedhar	3	260	28.09.2013

Sl. No.	Name of the book	Author	Publication house	1	Amount (in Rs.)	Date of Purchase
	Physics Vol-2		Publication			
9	B. Sc Practical Physics	C. L. Arora	S. Chand	2	930	28.09.2013
10	B. Sc Practical Physics	Harnam Singh,Hemne	S. Chand	1	850	28.09.2013
11	Classical mechanics	Gupta,Kumar,Sharma	Pragati Prakashan	1	270	28.09.2013
12	Classical mechanics	J. C. Upadhyaya	Himalaya Pubn.	1	350	28.09.2013
13	Electromagnetics	B. B. Laud	New Age	1	200	28.09.2013
14	Elements of Spectroscopy	Gupta, kumar & Sharma	Pragati Prakashan	1	310	28.09.2013
15	Feynman Lectures on Physics Vol-1	Feynman, Leighton, Sands	Pearson	2	475	28.09.2013
16	Fortran 77 & Numerical Analysis	C. Xavier	New Age	1	595	28.09.2013
17	Fundamentals of Statistical Mechanics	B. B. Laud	New Age	1	175	28.09.2013
18	Handbook of Electronics	Gupta, Kumar	Pragati Prakashan	1	625	28.09.2013
19	Heat, Thermodynamics and Statistical physics	Brijlal, Subrahmanyam, Hemne	S. Chand	1	450	28.09.2013
20	Mathematical Physics	B. D. Gupta	Vikash	1	625	28.09.2013
21	Mathematical Physics	B. S. Rajput	Pragati Prakashan	1	525	28.09.2013
22	Mathematical Physics	H. K. Dass	S. Chand	1	595	28.09.2013
23	Modern Physics: Concepts & Applications	S. Puri	Narosa	1	290	28.09.2013
24	Molecular Spectroscopy	S. Chandra	Narosa	1	235	28.09.2013

Sl. No.	Name of the book	Author	Publication house	1	Amount (in Rs.)	Date of Purchase
25	Nuclear Physics	R.R. Roy & B. P. Nigam	New Age	1	195	28.09.2013
26	Physics for Degree Students BSc 2nd year	C. L. Arora, P.S. Hemne	S. Chand	1	475	28.09.2013
27	Principles of Electronics	V.K. Mehta, R. Mehta	S. Chand	1	350	28.09.2013
28	Quantum Mechanics: 500 problems with solutions	Aruldhas	PHI	1	295	28.09.2013
29	Solid State Electronic Devices	B. G. Streetman	PHI	1	295	28.09.2013
30	Solid State Physics 5th Edn	S. O. Pillai	New Age	1	295	28.09.2013
31	Statistical Mechanics	Gupta, Kumar, Sharma	Pragati Prakashan	1	225	28.09.2013
32	Waves and Oscillations	Brijlal, Subrahmanyam	Vikash	1	365	28.09.2013
33	Waves and Oscillations	R. N. Choudhuri	New Age	1	200	28.09.2013
34	Waves and Oscillations	Waves and OscillationsS. N. SenNew Age1		195	28.09.2013	
			Amount (in R	(s.)		14,425
			Discount (in R	(s.)		1,442
		(s.)		12,983		

List of Encyclopaedia ordered

Sl. No.	Name of the Encyclopaedia	Rate (in Rs.)	Discount (in Rs.)	15,456	Date of Purchase
1	Encyclopaedia of Scientific Principles,Laws & Theory	19,320	3864		28.09.2013

List of Journals ordered

Sl. No.	Name of the Journal	Source	Annual Subscription Rate	24.09.2013
1	Current Science	Indian Academy of Sciences, Bangalore	1500	24.09.2013
2	Resonance-Journal of Science Education	Indian Academy of Sciences, Bangalore	500	24.09.2013
3	Bulletin of Materials Science	Indian Academy of Sciences, Bangalore	400	24.09.2013
4	Journal of Astrophysics and Astronomy	Indian Academy of Sciences, Bangalore	300	24.09.2013
5	Journal of Earth System Science	Indian Academy of Sciences, Bangalore	400	24.09.2013
6	Pramana- Journal of Physics	Indian Academy of Sciences, Bangalore	750	24.09.2013
	·	Amount (in Rs.)	3,850	
	Speed Post fee for sending	Subscription form (in Rs.)	39	
		Total Amount (in Rs.)	3889	

Grand Total Amount spent on Books, Encyclopaedia and Journals (in Rs.) = 54,507/-

(Rupees in words-Fifty Four thousand Five hundred and Seven only)

Lab Manuals generated:

1. Aim of the Experiment-To determine the horizontal component of the earth's magnetic field by plotting a graph between tan θ versus $1/d^3$ and comparing with the result obtain from the value of *MB* and *M/B*.

Apparatus required- Deflection Magnetometer, Vibration Magnetometer, Magnetic Needle, Spirit Level, Bar Magnet, Stop Watch, Slide callipers, Balance with weight box.

Theory:

If a bar magnet (NS) of magnetic moment M is placed near a magnetic needle (ns) so that the magnetic needle is on the end on position of the magnet and the axis of the bar magnet is at right angles to the earth's magnetic field, then the magnetic needle will be deflected from the magnetic meridian by a certain angle, say θ . In equilibrium, the needle will be acted upon by two equal and opposite forces forming a couple. One of the two forces is the horizontal

component of the earth's magnetic induction B acting along the magnetic meridian and the other force is due to the field F of the bar magnet (NS). If d is the distance of the needle from the centre of the magnet as shown in figure 1, then the deflection of the needle from the magnetic meridian is given by

Where l is the half of the magnetic length of the bar magnet = $\frac{1}{2}$ the actual length of the magnet × 0.85 (1a)

and μ_0 is the permeability of free space = $4\pi \times 10^{-7}$ Henry/metre.

If the same bar magnet is made to oscillate with small amplitude at the same place in a vibration magnetometer, with its axis parallel to the earth's magnetic field, then the timeperiod of oscillation of the bar magnet (assuming the suspension fibre to be torsionless and neglecting the moment of inertia of the stirrup) is

$$T = 2\pi \sqrt{\frac{l}{MB}} \qquad \dots \dots \dots (2)$$

where I is the moment of inertia of the bar magnet about the point of suspension. For a bar magnet of rectangular dimensions, the moment of inertia I is given by,

Where l' is the length, b' is the breadth and m is the mass of the bar magnet

From equation (3), we get,

$$MB = \frac{4\pi^2}{T^2}I \qquad \dots \dots \dots (4)$$

For a short bar magnet, $d \gg l$, equation (1) reduces

If a graph of $tan \theta$ versus $\frac{1}{d^3}$ is plotted, a straight line graph is obtained, with a slope of

Combining equations (4) and (6), we get

From equation (7), we get the value of *B* in tesla, since all the quantities are in SI units. The horizontal component of the earth's magnetic field *H* in $\frac{A}{m}$ is given by

$$H = \frac{B}{\mu_0} \qquad \dots \dots \dots (8)$$

To get the value of H from *MB* and $\frac{M}{B}$, we can know by using the following formula,

$$B = \sqrt{MB \div \frac{M}{B}} \qquad \dots \dots \dots \dots (9)$$

Knowing the value of B we can find the value of H using equation (8)



Figure 1

Procedure:

1. The mass (m') of the given magnet is determined by the balance, while its length (l') and breadth (b') are determined by using a slide calliper. The moment of inertia *I* of the magnet is then calculated by using the relation (3), while half of its magnetic length (l) is determined by using the relation (1a).

2. All magnets and magnetic substances are to be removed from the working table and the magnetometer is placed on the table with its two arms perpendicular to the magnetic meridian, i.e. perpendicular to the magnetic needle (*ns*). At this time, the pointer usually reads ($0^{\circ} - 0^{\circ}$) of the circular scale. If the pointer does not coincide with ($0^{\circ} - 0^{\circ}$) line the circular case of the magnetometer is to be slowly rotated to bring the pointer with ($0^{\circ} - 0^{\circ}$) line.

3. The magnet (NS) is then placed on the arm of the magnetometer at the east side of the needle, so that the length of the magnet is parallel to the arm of the magnetometer. The position of the magnet on the arm is adjusted until the pointer reads about 45° on the circular

scale. The reading d_1 and d_2 , corresponding to the two ends of the magnet, are noted from the meter-sale fixed on the arm. The distance d of the needle from the centre of the magnet is then given by the mean of d_1 and d_2 , i.e., $d = (d_1 + d_2)/2$.

4. Keeping this distances d of the needle from the centre of the magnet (NS) constant, the readings of the two ends of the pointer are noted from the circular scale when, (a) the two flat surfaces of the magnet are alternately touching the arm and (b) the N-pole and S-pole of the magnet are alternately pointing towards the needle. For each position of the magnet we are getting two readings corresponding to the two ends of the pointer hence for the four position of the magnet, as are indicated in (a) and (b), we shall get altogether eight readings.

5. The magnet (*NS*) is then transferred to the other arm of the magnetometer at the east side of the magnetic needle so that the distance of the needle from the centre of the magnet is again '*d*'. The operation (4) is the repeated when we get another set of eight readings. The mean of these 16 readings gives θ from which *M*/*B* is calculated by using the relation (1).

6. The magnet is now placed at other distances from the needle until the deflections of the pointer on the circular scale are about 43° and 47° respectively. By taking the two scale readings corresponding to the two ends of the magnet, we get the new distance of the needle from the centre of the magnet. Then the entire operations of (4) and (5) are repeated and M/B is calculated. The mean of these three values of M/B is found out.

7. The magnet is then suspended horizontally in the vibration magnetometer box and the box is rotated until the axial line of the magnet is parallel to the horizontal line marked on the plane mirror fixed to the base of the box. The magnet is then deflected by a very small angle by bringing momentarily an auxiliary magnet very near to it. The time taken by the magnet for its 20 complete oscillations is noted thrice. When the mean time for these three observations is divided by 20, we get the period T. These values of I and T, when put in the equation (4), we get *MB*. Knowing mean *MB* and *MB*, we can calculate *B* and hence *H*.

8. For each value of *d*, there are 16 values of θ . The mean of these deflections gives the true value of θ . A graph is then plotted by taking tan θ along the *y*-axis and $1/d^3$ along the *x*-axis. For small values of *d* (i.e., away from the origin), the graph is not a straight line, but it will be a straight line graph for large enough values of *d* (near the origin) for which the short-magnet approximation holds. Then the value of *d* is located on the graph beyond which the graph is a straight line. The slope η of the straight line is then found out and hence the value of *M/B* can be obtained from it.

Experimental results:

	G	eometric length	(l')	Geometric breadth (<i>b</i> ['])		
No. of	(in cm)			(in cm)		
Obs.	Main scale reading	Vernier scale reading	Vernier scale reading Total reading		Vernier scale reading	Total reading

Table 1- Determination of geometric length (l') and breadth (b') of the magnet:

Table 2	- Determination	of mass and	l moment	of inertia	(I) of the	magnet:
	2	01 111400 4114	*	01 11101 1110	(-) 01 0110	

Mass of the magnet	Mean <i>l</i> ' from table 1	Mean <i>b</i> ['] from table 1	Moment of inertia (in kg- m ²)
(kg)	(in m)	(in m)	$I=\frac{m}{12}\left(l^{\prime 2}+b^{\prime 2}\right)$

Table 3- Determination of time-period (T) of oscillation and *MB*:

No. of Obs.	Time for 20 oscillations (in seconds)	Mean Time for 20 oscillations (in seconds)	Time-period (in seconds) $T = \frac{t}{20}$	Moment of inertia (in kg- m ²) from table 2	$MB = \frac{4\pi^2}{T^2}I$ (in A-m ² -tesla)

	Mean of		Defl	ection o	of the ne	edle in c	legrees,	when th	ie magne	et is,	
No.	the scale reading for the two	Position of a	0	n the Ea magnet	ast-arm (tometer	of	0	n the W magnet	est-arm tometer	of	Mean
Obs.	ends of the magnet $d=(d_1+d_2)$ / 2 in m	of magnet	N-p pointi nee	oole ng the edle	S-p pointi nee	oole ng the edle	N-p pointi nee	oole ng the edle	S-p pointi nee	oole ng the edle	degrees (θ)
			End	End	End	End	End	End	End	End	
			Ι	Π	Ι	II	Ι	II	Ι	II	
1		Up									
		Down									
2		Up									
		Down									
3		Up									
		Down									

Table 4-Determination of θ from deflection magnetometer

Table 5- Values of $\tan \theta$ and $1/d^3$

No. of Obs.	θ (degree)	tan θ	<i>d</i> (m)	$1/d^3 ({\rm m}^{-3})$

Table 6 – Determination of *M/B* from the slope of the straight line portion of tan θ versus $1/d^3$ graph near the origin

$(\mu_0 = 4\pi \text{ x } 10^{-7} \text{ henry / m})$

$\Delta = \left(\frac{1}{d^3}\right)$ (from graph) (m ⁻³)	Δ (tan θ) (from graph)	Slope $\eta = \frac{\Delta(tan\theta)}{\Delta(\frac{1}{d_3})}$ (m ⁻³)	$M/B(=\frac{2\pi\eta}{\mu_0})$ (m ⁴ /henry)

Moment of inertia I (in kg-m ²) from table 2	Slope η (in m ³) from table 6	Time-period T (in seconds) from table 3	Earth's magnetic induction B(in tesla) (from equation 7)	Horizontal component of earth's magnetic field (in A/m) (H=B/ µ ₀)

Table 7- Determination of the horizontal component of the earth's magnetic field:

Table 8- Determination of H from the value of *MB* and *M/B*:

Value of MB	Value of <i>M/B</i>	Value of B	Value of H	Value of H	Difference in the
(from table 3)	(from table 6)	(from equation 8)	(from equation 9)	(from table 7)	value of H from the two procedures

Results:

The horizontal component of the earth's magnetic field=..... A/m.

By comparing the value of H obtained from the two methods with the true value of H it has been found that the first/second procedure gives better result.

Precautions and Discussions:

1. Before starting the experiment, all magnets, magnetic substances and irons should be removed from the working table. The magnetometers must be levelled before the experiment.

2. The error in the measurement of M/B would be the minimum when *d* is large and θ is 45° and the value of *d* should be large in comparison to the length of the magnet.

3. To bring the two arms of the magnetometer perpendicular to the magnetic meridian (*i.e.* to bring the axis of the magnet perpendicular to the magnetic meridian) the position of the arm is to be adjusted until equal deflections of the needle are obtained with N-pole and *S*-pole alternately pointing the needle. In the deflection magnetometer, θ should not be too small less than 5^o to avoid a large relative error in its measurement.

4. The magnetic needle should be made free, so that a small shift of the magnet may change the deflection of the needle. To minimise the effect of friction the magnetometer box should be tapped a little before taking the reading.

5. During the oscillation of the magnet the amplitude of the oscillation should be made small (not exceeding 10^{0}) and the oscillation of the suspension fibre should be avoided.

6. The magnet should be so placed on the cradle that its upper face is horizontal and the vertical axis of oscillation passes through its centre of gravity. The suspension fibre must be made twist-free and should be perfectly flexible.

7. For calculations of moment of inertia, the length and breadth of that face of the magnet should be measured which was horizontal during oscillation.

8. The moment of inertia of the cradle is assumed to be small compared with that of the magnet.

9. The experiment can also be performed by placing the magnet in 'Tangent B' position.

References

1. Chattopadhyay, D. R. An Advanced Course in Practical Physics. New Central Book Agency.

Ghosh, S. K. *A text-book of Practical Physics*. New Book Centre Agency.

2. Aim of the experiment- Determination of the thermal conductivity of a bad conductor by Lees' method:



Apparatus required:

Lees apparatus; two thermometers (1/10°C); specimen of a

bad conductor (glass) in the shape of a circular disc; steam chest; boiler; a stop watch; a screw gauge, etc.

The apparatus consists of a brass disc C supported by means of strings from a ring fixed on a retort stand as shown in the figure. The experimental material (bad conductor) is taken in the form of a circular slab (s) of the same diameter as that of C. It is of uniform thickness and placed on the disc C. a steam chamber A is placed on the slab (S) and the bottom of the steam chamber B is a thick circular metal plate. There are arrangements **Figurentryeandpointedf**ssteam in the chamber A. Diameters of B, C and S are all the same. Two thermometers T_1 and T_2 are inserted through two holes drilled in B and C to record temperatures of B and C respectively.

Theory:

When steam is passed through the inlet of chamber A (as shown in the diagram), the base B of A is warmed up and heat is conducted through the bad conductor (i.e., the specimen) and finally to the brass disc C, which is also warmed up. Soon a steady state is reached when the rate of flow of heat through S is equal to the heat lost from C.

If K is the coefficient of thermal conductivity of the material of the bad conductor, d its thickness and A, the area of cross-section of the specimen, then the quantity of heat (Q) conducted per sec through the bad conducting sheet is given by,

Where θ_1 and θ_2 are the temperatures of B and C in the steady state.

If M and s is the mass and specific heat of the material of the lower disc C and $\left(\frac{d\theta}{dt}\right)$ is the rate of cooling at temperature is θ_2 , then the heat radiated away per sec from C is

$$Q = Ms \left(\frac{d\theta}{dt}\right)_{\theta_2} \qquad \dots \dots (2)$$

From equation (1) and (2) we get,

$$KA \ \frac{(\theta_1 - \theta_2)}{d} = Ms \left(\frac{d\theta}{dt}\right)_{\theta_2}$$

$$K = \frac{M \cdot s \cdot d \cdot \left(\frac{dt}{dt}\right)_{\theta_2}}{A(\theta_1 - \theta_2)} \qquad \dots \dots \dots (3)$$

Measuring the quantities given in the above equation, one can determine the value of K of the bad conductor. The rate of cooling is found by heating the metal disc to a temperature about 10^{0} C above the steady temperature θ_{2} , it is then allowed to cool and temperature is noted after every 30 seconds till the temperature falls to about 10^{0} C below θ_{2} . A graph is then plotted between the temperature and time. A tangent is drawn at a point P corresponding to θ_{2} . The slope of the tangent gives the value of $\frac{d\theta}{dt}$ corresponding to temperature θ_{2} .

Procedure:

1. The mass (M) of the lower disc C is determined with the help of a spring balance. The diameter of the slab (S) is also determined by measuring its circumference and hence the area A. The specific heat (s) of the material of the slab is noted from the table of constants.

2. The thickness (d) of the slab is measured by a screw gauge at different positions and hence the mean of these values will give the thickness of the slab.

3. The two thermometers are then compared for any initial errors. The apparatus is set as shown in the diagram so that the flat surface of the disc is horizontal. The disc of the material is inserted in between the disc C and steam chamber A. The thermometers T_1 and T_2 are introduced in the respective position. Steam is passed from a boiler placed at a distance. Temperatures T_1 and T_2 are recorded at intervals of 5 minutes until steady temperatures are attained.
4. The supply of steam is cut off and the upper disc B, the chamber A and the sheet S are all removed. The lower slab C is then heated directly by a burner until the temperature is raised about ten degrees higher than the steady value recorded by T_2 . Then the burner is removed, and the disc C is allowed to cool down in the same conditions as during the first part of the experiment. The temperatures during cooling as indicated by T_2 are noted at intervals of half a minute.

5. From these data, the rate of cooling $\frac{d\theta}{dt}$ and the average temperature of the disc during the interval can be found out. A graph (as shown in figure 1) can be plotted with the rate of cooling $\frac{d\theta}{dt}$ along Y-axis and the corresponding average temperature θ along X-axis which would be a straight line. From this graph, the rate cooling $\frac{d\theta}{dt}$ at θ_2 , the steady temperature of C during the first part of the compriment, can be

C during the first part of the experiment, can be found out.

6. Another graph may be plotted with the temperature (θ) of the disc C recorded during cooling and the time (t). From this graph, a tangent is drawn at the point on the curve corresponding to θ_2 , the steady temperature attained by the lower disc during the static part of the experiment. From this tangent, the slope gives the value of $\frac{d\theta}{dt}$ at θ_2 .





Figure1: cooling rate-temperature



7. Knowing the values of M, s, $\left(\frac{d\theta}{dt}\right)_{\theta_2}$, A, d and $(\theta_1 - \theta_2)$ and also the value of r, the value of thermal conductivity K of the material of the slab can be obtained from equation (3).

Experimental data:

Mass of the lower disc $C(M) = \dots g$

Specific heat of the material of the disc C (s) = Cals/g 0 C

No. of Obs.	Circumference of the slab L (in cm)	Mean value of the circumference <i>L</i> (in cm)	Radius of the slab $r = \frac{L}{2\pi}$ (in cm)	Area of the slab $A = \pi r^2 (\text{in cm}^2)$
1.				
2.				
3.				

Table1- Determination of radius and hence area of the sheet:

Table 2- Determination of the thickness (d) of the slab by a screw gauge:

Least count of the screw gauge (L.C.) =

Error of the screw gauge (e) =.....

		Circular	scale reading	Total reading (in cm)	
	Linear		1			Mean value of
No.	scale	No. of circular	Value of Circular			the thickness of
of	reading (N)	scale divisions	scale divisions (in	Observed	Corrected	the slab d
Obs.	Obs. (in cm) scale divisions on the reference line (n)	$cm) = n \times L.C.$	$d_0 = N + n \times L.C.$	$d = d_0 + e$	(in cm)	
1.						
2.						
3.						

Table 3- Thermometer readings for steady-state temperatures:

Initial corrections between the thermometers=.....⁰C

Time in minutes	0	5	10	15	20	
Temperature $(\theta_1^0 C)$ of T_1						
Temperature $(\theta_2^0 C)$ of T_2						

Table 4-	Time-Temperature	record during	cooling	of the disc	C :
----------	-------------------------	---------------	---------	-------------	------------

No. of Obs.	Tim e in mins (t)	Temperatur e of the disc C in ⁰ C	Average temperature during the interval in ⁰ C	Rate of cooling of the disc C during the interval $\left(\frac{d\theta}{dt}\right)$	Rate of cooling $\left(\frac{d\theta}{dt}\right)$ at $\theta_2^0 C$ from graph 1(0 C per sec)	Rate of cooling $\left(\frac{d\theta}{dt}\right)$ at $\theta_2^0 C$ from graph 2(0 C per sec)
1.	0					
2.	1⁄2					
3.	1					
4.	11/2					
5.	2					
6.						

Calculations:

From equation (3)
$$K = \frac{M.s.d.\left(\frac{d\theta}{dt}\right)_{\theta_2}}{A(\theta_1 - \theta_2)}$$

Substituting the values of the quantities measured and obtaining the value of $\left(\frac{d\theta}{dt}\right)_{\theta_2}$ from the graph, the value of K can be obtained.

Result:

The value of the thermal conductivity of glass =

Precautions:

- 1. steady-state temperatures shown by the thermometers T_1 and T_2 should be recorded at least for ten minutes.
- 2. During cooling of the lower disc, readings of temperatures should be taken at an interval of half-a-minute or even more rapidly.
- 3. The lower disc should be heated by a burner to a temperature at least 10^{0} C above the steady temperature (θ_{2}^{0} C) shown during its static state.
- 4. Cooling of the lower disc must be allowed with the slab (S) placed on it.

- 5. The diameter of the experimental slab(S) must be large compared to the thickness (d) to minimize heat loss due to radiation.
- 6. The diameter of the slab (S) should be made equal to those of the upper and lower discs.
- 7. Care should be taken to screen off the apparatus from direct heating from the boiler.

References:

1. Chattopadhyay, D. R. *An Advanced Course in Practical Physics*. New Central Book Agency.

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2. Arora, C. L., B. Sc Practical Physics (Revised Edition), S. Chands Publication

3. To multiply two given matrices by using Fortran programming

Multiplication of two matrices:

Let *A* be an $m \times n$ matrix and *B* be another matrix of the order $p \times q$.

$$A = \begin{bmatrix} a_{11} & a_{12} & \dots & \dots & a_{1n} \\ a_{21} & a_{22} & \cdots & \cdots & a_{2n} \\ \dots & \dots & \dots & \dots & \dots \\ a_{m1} & a_{m2} & \cdots & \cdots & a_{mn} \end{bmatrix}$$

	[b ₁₁	b_{12}	•••	•••	b_{1q}^{-1}
	<i>b</i> ₂₁	b_{22}	•••	•••	b_{2q}
B =		•••	•••	•••	,
		•••			•••
	b_{p1}	b_{p2}	•••	•••	b_{pq}

We can evaluate the product of these two matrices provided the number of columns the matrix *A* is equal to the number of rows of the matrix *B* i. e. n = p.

Then the product of the two matrices can be written as X = AB

where *X* is another matrix of the order $m \times q$

Therefore

$$\begin{aligned} X \\ = \begin{bmatrix} a_{11}b_{11} + a_{12}b_{21} + \dots + a_{1n}b_{n1} & a_{11}b_{12} + a_{12}b_{22} + \dots + a_{1n}b_{n2} & \dots & a_{11}b_{1q} + a_{12}b_{2q} + \dots + a_{1n}b_{nk} \\ a_{21}b_{11} + a_{22}b_{21} + \dots + a_{2n}b_{n1} & a_{21}b_{12} + a_{22}b_{22} + \dots + a_{2n}b_{n2} & \dots & \dots & a_{11}b_{1q} + a_{12}b_{2q} + \dots + a_{1n}b_{nk} \\ & \dots & & \dots & & \dots & & a_{21}b_{1q} + a_{22}b_{2q} + \dots + a_{2n}b_{nk} \\ & \dots & \dots & & \dots & & \dots & \dots \\ a_{m1}b_{11} + a_{m2}b_{21} + \dots + a_{mn}b_{n1} & a_{m1}b_{12} + a_{m2}b_{22} + \dots + a_{mn}b_{n2} & \dots & \dots & a_{m1}b_{1q} + a_{m2}b_{2q} + \dots + a_{mn}b_{mn} \end{aligned}$$

In general the ij^{th} element of the matrix X is

$$x_{ij} = a_{i1}b_{1j} + a_{i2}b_{2j} + \dots + a_{in}b_{nj}$$
$$= \sum_{k=1}^{n} a_{ik} b_{kj}$$

All the $m \times q$ elements of the matrix X can be evaluated using the above equation.

Plan of the Program and Algorithm:

1. List of variables:

Input: Numbers of rows and columns m, n of the first matrix

Number s of rows and columns p, q of the second matrix

A and B are subscripted variables with two dimensional arrays.

Output: *X* is a two dimensional subscripted variable.

2. Formula:

$$x_{ij} = \sum_{k=1}^{n} a_{ik} \, b_{kj}$$

is the equation for evaluating each element of the product matrix

3. ALGORITHM:

- (1) Beginning of the program
- (2) Define variables m, n, p, q, A, B, X
- (3) input m, n, p, q
- (4) If $\neq p$, print multiplication not possible and go to step (3)

- (5) Start a loop for i=1 to m
- (6) Input a_{ij} of matrix A, row wise for j=1 to n, end of loop started at (5)
- (7) Start a loop for i=1 to p
- (8) Input b_{ij} of matrix B, row wise for j=1 to q, end of loop started at (7)
- (9) Start a loop for i=1 to m
- (10) Start a loop for j=1 to q
- (11) Initialize $x_{ij} = 0$
- (12) Start a loop for k=1 to n
- (13) Calculate $x_{ij} = x_{ij} + a_{ik} * b_{kj}$
- (14) End of loops started at (9), (10), (12)
- (15) Start a loop for i=1 to m
- (16) Print x_{ij} for j=1 to q, end of loop started at (15)
- (17) Stop execution
- (18) End

PROGRAM:

- C To multiply two given matrices integer m,n,p,q real a(10,10),b(10,10),x(10,10) open(2,file='matmultpln.out')
- 4 write(*,*)'enter the order of the first matrix'
 - read(*,*)m,n
 - write(*,*)'enter the order of the second matrix'
 - read(*,*)p,q
 - if(n.ne.p)then
 - write(*,*)'multiplication not possible: number of columns of
 - + first matrix not equal to number of rows of the second matrix'

```
goto 4
```

endif

write(*,*)'enter the elements of the first matrix, row wise'
do 6 i=1,m

```
6 read(*,*)(a(i,j),j=1,n)
```

write(*,*)'enter the elements of the second matrix row wise' do 8 i=1,p

```
8 read(*,*)(b(i,j),j=1,q)
do 10 i=1,m
do 10 j=1,q
x(i,J)=0.
```

do 10 k=1,n

- 10 x(i,j)=x(i,j)+a(i,k)*b(k,j) write(*,*)'The product matrix X = ' write(2,*)'The product matrix X = ' do 12 i=1,m write(*,14)(x(i,j),j=1,q)
 12 write(2,14)(x(i,j),j=1,q)
- 14 format(3x,10(3x,f8.2)) stop

```
end
```

Step by step Procedure (with examples):

The following procedure is for running the program using Windows based Force 2.0 compiler.

- 1. Start Force 2.0 compiler by double clicking on Force 2.0 shortcut on the desktop.
- 2. After a new file has opened up, save it with any name (eg. 'matmult.f')
- 3. You can now type the Program as given above.
- 4. To compile the Program click Run on the Menu bar and then click Compile.
- 5. To Run the Program click Run on the Menu bar and then click Run.
- 6. Immediately a new window 'name.exe' will open up displaying 'enter the order of the first matrix'
- 7. Now from the keyboard enter the values of matrix order, m and n.
- 8. Then you will be asked to enter the order of the second matrix viz. p and q.
- 9. If $n \neq p$ the message 'multiplication not possible' and you will be asked to enter the orders of another pair of matrices.
- 10. Now if n = p, the instruction to enter the elements of the first matrix will appear on the screen.
- 11. Type all the elements of the first row keeping as pace or using a coma between two consecutive numbers and press the 'enter' key. Then enter all the elements of the second row and so on.
- 12. When entry of first matrix is done, you will be asked to enter the elements of the second matrix and this is to be done as in step 11.
- 13. Now calculation of each element of the product matrix will be performed and the output of the program will be displayed on the screen.

- 14. To view the output file 'matmultpln.out' click file then click open on the menu bar. Then enter the file name ('matmultpln.out') and click open.
- 15. You can verify the results by calculating manually some of the elements of the product matrix taken at random from different rows and columns. Compare these results with the computer out

Bibliography

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4. To compute the roots of a quadratic equation by using Fortran programming

Quadratic Equation and the different types of roots:

A quadratic equation is an equation that has the following form

$$ax^2 + bx + c = 0$$

where a,b,c are the constants known as the coefficients and x is an unknown variable.

In general this equation has two solutions i.e. two roots or two values of x that satisfy it. These are

$$x_1 = \frac{-b + \sqrt{b^2 - 4ac}}{2a}$$
 and $x_2 = \frac{-b - \sqrt{b^2 - 4ac}}{2a}$

The quantity $b^2 - 4ac$ is denoted by 'D' and is called the discriminant because it can discriminate between the possible types of solutions.

- 1. When D > 0, the two roots x_1 , x_2 are real and distinct.
- 2. When D = 0, the two roots of the equation become

$$x_1 = x_2 = \frac{-b}{2a}$$

i.e. the roots of the equation are real and equal.

3. When D < 0, the two roots are complex quantities and conjugate to each other.

The real part of the roots is
$$\frac{-b}{2a}$$
 and the imaginary part is $\frac{\sqrt{b^2 - 4ac}}{2a}$

Plan of the Program and Algorithm:

1. list of variables:

Coefficients a, b, c are the input constants

Roots x_1 and x_2 are the output variables. For complex conjugate roots, real part (RP) and imaginary part (IP) of the roots are the output variables.

2. Formulae:

(a) Discriminant $D = b^2 - 4ac$ to determine the types of solutions

(b)
$$x_1 = \frac{-b + \sqrt{b^2 - 4ac}}{2a}$$
 and $x_2 = \frac{-b - \sqrt{b^2 - 4ac}}{2a}$ for real roots

(c)
$$RP = \frac{-b}{2a}$$
 and $IP = \sqrt{|b^2 - 4ac|}$ for complex conjugated roots

3. ALGORITHM:

- (1) Beginning of the program
- (2) Define variables x1, x2,x, RP, IP
- (3) Input coefficients a, b, c
- (4) Calculate Discriminant $D = b^2 4ac$
- (5) If D < 0 go to step (6)
 - If D = 0 go to step (9)
 - If D > 0 go to step (12)

(6) Calculate
$$RP = \frac{-b}{2a}$$
 and $IP = \sqrt{|b^2 - 4ac|}$

- (7) Print RP and IP
- (8) Go to step (14)
- (9) Calculate $x = \frac{-b}{2a}$
- (10) Print x
- (11) Go to step (14)

(12) Calculate
$$X1 = \frac{-b + \sqrt{b^2 - 4ac}}{2a}$$
 and $X2 = \frac{-b - \sqrt{b^2 - 4ac}}{2a}$

- (13) Print X1, X2
- (14) Stop execution
- (15) End

PROGRAM:

C Program To compute the roots of a quadratic equation Real x1,x2,x,rp,ip open (1,file='quad.out') write(*,*)'enter coefficients a,b,c' read(*,*)a,b,c d=b**2-4*a*c if(d)2,4,6
2 rp=-b/(2.*a)

- ip=sqrt(abs(d))/(2.*a) write(*,8)rp,ip write(1,8)rp,ip
- 8 format(3x, 'The roots x1, x2 are complex conjugates:',//,3x, 'Real

```
+ Part=',f8.2,//,3x,'Imaginary Part=',f8.2)
go to 14
```

- 4 x=-b/(2.*a) write(*,10)x write(1,10)x
- 10 format(3x,'Roots x1,x2 are equal:',//,3x,'Root x=',f8.2) go to 14
- 6 x1=(-b+sqrt(d))/(2.*a) x2=(-b-sqrt(d))/(2.*a) write(*,12)x1,x2 write(1,12)x1,x2
- 12 format(3x, 'The roots are real and distinct:',//,3x, 'x1=',f8.2,//

```
+ ,3x,'x2=',f8.2)
14 stop
end
```

Step by step Procedure (with examples):

The following procedure is for running the program using Windows based Force 2.0 compiler.

- 16. Start Force 2.0 compiler by double clicking on Force 2.0 shortcut on the desktop.
- 17. After a new file has opened up, save it with any name (eg 'quad.f')
- 18. You can now type the Program as given above.
- 19. To compile the Program click Run on the Menu bar and then click Compile.
- 20. To Run the Program click Run on the Menu bar and then click Run.
- 21. Immediately a new window 'name.exe' will open up displaying 'enter coefficients a, b, c'
- 22. Now from the keyboard enter the values of a, b, c (for eg 2. 3. 4.)
- 23. The output of the program will be displayed on the screen.
- 24. To view the output file 'quad.out' click file then click open on the menu bar. Then enter the file name ('quad.out') and click open.
- 25. You can verify the results by calculating manually and compare with the computer output.

Bibliography

- C. Xavier, FORTRAN 77 and Numerical Analysis, New Age International (2001)
- S. Lipschutz & A. Poe, Programming with Fortran

DEPARTMENT OF PHYSICS

YEAR No. OF APPLICANTS VIS-À-VIS SANCTIONED SEATS		STUDENTS	CUT OFF PERCENTAGE (%)	DETAILS OF STUDENTS ADMITTED				NTS	DROP OUT INCREASE IN THE PROPORTION		PASS	POSITIONS SECURED IN	PG ADMISSIONS
		ADMITTED		G	ST	SC	OBC	PH	RATE	OF HANDS ON EXPERIMENTAL WORK	PERCENTAGE	UNIVERSITY EXAMINATIONS	TO UNIVERSITY
		13		0	13	0	0	0	2 OUT OF 13	Nil	81.80%	NIL	NIL
2011 15	15	7	45	1	5	1	0	0	2 OUT OF 13		50%		
		1		0	1	0	0	0	NIL		100%		
		14		3	11	0	0	0	NIL	Nil	86%	NIL	3
2012	25	9	45	0	9	0	0	0	NIL		67%		
		3		1	2	0	0	0	NIL		100%		
2013	15	11	45	0	11	0	0	0	NIL	4 Expts + One Hands on Training	RESULTS AWAITED	RESULTS AWAITED	RESULTS AWAITED

DEPARTMENT OF ZOOLOGY

Activities of Faculty:

Activities	Name & Date of the Workshop/Seminar	Name of faculty	Organizing Dept./ Host
i.Seminar/ Workshop / Summer school attended	i)Summer School Dt- 22 nd August to 11 th September	Mr.Liston Marbaniang	Organized by Academic Staff College, NEHU, Shillong.
	 ii)A two day Workshop on "Faculty Training and Motivation and Adoption of Schools and Colleges by CSIR Labs" Dt21st &22nd March2013 	The faculty of the department	Organized by CSIR- NEIST, Jorhat, Assam. Venue- St. Edmund's College,Shillong.
	 iii)One day Seminar on "Youth Development" held as a part of Platinum Jubilee celebrations of St. Mary's College Dt6th June2013 	The Faculty of the department	Organized by St.Marys College ,Shillong
	iv)One day workshop on "Instrumental Techniques and their Applications" Dt 18 th July 2013	Mrs.A.P. Shabong &Dr.(Mrs.)Neerja Mishra	Organized by department of Chemistry, St. Mary's College, Shillong.
	v)A National Workshop on " Maintenance of Opto-Analytical Instruments " Dt14 th to 21 st September 2013	All the faculty of the department	Organized by department of Chemistry, St. Anthony's College in association with Western Regional Instrumentation Centre, Mumbai.

	vi) A two day International Workshop on: Women in Leadership- the unknown women . Dt: 23 rd &24 th October,2013.	Mrs. A.P. Shabong	Organized by the German Development Corporation (GIZ) and facilitated by - Academy of Human Excellence at the Asian confluence, based at Baroda,Gujrat.
ii.Training	i)One day programme on "	Mrs.A.P.Shabong	Organized by Meghalaya
programm	International Biodiversity Day"	&	Biodiversity Board &
es/Other Scientific activities	Dt22 nd May2013	Dr.(Mrs.)N.Mishra	Dept. of Forests & Environment, Meghalaya
attended			Dt22 May2015
	ii) Celebration of "Golden Jubilee of teaching Science in Shillong College"Dt7th June2013	Mrs.A.P.Shabong & Dr.(Mrs)N.Mishra	Organized by the Science Faculty of Shillong College,Shillong.
	 iii) Three day training workshop on "Biological Techniques" Dt. 10th -12th October,2014 	Ms. Y.B.Nongrum & Mr. L. Marbaniang	DBT, Biotech Hub, Synod College, Shillong

Student Studistics.												
Year	No. of applicants vis-à-vis sanctioned	Students admitted	Cut off %	G	Deta ST	admi SC	students tted OBC	PH	Drop out rate	Pass Percentage	Positions secured in University	Pg Admissions to
	seats											university
	1 st Year- 31	31	45%	1	30	-	-	-	45.16%(14 dropped)	94.1%(17 Appeared,16 passed)	1 (Puthi	
2011 2 ⁿ	2 nd Year- 19	18	NA	4	14	-	-	-	15.78%(3 dropped)	93.3%(15 appeared,14 passed)	Lalchawisang- 1 st class 1 st	4
	3 rd Year- 10	10	NA	-	10	-	-	-	0%	100%	position)	
	1 st Year- 35	35	50%	2	33	-	-	-	45.71%(16 dropped)	94.1%(16 appeared, 15 passed)	3 (Pinky Sultana-1 st	
2012	2 nd Year- 17	17(16+1 repeater)	NA	1	16	-	-	-	0%	86.66%(15 appeared, 13 passed)	class 2 nd , Sanjana Sinha-1 st class	6
	3 rd Year- 16	16(14+2 repeaters)	NA	3	13	-	-	-	0%	100%	7 th , Debora Mawrie-1 st class 8 th)	
	1 st Year- 35	35	55%	2	33	-	-	-	11.42%(4 dropped)	Result Awaited(RA)		Posult
2013	2 nd Year- 15	15	NA	2	13	-	-	-	0%	RA	Result Awaited(RA)	Awaited
	3 rd Year- 13	13	NA	-	13	-	-	-	0%	RA		

Student Statistics:

Student Activities:

Activities	Purpose	Class	Date
1.Industrial visit			4
i)Visit to Sericulture Farm	To study the method of rearing of silk moths and extraction of silk	B.Sc. IIIrd Year	12 th Nov., 2012 & 5 th July, 2013
ii) Visit to Animal Husbandry & Veterinary Department ,Govt. of Meghalaya	To study the zoonotic diseases that affect livestock	B.Sc. IIIrd Year.	18 th June, 2013
2.Research Labs			
i)Visit to Institutional Biotech Hub,Synod College , Shillong	To understand the basic concept of Biotechnology and Demonstration of DNA Separation and PCR	B.Sc. IInd Year & B.Sc.IIIrd Year	7 th June2013
ii)Visit to Sophisticated Analytical Instrumentation Facility ,NEHU, Shillong	To understand the working mechanism and applications of some of the instruments used in biological sciences	B.Sc. IInd Year	16 th July, 2013
3.Training Programme attended			
i)Visit to Hatchery unit,Dept. of Fisheries,Mawpun, Govt. of Meghalaya	Induced Breeding Techniques in carps	B.Sc. IIIrd Year	3 rd July 2013

4.Seminar /Workshops attended	1.One day seminar on "Youth Development" organized by	BSc. IIIrd year Honours	6 th June 2013
	St. Marys College, Shillong.	students	
	2. A two day National seminar on "Burning Enviornmental Issues : Risk to Biodiversity and Human Health with special reference to North East India" organized by St. Mary's College, Shillong.	BSc 2 nd year and Final year students	15 th and 16 th May 2013.

3.Projects undertaken	Торіс	Class / Name of the student	Submission date
1.Group Project work	i) Study of	B.Sc. Final Year	Submitted in
	Sericulture Practices	Students	Dec.2012
	in Meghalaya		
2.Individual projects	i) DDT and its effects	Pinky Sultana	NovDec.2012
taken by B.Sc. Final			
Year students under			
nrogramma			
	ii) Mycoplasma	Genevieve Lakadong	
	Pneumonia		
	111) Huntingtons	Euginia Tete	
	Chorea	Syiemlieh	
	iv) Ecological		
	Importance of	Minee Bhuyan	
	wetlands		

v) Disorders of	Silvary Khongjah	
Pancreatic Endocrine		
function		
Tunetion		
·	Linda Sailo	
v1) Prions		
vii) Living boolthy	Aitihun Marthong	
with Asthma		
with Astillia		
viii) Programmed cell	Vanlalduhsaki	
death		
ix) ADD (Attention		
Deficit Disorder)	Suzie S. Nongbet	
x)Fish Farm		
management	x 11 1 1	
	Lanangaibiang Thabah	

Name of the guest speaker	Designation	Date	Host Institute	Торіс
Capt.Dapple.D.Wankhar	Captain(nursing)	12.7.13	AFMC, Pune	Career Prospects and Scope of B.Sc. nursing in Armed Forces
Mr.Jeremy Syiem	Junior Research Fellow, under DBT scheme	4.6.13	Synod College, Shillong	Basic Techniques in Biotechnology

Guest Lectures /Invited lectures organized:

Interdepartmental activities :

i)Department of Zoology, Chemistry, Geography and Botany organized a National Seminar on

"Burning Environmental Issues: Risk to Biodiversity and Human health with special references to North East India" on $15^{th} - 16^{th}$ May,2013.

ii) Department of Zoology and Botany held an annual programme on 5th June to commemorate World Environment Day.

Training Course attended by laboratory staff:

Name of Laboratory staff	Organizing Department	Title of workshop	Date
Mrs. Bissina	North Eastern Hill	Maintenance of	24th to 28th Sept.
Marbaniang	University in	Electronic Laboratory	2012
	collaboration with	Instruments	
	Western Regional		
	Instrumentation		
	Centre, Mumbai.		

Procurement of new equipment:

List of equipments, books and journals procured under DBT support

1. List of equipments:

Sl. No.	Product Name	Company	Model	Quantity	Dealer	Price (inclusive of VAT) in Rs	Date of Receipt	Date of Installatio n
1.	LCD	Epson	EB-XO2	1	Ambitious		19/3/13	
	Projector				Enterprise	73,775		
2.	CCD	DeWinte	Digieye	1	Ambitious		19/3/13	12/7/13
	Camera	r			Enterprise			
3.	Desktop	HP	P-2 1403	1	Hue		20/3/13	20/3/13
	with				Service			
	monitor				Pvt. Ltd	-		
4.	Laptop	HP	G6-	1	Hue		20/3/13	20/3/13
	with carry		2204TX		Service	95,356		
	case				Pvt. Ltd	-		
5.	Laserjet	HP	M1536D	1	Hue		20/3/13	20/3/13
	Printer		NF		Service			
					Pvt. Ltd			
6.	UPS	Intex	600VA	1	Hue		20/3/13	20/3/13
					Service			
					Pvt. Ltd			
7.	DSLR	Canon	EOS1100	1	Highland	38,990	22/12/12	22/12/12
	Camera		D		Studio			
8.	UV Filter	Canon	-	1	Highland	1150	22/12/12	22/12/12
					Studio			
9.	Projection	Wesmox	MP-385A	1	Sun		8/3/13	15/7/13
	Microscop				Scientific			
	e					-		
10.	Student	Olympus	HSA	15	Sun	1,73087	8/3/13	8/3/13
	Monocular				Scientific			
	Microscop							
	е							
11.	Haemocyt	Neubaeu		5	Sun		28/5/13	28/5/13
	ometer	r			Scientific	21,281		
12.	Haemomet	Neubaeu		5	Sun		28/5/13	28/5/13
	er	r			Scientific			

Sl. No.	Title of the Book	Author	Publisher	Dealer	Quantity	Amount	Date of Receipt
1.	Immunology& Medical	Sinha,S.K.	Oxford	Eastern	1	1,800	16/2/13
	Zoology			Book			
			~	House			
2.	Biological Sciences	Mitra, S.	Campus	Eastern	1	1,350	16/2/13
			BOOKS	BOOK			
2	Coll Diology	Cunto/Iongin	Agno	House	1	1 800	16/2/12
5.	Cell Diology: Fundamentals &	Gupta/Jangir	Agro	Book	1	1,000	10/2/13
	Applications			House			
4	Water & Sewage	Theroux	Agro	Eastern	1	695	16/2/13
-10	Water & Bewage	Incidux	11gi U	Book	1	070	10/2/10
				House			
5.	Animal Ecology	Bhaskar, H. V.	Campus	Eastern	1	825	16/2/13
			Books	Book			
				House			
6.	Animal Embryology	Sandhu, G.S.	Campus	Eastern	1	950	16/2/13
			Books	Book			
				House			
7.	Basic Concepts of	Singh, V.B.	ALP Books	Eastern	1	850	16/2/13
	Zoology			Book			
				House			
8.	H.B. of Cell Biology	Sharma ,P.	Wisdom	Eastern	1	995	16/2/13
				Book			
				House			
9.	H.B. of Modern	Bahl,B.	Cyber Tech	Eastern	1	800	16/2/13
	Immunology			Book			
10			A /1 /1	House	4	450	1 (10 (1 0
10.	T.B. of Biomedical	Venkatesan, P.	Atlantic	Eastern	1	450	16/2/13
	Laboratory Techniques		Publications	BOOK			
11	T.D. of Laboratory	Cogwami C	Wisdom	House	1	005	16/2/12
11.	T.D. Of Laboratory	Goswann, C.	vv isuoili	Book	1	995	10/2/13
	reeninques			House			
12.	Applied Fisheries and	Singh, B.K.	Swastik	Eastern	1	895	16/2/13
12.	Aquaculture	Singh, D.IX.	5 wustik	Book	1	070	10/2/10
				House			
13.	H.B. of Fisheries and	Sharma, O.P.	Agrotech	Eastern	1	500	16/2/13
	Aquaculture	,		Book			
	-			House			
14.	Intro. To Animal	Jindal, S.K.	Nipa	Eastern	1	795	16/2/13

ii) Procurement of Books and Journals:

Sl. No.	Title of the Book	Author	Publisher	Dealer	Quantity	Amount	Date of Receipt
	Physiology			Book House			
15.	Insect Pest management: Ecological Concepts	Sathe, T. V.	Daya	Eastern Book House	1	700	16/2/13
16.	Animal Behaviour	Bhaskar, H.V.	Campus Books	Eastern Book House	1	900	16/2/13
17.	Chordates (2 Vols.)	Bhaskar, H.V.	Campus Books	Eastern Book House	1	1,850	16/2/13
18.	Molecular Genetics & Biotechnology	Srivastava,S.	Swastik	Eastern Book House	1	795	16/2/13
19.	Invertebrates	Misten	Ivy	Eastern Book House	1	250	16/2/13
20.	H.B. of Parasitology	Awsathi, A.K.	Wisdom	Eastern Book House	1	850	16/2/13
21.	Invertebrates (2 Vols.)	Vardhan, H.	Campus Books	Eastern Book House	1	1,750	16/2/13
22.	Research in Zoology	Pande, P.	Swastik	Eastern Book House	1	800	16/2/13
23.	Ponds & Fish Culture	Hall, C.B.	Agro	Eastern Book House	1	595	16/2/13
24.	Evolutionary Biology	Sharma, A.	Adhyayan	Eastern Book House	1	750	16/2/13
25.	Genetics	Elrod and stansfield	Tata Mcgraw-Hill	Galaxy Book Centre	1	505	28/3/13
26.	Genetics & Molecular Biology	D.R.Hyde	Tata Mcgraw-Hill	Galaxy Book Centre	1	780	28/3/13
27.	Vertebrates: Comparative Anatomy, Function,Evolution	Kardong, K.V.	Tata Mcgraw-Hill	Galaxy Book centre	1	1000	31/5/13

Sl. No.	Title of the Book	Author	Publisher	Dealer	Quantity	Amount	Date of Receipt
28.	An Introduction to Practical Biochemistry	D.T Plummer	Tata Mcgraw-Hill	New Book Centre	1	250	15/3/13
29.	Animal Behaviour	H.S.Gunderia, H.S.Singh	S.Chand	New Book Centre	1	135	15/3/13
30.	Cell Biology	C.B.Powar	Himalaya Publishing House	New Book Centre	2	550	15/3/13
31.	Chordate Embryology	Verma & Agarwal	S.Chand	New Book Centre	1	200	15/3/13
32.	Economic Zoology	Shukla & Upadhyay	Rastogi	New Book Centre	1	245	15/3/13
33.	Invertebrate Zoology	Jordan &Verma	S.Chand	New Book Centre	1	545	15/3/13
34.	Chordate Zoology	Jordan &Verma	S.Chand	New Book Centre	1	450	15/3/13
35.	Experimental Biology	Abhijit Dutta	Narosa	New Book Centre	1	260	15/3/13
36.	Practical Zoology Invertebrates	S.S.Lal	Rastogi	New Book Centre	1	260	15/3/13
37.	Practical Zoology Vertebrates	S.S.Lal	Rastogi	New Book Centre	1	260	15/3/13
38.	Fundamental Ecology	Odum & Barret	Brookes/Cole Cengage Learning	New Book Centre	1	599	15/3/13
39.	Endocrinology	Hadley & Levine	Pearson	New Book Centre	1	750	15/3/13
40.	Evolution,Comparative Anatomy, Biometry,Economic Zoology & Animal Development Vol5	Pandey	Tata Mcgraw-Hill	Galaxy Book Centre	1	265	1/2/13

Sl. No.	Title of the Book	Author	Publisher	Dealer	Quantity	Amount	Date of Receipt
41.	Biochemistry,	Pandey	Tata	Galaxy	1	325	1/2/13
	Physiology &		Mcgraw-Hill	Book			
	Endocrinology Vol3			Centre			
42.	Animal Diversity Vol 1	Pandey	Tata	Galaxy	1	325	1/2/13
			Mcgraw-Hill	Book			
				Centre			
43.	Cytology, Genetics &	Pandey	Tata	Galaxy	1	325	1/2/13
	Molecular Genetics		Mcgraw-Hill	Book			
	Vol2			Centre			
44.	Life-The Science of	Purves et al	W.H.	Book	1	6500	25/2/13
	Biology with CD		Freeman &	Friends			
			Co.				
45.	Animal Physiology	Sherwood et al	Brooks/Cole	Book	1	7900	8/6/13
			Cengage	Friends			
			Learning				
46.	Cells	Lewin et al	Jones	Book	1	4590	8/6/13
			Bartlett	Friends			

B) Chemicals , Glassware, Classwork material

Sl. no	Product Description	Company	Quantity	Dealer	Price	Date of Receipt
1.	DPX Mountant	Merck	4x250ml	Sun Scientific	960.00	14/03/13
2.	Folin's Reagent	Merck	3x100ml	Sun Scientific	1050.00	14/03/13
3.	Acetone	Himedia	2x500ml	Sun Scientific	600.00	14/03/13
4.	Chloroform	Himedia	2x500ml	Sun Scientific	740.00	14/03/13
5.	Olive Oil	Himedia	1x250ml	Sun Scientific	1090.00	14/03/13
6.	Butanol	Himedia	1x2.5L	Sun Scientific	1355.00	14/03/13
7.	Hydrochloric acid	Himedia	2x500ml	Sun Scientific	450.00	14/03/13

Sl. no	Product Description	Company	Quantity	Dealer	Price	Date of Receipt
8.	Xylene low in sulphur	Himedia	1x2.5L	Sun Scientific	1080.00	14/03/13
9.	Ferric chloride hexahydrate	Himedia	1x500g	Sun Scientific	548.00	14/03/13
10.	Measuring Cylinders 10ml	Borosil	4 nos.	Sun Scientific	840.00	14/03/13
11.	Measuring Cylinders 50ml	Borosil	12 nos.	Sun Scientific	3300.00	14/03/13
12.	Measuring Cylinders 100ml	Borosil	10 nos.	Sun Scientific	3150.00	14/03/13
13.	Conical flask 25ml	Borosil	12 nos.	Sun Scientific	720.00	14/03/13
14.	Conical flask 50ml	Borosil	12 nos.	Sun Scientific	720.00	14/03/13
15.	Conical Flask 100ml	Borosil	6 nos.	Sun Scientific	360.00	14/03/13
16.	Measuring cylinders 10ml	Borosil	8 nos.	Sun Scientific	1680.00	28/05/13
17.	Slides	Abdos	50pkts(1Box)	Sun Scientific	2400.00	14/03/13
18.	Cover Slips 18mm square	Blue Star	20 pkts(1 box)	Sun Scientific	1300.00	14/03/13
19.	Cover Slips 2mm	Blue Star	20 pkts(1 box)	Sun Scientific	140.00	14/03/13
20.	Nauplius larva	Sc.House	6 phials	Sun Scientific	132.00	4/04/13
21.	Spicules of sponges	Sc.House	6 phials	Sun Scientific	132.00	4/04/13
22.	Mysis larva	Sc.House	6 phials	Sun Scientific	132.00	4/04/13

Sl. no	Product Description	Company	Quantity	Dealer	Price	Date of Receipt
23.	Scoliodon	Sc.House	138 pcs	Sun Scientific	4140.00	27/09/12
24.	Mysis	Sc.House	6 phials	Sun Scientific	150.00	27/09/12
25.	Glass slides	Blue Star	10 pkt	Ambitious Enterprise	600.00	19/01/13
26.	Blood Lancets		6 pkts	Ambitious Enterprise	2400.00	19/01/13
27.	Glass Droppers		12 nos.	Ambitious Enterprise	120.00	19/01/13
28.	Aluminium Foil		1 roll	Ambitious Enterprise	42.00	19/01/13
29.	Phenolphthalein Indicator Solution		2 x125ml	Ambitious Enterprise	196.00	19/01/13
30.	Measuring Cylinder 500ml	Borosil	5	Sun Scientific	3775.00	28/5/13
31.	Measuring Cylinder 1000ml	Borosil	5	Sun Scientific	5725.00	28/5/13
32.	40X Objective HAS/HB	Olympus	10	Sun Scientific	19159.00	8/3/13

Standard Operating Procedure

Department of Zoology, St. Mary's College, Shillong

ECOLOGY

List of practicals:

- 1. Estimation of oxygen in different water samples.
- 2. Estimation of free carbon dioxide in water samples.
- 3. Estimation of Total alkalinity of water samples.
- 4. Estimation of total hardness of water samples.
- 5. Estimation of pH and temperature of water samples.
- 6. Qualitative study of plankton from fresh water samples
- 7. Quantitative estimation of plankton from fresh water samples

1. ESTIMATION OF O2 IN DIFFERENT WATER SAMPLE.

<u>Principle</u>: The amount of oxygen present in water sample is determined indirectly by the amount of iodine present. In the iodometric titration, the amount of O_2 present. The method is based on the oxidation-reduction reaction.

The overall reaction is:

 $MnSO_4 + 2KOH \longrightarrow Mn(OH)_2 + K_2SO_4$

In the presence of O_2 , in an alkaline solution media $Mn(OH)_2$ is oxidized to $Mn(OH)_3$ which is yellow precipitate.

 $4Mn(OH)_2 + O_2 + H_2 \Theta \longrightarrow 4Mn(OH)_3$

The yellow precipitate of Mn(OH)₃ is dissolved upon the addition of conc. H₂SO₄.

 $4Mn(OH)_2 + 6H_2SO_4 \rightarrow 4MnSO_4 + 2H_2O + 2SO_4^{2-}$

The $SO_4^{2^2}$ generated reacts with KI to form K_2SO_4 and I_2 is liberated. The liberated I_2 is proportional to the amount of O_2 present in the water sample.

 $2SO_4^{2-} + KI \longrightarrow 2K_2SO_4 + 2I_2$

The liberated I_2 is determined by titrating against 0.025 (N) Na₂S₂O₃ using starch solution as indicator. The end point is marked when the blue colour complex changes to colourless solution.

REQUIREMENTS

Conical flasks, pipette, burette, dropper, measuring cylinder, BOD bottles.

PROCEDURE

- 1. Water samples from different sources were collected in 125 BOD bottles, taking care that no air bubbles are formed.
- 2. To this 1ml of MnSO₄ and 1ml of alkaline iodide (KI) were added.
- 3. The mixture forms a brown precipitate. The mixture was allowed to stand for 10 minutes for the precipitate to settle down.
- After the precipitate has settled down at the bottom, 1 ml of conc. H₂SO₄ was added. The precipitate dissolves and a brown solution is observed.
- 5. 25 ml of the fixed solution was taken in a conical flask and 1 or 2 drops of starch solution was added.
- The solution was titrated against the 0.025 (N) Na₂S₂O₃ till the colourless was obtained. The volume of titrant used was noted down.
- 7. The experiment was repeated twice or thrice for each of the water sample.

OBSERVATION

Water Sample	No. of Observation	Vol. of water taken	Vol. of 0.025 (N) Na ₂ S ₂ O ₃			
			Initial Reading(ml)	Final Reading(ml)	Difference	Mean (ml)
Sample 1						
Sample 2						
Sample 3						

CALCULATION

8 x 1000 x N x Vol. of Titrant

Dissolved O₂ (DO) mg/l = -----

Vol. of sample taken

Where, N = Normality of titrant

2. ESTIMATION OF FREE CO2 IN WATER SAMPLES

<u>**Principle**</u>: Na_2CO_3 from carbonic acids. The formation of which is indicated by indicators. CO_2 undergo or chemical reaction, it does not remain as such.



It is the normal practice to distinguished free CO_2 as the concentration of CO_2 and H_2 CO_3 which is estimated by the titration sample with standard alkali titrate to pH - 8.5

REQUIRED MATERIALS

Water sample, 0.045 (N) Na₂CO₃, Phenolphthalein, measuring cylinder, pipette, burette, conical flask.

PROCEDURE

- 1. 50 ml of water sample was taken in a conical flask. To it 2 3 drops of phenolphthalein (if colour turns pink free CO₂ absent, if no colour or remain colourless, free CO₂ is present)
- 2. This was then titrated against 0.045 (N) Na₂CO₃ till a light pink colour appears. It is the end point and volume of titrant used was noted.
- 3. The above procedure was repeated for three samples.
- 4. Each sample was tested three times.

OBSERVATION

Vol. of water	Vol. of 0.045 (N) Na ₂ CO ₃ used			Moon
sample taken	Initial Reading	Final Reading	Difference	Iviean
	1.			
Sample 1	2.			
	3.			
	1.			
Sample 2	2.			
	3.			
	1.			
Sample 3	2.			
	3.			

CALCULATION

Vol. of titrant used Free CO₂ = ----- x 1000 mg/l

Vol. of sample taken

3. ESTIMATION OF TOTAL ALKALINITY OF WATER SAMPLES

<u>Principle</u>: In most fresh water ecosystem, bicarbonate and carbonate ions are present in considerable amount. These salts get hydrolysed in solution and produced hydroxyl ion consequently raising the pH.

$$H^+ + HCO_3^- + H_2O$$
 $H^+ + H_2CO_3 + OH^-$

Alkalinity of water sample depends upon the amount of CO_3^- and HCO_3^- ions present in it. Normally, CO_2 divided in water are combined to form H_2CO_3 which dissociate to H^+ and HCO_3^- . The alkalinity is determined by titrating the sample against a standard solution of a strong acid using Phenolphthalein (alkalinity due to carbonates and hydrides), and methyl orange indicator (alkalinity due to carbonate).

REQUIREMENT

0.02 (N) HCL/ H₂SO₄, Phenolphthalein, Methyl Orange, water sample, conical flask, measuring cylinder, pipette, burette, etc.

PROCEDURE

- 1. 50 ml of H_2O is taken in a conical flask.
- 2 drops of Phenolphthalein is added to water sample, a pink colour appears and is titrated against 0.02 (N) HCL/ H₂SO₄ till the colour disappears.
- 3. 2-3 drops of methyl orange is added to the sample and titrated with the same titrant with the same titrant till a pink colour appears.
- 4. The above procedure is repeated 3 4 times for each sample.

OBSERVATION

Vol. of water	Vol. of 0.02 (N) H_2SO_4 used ml			Moon
sample taken	Initial Reading	Final Reading	Difference	Ivicali
Sample 1	1.			
	2.			
	3.			
	1.			
Sample 2	2.			
	3.			

CALCULATION

Vol. of titrant used

Total Alkalinity = ----- x 1000 mg/l

Vol. of sample taken

4. ESTIMATION OF TOTAL HARDNESS OF WATER SAMPLES

<u>PRINCIPLE</u>: Eriochrome black T forms wine red complex compound with metal ions $(Ca^{2+} and Mg^{2+})$. The disodium salt of EDTA extracts the metal ions from the dye- metal ion complex as colourless chelate complexes leaving a blue colour aqueous solution of the dye.

REQUIREMENT

Conical flask, pipettes, droppers, and burettes,

Standard EDTA titrant (0.01M), Eriochrome black T indicator and ammonia buffer solution.

PROCEDURE:

- 1. 50 ml of the water sample was taken in a conical flask. Then 1 ml of ammonia buffer solution and 5 drops of indicator added to it. The colour of the sample turns wine red.
- 2. It was then titrated with EDTA titrant, until a blue colour appear.
- 3. The reading was noted and the total hardness is calculated.
- 4. The above procedure was repeated three times.

Samples	No of observation	Initial Burette	Final Burette	Mean (ml)
		Reading (ml)	Reading (ml)	
1				
2				
3				

OBSERVATION

CALCULATION

Vol. of titrant used

Total hardness mgl^{-1} as $CaCO_3 = ----- X 1000$

Vol. of sample taken

5. ESTIMATION OF pH & TEMPERATURE OF GIVEN WATER SAMPLE

INTRODUCTION:

Hydrogen ions (H^+) and Hydroxyl ions (OH^-) results from the ionization of water therefore any change in concentration of one these ions brings about the change in the concentration of other. Therefore a simple scale of numbers called the pH scale is used to measure the acidity or alkalinity of water and the number express the concentration of H^+ indirectly.

pH can be defined as the negative logarithm of H⁺ concentration and is mathematically express as:

$pH=-log[H^+]$

Where $[H^+]$ is the amount of H^+ in moles per litre of the solution of the solution.

In pure water 1/10,000,000 of the molecules are ionized. These numbers are written as 10^{-7} and the pH of water is said to be 7. In pure water the number of H⁺ and OH⁻ are equal; therefore, it is neither acidic nor alkaline but neutral. If the H⁺ is increased 100 folds then the concentration will be 1/1,000,000,000 or 10^{-9} or pH= 9. This is an alkaline solution. The pH scale runs from 1 - 14 with 7 indicating as neutral. The number below 7 indicates acidic medium and above 7 indicates alkaline medium.

PROCEDURE:

1. The pH of water sample is measured electrochemically from a pH meter. These instruments are used with electrodes that are charge in the electric property of the solution. This charge reach electrode and accuracy is greatest in the middle pH range. However, the instruments are provided with suitable devices to measure extremes of pH very accurately.

- 2. The pH is standardized with a standard buffer solution. The instrument is adjusted for temperature, following the manufacturer instruction at the pH value of the standard and after the warm up time.
- 3. The instrument is balance to eliminate asymmetrical potential. The buffer is removed and the electrode is washed carefully with the distilled water.
- 4. The electrode is place in a beaker containing the sample(s) and the pH reading is taken.

Sl. No.	Sample	рН
1		
2		

5. Qualitative study of plankton from fresh water samples

Requirement: Microscope, plankton sample, dropper, slide.

Introduction: The term plankton was coined by Victor Henson in 1887 to designate the suspended microscopic materials minute organisms and detritus in water which wander at the mercy of winds and tides. However, the use of the term has been confined to designate only the microscopic, free floating organisms which depending on their nature are divided into two major groups, namely phytoplankton and zooplankton.

On the basis of their size they have been classified as ultra (0.5 - 10 um), nano (10 - 50 um), micro or net (50 - 500 um), and macroplankton (> 500 um).

Procedure:

- 1. Shake the concentrated plankton sample and put one drop on a clean slide with the help of a dropper.
- 2. Put the slide under the microscope and observe the plankton in the sample.
- 3. Record plankton present, identify them with the help of textbooks available.

6. Quantitative estimation of plankton from fresh water samples.

Requirement: Microscope, Sedgewick – Rafter cell, pipette or standard dropper, cover slip.

Procedure:

- 1. Put the coverslip diagonally on the cell cavity.
- 2. Shake the sample gently and transfer quickly 1 ml. of it into the cavity with the help of a graduated pipette.
- 3. Adjust the coverslip properly to cover the cavity without air bubbles.
- 4. Focus on one edge of the cavity and move the slide horizontly, simultaneously counting the plankton till the other edge.

Observation: Phytoplankton / Zooplankton

Sl. No.	Name of species	Counting of each species observed using tally mark	Total no. of each species.

Community similarity and species diversity are calculated using Sorensen Similarity Index and Shannon Weiner's Diversity Index.
Standard Operating Procedure

Department of Zoology, St. Mary's College, Shillong.

BIOCHEMISTRY & MOLECULAR BIOLOGY

List of practicals:

- 1. Estimation of DNA using diphenylamine reagent.
- 2. Estimation of RNA using orcinol reagent.
- 3. Demonstration of gel electrophoresis for separation of DNA.
- 4. Demonstration of radial immunodiffusion.
- 5. Estimation of glucose using anthrone reagent.
- 6. Estimation of protein by Lowry's method.
- 7. Detection and Separation of amino acids by paper chromatgraphy.
- 8. Estimation of Ascorbic acid content of lemon.
- 9. Qualitative tests for carbohydrates, proteins and fats.

AIM: Estimation of DNA using diphenylamine reagent

PRINCIPLE:

When DNA is treated with diphenylamine under acid conditions, a blue compound is formed with a sharp absorption maximum at 595 nm. This reaction is given by 2-deoxypentoses in general and is not specific for DNA. In acid solution, the straight form of the deoxypentose is converted to the highly reactive β -hydroxyevulin aldehyde which reacts with DNA to produce a blue coloured complex. In DNA only the deoxyribose of purine nucleotides reacts so that the value obtained represents only one half of the the deoxyribose present.

CHEMICALS REQUIRED:

- 1) **Diphenylamine reagent:** Dissolve 1 g in 100ml of glacial acetic acid and add 2.5 ml of conc. H₂SO₄. Solution should be prepared fresh.
- 2) **Standard DNA solution (400µg/ml):** Add 40 mg of DNA to 100 ml of 0.9 % NaCl solution.
- 3) Unknown DNA sample or nucleic acid extract.
- 4) Distilled water

OTHER REQUIREMENTS:

- 1) Colorimeter/spectrophotometer
- 2) Cyclomixer
- 3) Test tubes, stands, beakers ,pipettes
- 4) Water bath, marbles

PROCEDURE:

- Different volumes of standard DNA sample (0.1 ml, 0.2 ml, 0.4 ml, 0.6 ml, 0.8 ml & 1.0 ml) and unknown sample (0.25 ml & 0.5 ml) are taken in different test tubes in duplicate.
- 2) The volume in each test tube is made to 1 ml by adding distilled water.
- 3) A blank test tube is also prepared by taking 1 ml of distilled water.
- 4) To each test tube, 2 ml of diphenylamine reagent is added and the solution is mixed thoroughly in a cyclomixer.
- 5) The test tubes are kept in a boiling water bath for 10-15 minutes and then cooled.
- 6) The optical density is then taken at 595 nm in a colorimeter/ spectrophotometer against the blank tube and then proceeded for the solution in the other tubes.

OBSERVATIONS:

Sl. No.	Volume of Standard DNA taken (Volume of distilled water added to make	Amount of DNA (µg)	Volume of diphenylamine reagent added	Optical density at	Mean optical density at
	ml)	1 ml (ml)		(ml)	595 IIII	595 nm
1.	0.1	0.9	40			
2.	0.1	0.9	40			
3.	0.2	0.8	80			
4.	0.2	0.8	80			
5.	0.4	0.6	160	2 ml in all		
6.	0.4	0.6	160	test tubes		
7.	0.6	0.4	240			
8.	0.6	0.4	240			

TABLE: For standard DNA (400µg / ml)

9.	0.8	0.2	320
10.	0.8	0.2	320
11.	1.0	-	400
12.	1.0	-	400
13.	Blank	1.0	-

TABLE: For unknown DNA sample

Sl.	Volume of	Volume of	Volume of	Optical density	Mean optical
No.	unknown DNA	distilled water	diphenylamine	at 595 nm	density at
	sample taken	added to make	reagent added (595 nm
	(ml)	1 ml (ml)	ml)		
1.	0.25	0.75	2 ml in all test		
2.	0.25	0.75	tubes		
3.	0.5	0.5			
4.	0.5	0.5			

GRAPH:

CALCULATIONS:

From the standard graph, **A** O.D. \equiv **B** µg of DNA

Therefore, $1 \text{ O.D.} \equiv \mathbf{B} / \mathbf{A} \mu g \text{ of } DNA = \mathbf{C} \mu g \text{ of } DNA$

• For unknown sample 1 (0.25 ml), O.D. value is **D**

Since, $1 \text{ O.D.} \equiv \mathbf{C} \mu g \text{ of DNA}$

Therefore, **D** O. D. \equiv **C** x **D** = **E** μ g of DNA

0.25 ml of unknown sample contains $\mathbf{E} \ \mu g$ of DNA

Therefore, 1ml of unknown sample contains $E/0.25 = F \mu g$ of DNA

• For unknown sample 2 (0.5 ml), O.D. value is G

Since,	$1 \text{ O.D.} \equiv \mathbf{C} \ \mu \mathbf{g} \ \mathbf{o}$	of DNA
--------	------------------------------------------------------------------	--------

Therefore, **G** O. D. \equiv **C** x **G** = **H** μ g of DNA

0.5 ml of unknown sample contains H µg of DNA

Therefore, 1ml of unknown sample contains $H/0.5 = J \mu g$ of DNA

Amount of DNA / ml in the unknown sample = $\mathbf{F} + \mathbf{J} = \mathbf{K} \mu g$

RESULT: The concentration of DNA in the unknown sample is found to be $K \mu g /ml$.

DISCUSSION:

AIM: Estimation of RNA using orcinol reagent

PRINCIPLE:

This is a general reaction for pentose (ribose) and depends on the formation of furfural. When the pentose undergoes decomposition on heating with concentrated HCl, furfural is formed which reacts with the orcinol reagent in the presence of of $FeCl_3$ to give ablue green coloured complex. Orcinol is a natural phenolic organic compound. It should be noted that the positive result for the orcinol test for RNA (pentoses) in general is a blue coloured compound which shows maximum absorbancy at 665 nm.

CHEMICALS REQUIRED:

- 1) Orcinol reagent: Dissolve 100 mg of ferric chloride (FeCl₃.6 H₂0) in 100 ml of concentrated HCl and add 3.5 ml of 6 % orcinol (W/v in alcohol).
- **2)** Standard RNA solution (200 μg/ ml): Add 20μg of RNA to 100 ml of saline or distilled water.
- 3) Unknown RNA sample or nucleic acid extract.
- 4) Distilled water

OTHER REQUIREMENTS:

- 1) Colorimeter/spectrophotometer
- 2) Cyclomixer
- 3) Test tubes, stands, beakers ,pipettes
- 4) Water bath, marbles

PROCEDURE:

- Different volumes of standard RNA sample (0.1 ml, 0.2 ml, 0.4 ml, 0.6 ml, 0.8 ml & 1.0 ml) and unknown sample (0.25 ml & 0.5 ml) are taken in different test tubes in duplicate.
- 2) The volume in each test tube is made to 1 ml by adding distilled water.
- 3) A blank test tube is also prepared by taking 1 ml of distilled water.
- 4) To each test tube , 1.5 ml of orcinol reagent is added and the solution is mixed thoroughly in a cyclomixer.
- 5) The test tubes covered with marbles are kept in a boiling water bath for 20 minutes and then cooled.
- 6) The optical density is then taken at 665 nm in a colorimeter/ spectrophotometer against the blank tube and then proceeded for the solution in the other tubes.

OBSERVATIONS:

Sl.	Volume of	Volume of	Amount of	Volume of	Optical	Mean
No.	Standard	distilled water	RNA (µg)	orcinol	density at	optical
	RNA taken (added to make		reagent	665 nm	density at
	ml)	1 ml (ml)		added (ml)		665 nm
1.	0.1	0.9	20			
2.	0.1	0.9	20			
3.	0.2	0.8	40			
4.	0.2	0.8	40			
5.	0.4	0.6	80	1.5 ml in all		
6.	0.4	0.6	80	test tubes		
7.	0.6	0.4	120			
8.	0.6	0.4	120			
9.	0.8	0.2	160			
10.	0.8	0.2	160			
11.	1.0	-	200			
12.	1.0	-	200			
13.	Blank	1.0	-			

TABLE: For standard RNA (200µg / ml)

TABLE: For unknown RNA sample

S1.	Volume of	Volume of	Volume of	Optical density	Mean optical
No.	unknown RNA	distilled water	orcinol reagent	at 665 nm	density at
	sample taken	added to make	added (ml)		665 nm
	(ml)	1 ml (ml)			
1.	0.25	0.75	1.5 ml in all test		
2.	0.25	0.75	tubes		
3.	0.5	0.5			
4.	0.5	0.5			

GRAPH:

CALCULATIONS:

From the standard graph, **A** O.D. \equiv **B** µg of RNA

Therefore, $1 \text{ O.D.} \equiv \mathbf{B} / \mathbf{A} \ \mu \text{g of DNA} = \mathbf{C} \ \mu \text{g of RNA}$

• For unknown sample 1 (0.25 ml), O.D. value is **D**

Since, 1 O.D. \equiv **C** µg of RNA

Therefore, **D** O. D. \equiv **C** x **D** = **E** μ g of RNA

0.25 ml of unknown sample contains $\mathbf{E} \mu g$ of RNA

Therefore, 1ml of unknown sample contains $E/0.25 = F \mu g$ of RNA

• For unknown sample 2 (0.5 ml), O.D. value is G

Since, $1 \text{ O.D.} \equiv \mathbf{C} \mu g \text{ of RNA}$

Therefore, **G** O. D. \equiv **C** x **G** = **H** µg of RNA

0.5 ml of unknown sample contains $H \mu g$ of RNA

Therefore, 1ml of unknown sample contains $H/0.5 = J \mu g$ of RNA

Amount of RNA / ml in the unknown sample = $\mathbf{F} + \mathbf{J} = \mathbf{K} \mu g$

2

RESULT: The concentration of RNA in the unknown sample is found to be $K \mu g /ml$.

DISCUSSION:

AIM: DEMONSTRATION OF GEL ELECTROPHORESIS SEPARATION OF DNA

PRINCIPLE:

Electrophoresis is a technique used in the separation of charged macromolecules in a solution by the application of an electric current. Electrophoresis was developed by Arne Tiselius and is used in medicine, biochemistry, molecular biology and immunology to separate, identify, characterize and measure the amount of protein, nucleic acid and even small cell organelles. The molecules to be separated are loaded in a gel usually made up of agarose. Agarose is a natural product obtained and purified from red seaweed. It is a polysaccharide that dissolves in water on boiling and forms a gel when cooled to 40°C. the agarose on cooling undergoes polymerization i.e. sugar polymers cross link with each other and cause the solution to form a porous gel whose pore size is determined by the concentration of the agarose used.

Tris-Acetate EDTA (TAE) and Tris- Borate EDTA (TBE) are two buffers used to transmit the electrical current to the DNA molecules current to the DNA molecules in the gel.

DNA isolate from bacterial cells may either be whole or fragmented into various sizes. To separate the different sized DNA molecules, the DNA sample are loaded at the cathode end and during electrophoresis, they move towards the anode end. Based on the molecular mass, the DNA molecules will be separated along the length of the gel.

<u>Staining</u>: DNA molecules separated by electrophoresis can be viewed after staining with Ethidium Bromide (EtBr) and exposed to UV light. EtBr is a planar molecule that intercalates between the DNA bases. It absorbs UV light and emits a bright orange colour. Hence any DNA present in the agarose gel after electrophoresis will contain EtBr and appear as an orange band in UV light. Etbr is however, carcinogenic and should be handled with gloves and disposed off carefully.

REQUIREMENTS:

Electrophoresis buffer tank, 1X TAE, water bath, hot plate, UV Transilluminator, stirring rod, micropipette, etc.

PROCEDURE:

Preparation of Agarose Gel:

- 1. Weight 0.5 g of Agarose powder and transfer it to a clean beaker.
- 2. Add 50 ml of TAE buffer to the agarose powder and heat in a microwave oven to dissolve the agarose gel.
- 3. Add about 1ml of EtBr from a stock of $20\mu g/ml$ into the molten agarose when the temperature of the agarose is just about 55°C.
- 4. Pour the agarose into the gel tray of the electrophoresis tank or template filled with combs.
- 5. Leave the gel undisturbed for some time to cool and set. Removed the combs.

Electrophoresis:

- 1. Place the agarose gel in the buffer tank with the wells towards the cathode end.
- 2. Pour TAE buffer in the tank till it just covers the gel.
- 3. Mix 25µl of the DNA with 2.5µl of gel loading buffer.
- 4. Load the DNA into the wells of the gel with the help of a micropipette.
- 5. Connect the electrodes from the electrophoresis tank to a power supply of 100V.
- 6. Leave the set up for about 1-2 hours to allow the DNA molecules to electrophorese.

UV Transillumination:

- 1. After electrophoresis, take the gel out of the buffer and place it on a UV transilluminator.
- 2. Switch on the UV transilluminator and observed for the presence of orange DNA bands.

Observation:

DNA molecules will appear in the form of bright orange bands in the agarose gel on exposure to UV light.

Precautions:

- 1. EtBr must be handle with gloves as it is carcinogenic.
- 2. One should not expose to oneself to UV.
- 3. Wash Hands after work.
- 4. EtBr must be properly disposed off.

AIM: DEMONSTRATION OF RADIAL IMMUNODIFFUSION

Principle:

The interaction between an antibody and an antigen is the main reaction in an immune response. When a soluble antigen is bound by an antibody, the antigen-antibody complex forms a precipitate and the reaction is called precipitation.

Radial Immunodiffusion, also called Mancini method, is a technique by which we can determine the interaction between an antigen and an antibody as well as find out the quantity of an antigen in a given sample. In this test, antigens are placed in a small well made in an agarose gel that contains antibodies. The antigens diffuse out radially from the wells into the agarose gel. When the diffused antigens meet the antibodies in the gel they will interact with each other. The interaction of antigens and antibodies is however defined by the specificity or ability of the antibody to identify the antigen. The antigen- antibody complex formed by the interaction can be observed as a precipitin ring around the well.

<u>Requirement</u>:

Agarose, 1X assay buffer, template, gel punch syringe, antigen samples, antiserum, clean glass slides, petri dish, cotton wool, micropipette, micropipette tips, etc.

PROCEDURE:

- 1. Weight 0.1 g of agarose powder and completely dissolve it in 10 ml of 1X assay buffer by heating the buffer. This will give a 1% agarose.
- 2. The molten agarose is allowed to cool to about 55°C.
- 3. 200µl of the antiserum is added to the agarose solution and mixed well by stirring.
- 4. The agarose solution containing the antiserum is poured onto a clean glass slides placed on a horizontal surface and then left undisturbed to allow the agarose solution to solidify into a gel.
- 5. Wells are cut in the agarose gel using a gel puncher.
- 6. $20\mu l$ of the antigen samples are added into the wells.
- 7. The gel is then placed in a petri dish containing some wet cotton wool and left undisturbed overnight at room temperature.

OSERVATION:

A precipitin ring is seen around the wells in which the antigen samples have been added.

DISCUSSION.

AIM: Estimation of glucose by anthrone reagent

PRINCIPLE:

In the anthrone assay, glucose is dehydrated by concentrated H_2SO_4 to form 5hydroxyl methyl furfural, which in turn condenses with anthrone to form a bluish-green coloured complex, the intensity of which is proportional to the amount of glucose present in the sample. The bluish-green coloured complex so formed has a maximum absorbancy at 620 nm and can be measured by using a spectrophotometer.

REQUIREMENTS:

- 1) Test tubes, test tube stands and holders
- 2) Pipettes, measuring cylinders& conical flasks
- 3) Water bath
- 4) Cyclomixer
- 5) Spectrophotometer

CHEMICALS REQUIRED:

1) Anthrone reagent: Prepared by dissolving 200 mg (0.20 g) of anthrone in 100 ml of concentrated H_2SO_4 .

- Standard glucose (200 µg/ ml): Prepared by dissolving 20 mg of glucose (0.02 g) in 100 ml of distilled water.
- 3) Unknown glucose sample
- 4) Distilled water

PROCEDURE:

- Different volumes of standard glucose sample (0.1 ml, 0.2 ml, 0.4 ml, 0.6 ml, 0.8 ml & 1.0 ml) and unknown sample (0.25 ml, 0.5 ml & 1 ml) are taken in different test tubes in duplicate.
- 2) The volume in each test tube is made to 1 ml by adding distilled water.
- 3) A blank test tube is also prepared by taking 1 ml of distilled water.
- 4) To each test tube, 4 ml of anthrone reagent is added and the solution is mixed thoroughly in a cyclomixer.
- 5) The test tubes covered with marbles are kept in a boiling water bath for 10 minutes and then cooled.
- 6) The optical density is then taken at 620 nm in a colorimeter/ spectrophotometer against the blank tube and then proceeded for the solution in the other tubes.

OBSERVATIONS:

~ 4						
SI.	Volume of	Volume of	Amount of	Volume of	Optical	Mean
No.	Standard	distilled water	glucose	anthrone	density at	optical
	glucose taken	added to make	(µg)	reagent	620 nm	density at
	(ml)	1 ml (ml)		added (ml)		620 nm
1.	0.1	0.9	20			
2.	0.1	0.9	20			
3.	0.2	0.8	40			
4.	0.2	0.8	40			
5.	0.4	0.6	80	4 ml in all		
6.	0.4	0.6	80	test tubes		
7.	0.6	0.4	120			
8.	0.6	0.4	120			
9.	0.8	0.2	160			
10.	0.8	0.2	160			
11.	1.0	-	200			
12.	1.0	-	200			
13.	Blank	1.0	-			

TABLE: For standard glucose (200µg / ml)

S1.	Volume of	Volume of	Volume of	Optical density	Mean optical
No.	unknown	distilled water	anthrone reagent	at 620 nm	density at
	glucose sample	added to make	added (ml)		620 nm
	taken (ml)	1 ml (ml)			
1.	0.25	0.75			
2.	0.25	0.75			
3.	0.5	0.5	4 ml in all test		
4.	0.5	0.5	tubes		
5.	1.0	-			
6.	1.0	-			

TABLE: For unknown glucose sample

GRAPH:

CALCULATIONS:

From the standard graph, **A** O.D. \equiv **B** µg of glucose

Therefore, $1 \text{ O.D.} \equiv \mathbf{B} / \mathbf{A} \mu g \text{ of } DNA = \mathbf{C} \mu g \text{ of glucose}$

• For unknown sample 1 (0.25 ml), O.D. value is **D**

Since, 1 O.D. \equiv **C** µg of glucose

Therefore, **D** O. D. \equiv **C** x **D** = **E** μ g of glucose

0.25 ml of unknown sample contains $\mathbf{E} \mu g$ of glucose

Therefore, 1ml of unknown sample contains $E/0.25 = F \mu g$ of glucose

• For unknown sample 2 (0.5 ml), O.D. value is G

Since, $1 \text{ O.D.} \equiv \mathbf{C} \mu g \text{ of glucose}$

Therefore, **G** O. D. \equiv **C** x **G** = **H** μ g of glucose

0.5 ml of unknown sample contains $H \mu g$ of glucose

Therefore, 1ml of unknown sample contains $H/0.5 = J \mu g$ of glucose

• For unknown sample 3 (1.0 ml), O.D. value is **K**

Since, $1 \text{ O.D.} \equiv \mathbf{C} \ \mu g \text{ of glucose}$

Therefore, **K** O. D. \equiv **C** x **K** = **L** µg of glucose

Therefore, 1ml of unknown sample contains $L \underline{\mu g}$ of glucose

Amount of glucose/ ml in the unknown sample $= \mathbf{F} + \mathbf{J} + \mathbf{L} = \mathbf{M} \mu g$

RESULT: The concentration of glucose in the unknown sample is found to be $M \mu g /ml$.

DISCUSSION:

AIM: Estimation of protein by Lowry's method

PRINCIPLE:

Lowry's method of protein estimation is the most widely used and accepted method for accurated etermination of protein concentration. In the first step of the reaction, protein binds to copper in alkaline medium and produces Cu^{2+} . In the second step, Cu^{2+} catalyses oxidation of aromatic amino acids by reducing phosphomolyb denotunstate to a blue coloured complex. The strong blue colour so formed depends on tyrosine and tryptophan content of protein and to a lesser extent cysteine and other residues in protein.

REQUIREMENTS:

- 1) Test tubes, test tube stands and holders
- 2) Pipettes, measuring cylinders& conical flasks
- 3) Cyclomixer
- 4) Spectrophotometer

CHEMICALS REQUIRED:

- **1)** Alkaline Na ₂ CO ₃: Dissolve 4 g of Na ₂ CO ₃ in 200 ml of 0.1 N NaOH (40 mg in 1L; 4 mg in 100 ml; 8 mg in 200 ml).
- 2) CuSO₄ –Na-K Tartarate Solution: Mix 1% CuSO₄.5H₂O and 2% Na-K tartarate in the ratio 1:1. This solution has to be prepared fresh.
- 3) Alkaline solution: Mix 50ml of solution 1 with 1 ml of solution 2.
- 4) Folin-ciocalteau reagent: Diluted two times with water.
- 5) Standard protein solution (200µg/ml): Prepared by dissolving 20 mg of bovine serum albumin (0.02 g) in 100 ml of distilled water.
- 6) Unknown protein sample
- 7) Distilled water

PROCEDURE:

- 1) Different volumes of standard protein sample (0.1 ml, 0.2 ml, 0.3 ml, 0.4 ml & 0.5 ml) and unknown sample (0.1,0.25 ml n & 0.5 ml) are taken in different test tubes in duplicate.
- 2) The volume in each test tube is made to 0.5 ml by adding distilled water.
- 3) A blank test tube is also prepared by taking 0.5 ml of distilled water.
- 4) To each test tube , 2.5 ml of alkaline reagent is added , the solution is mixed thoroughly in a cyclomixer and allowed to stand for 10 minutes.
- 5) After 10 minutes, 0.25 ml of Folin-ciocalteau reagent is added to each test tube, followed by mixing in a cyclomixer.
- 6) The test tubes are then kept at room temperature for 30 minutes .
- 7) The optical density is measured at 630 nm in a colorimeter/ spectrophotometer against the blank tube and then proceeded for the solution in the other tubes.

OBSERVATIONS:

Sl. No.	Volume of Standard protein taken (ml)	Volume of distilled water added to make 0.5 ml (ml)	Amount of protein (µg)	Volume of alkaline reagent added (ml)	Volume of Folin- ciocalteau reagent added (ml)	Optical density at 620 nm	Mean optical density at 620 nm
1.	0.1	0.4	20				
2.	0.1	0.4	20				
3.	0.2	0.3	40		0.25 ml		
4.	0.2	0.3	40				
5.	0.3	0.2	60	2.5 ml in			
6.	0.3	0.2	60	all test	in all test		
7.	0.4	0.1	80	tubes	tubes		
8.	0.4	0.1	80				
9.	0.5	-	100				
10.	0.5	-	100				
11.	Blank	0.5	-				

TABLE: For standard protein (200µg / ml)

Sl.	Volume of	Volume of	Volume of	Volume of	Optical	Mean
No.	unknown	distilled	alkaline	Folin-	density at	optical
	protein	water added	reagent added	ciocalteau	620 nm	density at
	sample taken	to make 0.5	(ml)	reagent		620 nm
	(ml)	ml (ml)		added (ml		
)		
1.	0.1	0.4				
2.	0.1	0.4				
3.	0.25	0.25	2.5 ml in all	0.25 ml in		
4.	0.25	0.25	test tubes	all test tubes		
5.	0.5	-]			
6.	0.5	-				

TABLE: For unknown protein sample

GRAPH:

CALCULATIONS:

From the standard graph, **A** O.D. \equiv **B** µg of protein

Therefore, $1 \text{ O.D.} \equiv \mathbf{B} / \mathbf{A} \mu g \text{ of } DNA = \mathbf{C} \mu g \text{ of protein}$

• For unknown sample 1 (0.1 ml), O.D. value is **D**

Since, 1 O.D. \equiv **C** µg of protein

Therefore, **D** O. D. \equiv **C** x **D** = **E** μ g of protein

0.1 ml of unknown sample contains $\mathbf{E} \ \mu g$ of protein

Therefore, 1ml of unknown sample contains $E/0.1 = F \mu g$ of protein

• For unknown sample 2 (0.25 ml), O.D. value is G

Since, $1 \text{ O.D.} \equiv \mathbf{C} \mu g \text{ of protein}$

Therefore, **G** O. D. \equiv **C** x **G** = **H** µg of protein

0.25 ml of unknown sample contains $H \mu g$ of protein

Therefore, 1ml of unknown sample contains $H/0.25 = J \mu g$ of protein

• For unknown sample 3 (0.5ml), O.D. value is **K**

Since, $1 \text{ O.D.} \equiv \mathbf{C} \mu g \text{ of protein}$

Therefore, **K** O. D. \equiv **C** x **K** = **L** µg of protein

0.5 ml of unknown sample contains $L \mu g$ of protein

Therefore, 1ml of unknown sample contains $L / 0.5 = M \mu g$ of protein

Amount of protein/ ml in the unknown sample $\underline{= F + J + M} = N \ \mu g$

RESULT: The concentration of protein in the unknown sample is found to be $N \mu g /ml$.

DISCUSSION:

AIM: Detection and separation of amino acids by paper chromatography

INTRODUCTION:

Chromatography in modern era forms an important tool for detection of aminoacids, proteins, carbohydrates, plant pigments, etc.. The term 'chromatography' (Greek- to unite in colours) was coined by Mikhael Tsvet, a Russian scientist in 1900. Later Neubaur (1936), separated neutral amino acids and observed that the partition coefficient of acetylated amino acids between water and immiscible organic solvents differed for various amino acids.

PRINCIPLE:

The method of paper chromatography consists in applying a small drop of solution containing substances to be separated to a strip of filter paper, a short distance from one end,. The drop is allowed to dry and the end of the paper nearest to the spot is placed in the developing solutionwater containg organic solvents so that the solvent flows past the spot by capillary action along the length of the paper.

In paper chromatography, separation of the components of a mixture is a function of their affinity for a stationary solid phase (paper) in a moving liquid phase (solvent). The main factor for separation of compounds is the coefficient of the distribution of compounds governed by their tendency to distribute themselves between the solvent (liquid phase) and adsorption on the solid phase. Some compounds in a mixture travel almost as far as the solvent does; some stay much closer to the base line. The distance travelled relative to the solvent is a constant for a particular compound –as far as everything is kept constant- type of paper and the exact composition of the solvent, for example.

The distance travelled by the compound relative to the solvent is called the $R_{\rm f}$ value. For each compound it can be worked out using the formula:

 $R_f =$ <u>Distance travelled by the compound/solute</u> Distance travelled by the solvent

REQUIREMENTS:

- 1) Chromatography chamber
- 2) Butanol, glacial acetic acid and water in the ratio 4:1:1
- 3) Whatman paper
- 4) 0.1% Ninhydrin
- 5) Capillary tubes
- 6) Samples of amino acids to be separated
- 7) Hair dryer

PROCEDURE:

- 1) All glass ware is cleaned before starting the experiment and dried in the oven.
- 2) The rectangular glass jar is taken and is filled with the solvent i.e., butanol, glacial acetic acid and water in the ratio 4:1:1.
- 3) The whatman chromatogaraphy paper is cut into strips and is length is taken according to the height of the glass jar.
- 4) A line is drawn about 1 inch from the bottom of the paper and three small circles are marked on the line separated separated by a little distance from each other.
- 5) A small amount of sample is applied to the marked spots(different samples on different spots) on the paper (chromatogram).
- 6) The paper is hung in a string fastened to the edge of the glass jar.
- 7) The bottom edge of the paper is dipped in the solvent keeping the pencil marks above the solvent.
- 8) The glass chamber is then closed with a glass lid, and the chromatogram is left to develop for 2 hours.
- 9) After 2 hours the chromatogram is removed and the position of the solvent is marked. the chromatogram is then allowed to dry after which it is sprayed uniformly with 0.1% ninhydrin and dried with a hair dryer at 90°C for 2-5 minutes.
- 10) The spots formed are circled with a pencil and the R_f values are calculated and compared with standard values of amino acids.

OBSERVATIONS:

According to Morris (1964), R_f value is the relation of the distance travelled by a compound to that by the solvent front. R_f value is defined as the linear rate of movement of the solute zone to the linear rate of movement of the solvent.

 $R_{f} = \frac{Distance\ travelled\ by\ the\ compound/solute\ from\ origin}{Distance\ travelled\ by\ the\ solvent\ from\ origin}$

Thus, in the experiment ,in two hours, distance travelled by the compound in the sample and that travelled by the solvent is measured and the R_f is calculated and compared with standard values to find out the amino acid present in the sample.

TABLE: Standard R_f values of amino acids

Amino Acid	R _f value

RESULTS:

DISCUSSION:

Aim : Estimation of Ascorbic acid content of lemon.

Requirements:

Lemon, burette, conical flask, measuring cylinder, pipette, balance, weight box, beaker, **Reagents**: 1.Standard Ascorbic acid solution (0.5mg/ml; prepare just before use) 2. 2,6-dicholorophenolindophenol (0.25% i.e.,50mg in 100ml.)

Principle: Ascorbic acid is a strong reducing agent .It can reduce the blue coloured dye 2,6dichlorophenolindophenol to a colourless compound.In this reaction ascorbic acid gets oxidized to dehydroascorbic acid.



Procedure:

1. 1ml. of the dye 2,6-dichlorophenolindophenol was taken in a conical flask.

2.Standard ascorbic acid was taken in a burette.The dye was titrated against standard ascorbic acid. The end point indicated by the disappearance of the blue colour.

3. The same was repeated three times.

4. The weight of the lemon was taken and the juice was extracted and filtered.

5. The volume of the lemon juice filtrate was measured.

6.The lemon juice was taken in a burette and titrated against 1ml. of of the dye.The end point was marked by the appearance of lemon yellow colour.

7. The step number 6 is repeated three times and the mean was taken.

Calculation:

Strength of ascorbic acid = 0.5mg/ml.

A ml. of standard ascorbic acid is required to neutralized 1ml. of dye

1ml. of standard ascorbic acid is =0.5mg ascorbic acid

.. A ml. of standard ascorbic acid =0.5x Aml. =0.5x A mg. of Ascorbic acid

For lemon:

Weight of the lemon =**B** gms.

Volume of lemon juice= \mathbf{C} ml

D ml. of lemon juice can neutralize 1 ml. of dye

- . D ml. of lemon juice = 0.5x A mg. ascorbic acid
- .. 1ml.of lemon juice can neutralize =0.5Amg. /D mg of Ascorbic acid
 - . . C ml of lemon juice can neutralize=0.5xAmg x C /D mg of ascorbic acid

B gm. of lemon = 0.5x **A** mg.x **C/D** mg of ascorbic acid

... 1 gm. of lemon = $\underline{0.5 \times A \mod x C / D}$ mg. of ascorbic acid

B

Result : 1 gm of lemon contains $0.5 \times A \mod x C / D$ mg. of ascorbic acid.

Discussion:

B

0.5xaml

AIM: QUALITATIVE TESTS FOR CARBOHYDRATES, PROTEINS & FATS

A: TESTS FOR CARBOHYDRATES

Apparatus and Glassware: Test tubes, test tube holder, test tube stand, spirit lamp.

Reagents required:

- 1. Fehling's reagent.
- 2. Benedict's reagent
- 3. Saturated solution of picric acid
- 4. 40% sodium hydroxide solution.

TEST SOLUTIONS: 1.1% Glucose

2.1% Starch

General Principle: Carbohydrates are group of compounds containing C, H, O which generally fit in the empirical formula (CH₂O) n. These include monosaccharides, disaccharides and polysaccharides.

All monosaccharides and some disaccharides including maltose and lactose are reducing sugars, having <u>aldehyde</u> or <u>ketone</u> gp. possessing reducing property.

The first test solution is of 1% glucose. Glucose has the aldehyde (CHO) gp. which has the reducing power.

Two common tests for reducing sugars are Fehling's test & Benedict's test. Both the tests make use of the ability of glucose to reduce copper from a valency of 2 to 1. Both tests involve use of an alkaline solution of copper (II) sulphate ($CuSO_4$) which is reduced to insoluble copper (I) oxide (Cu_2O).

The reaction is as follows:

$$CuSO_4 + 2NaOH \rightarrow Cu (OH)_2 + Na_2SO4 \qquad 2 Cu (OH)_2 \underline{reduction} Cu_2O + 2H_2O + 2H_$$

red ppt of Cuprous oxide.

Reduction of blue CuSO₄ solution is accompanied with liberation of oxygen & sugar itself undergoes oxidation

Fehling's Test: This test is not as convenient as Benedicts test because Fehling's solution A & B have to be kept separate and mixed just prior to the test.

Benedict's Test: Benedict's method modifies the Fehling's test to produce a single solution which is more convenient to use and also more stable. The final precipitate would appear green ntto yellow to orange to red – brown with increasing amounts of reducing sugar.

Picric acid Test: Saturated solution of Picric acid is used. Glucose acts as reducing agent in a weakly acid solution.

Picric acid is reduced to **Picramic acid** with the formation of a red precipitate

TEST SOLUTION: 0.2% STARCH

General Principle:Starch is a storage polysaccharide in plants.Starch is a homopolysaccharide consisting of many no. of D glucopyranose units.It does not contain a free aldehyde (CHO) or keto (C=O) group thus is a non reducing substance.It does not show colour reactions with reagents like Benedicts and Fehlings exhibited by reducing sugars.When starch is heated with conc.HCl, it is hydrolysed to maltose a reducing sugar which is able to form coloured ppt. with the above mentioned reagents.

Starch forms a blue coloured complex with Iodine.

found to conatin a test solution was the analysis systematic ftersA :clusionCon polysacchrdride

Sl No	Experiments	Observations	Inference
1	Fehling's test: 5ml of fehling solution + 1ml (5drops) of test solution are mixed and boiled	Brick red precipitate of cuprous oxide (Cu2O) is formed	Presence of reducing sugar

2.	Benedict's test: 2ml of benedict's reagent + 5 drops (1ml) of test solution and boiled vigorously.	A red precipitate is formed	Reducing sugar is confirmed.
3	Picric Acid test 2ml.test solution + 1 ml of picric acid and 1 ml of 40% NaOH solution & boiled for 1 – 2 mins	A red colour appears in the solution	Reducing sugar is present

Discussion:

The carbohydrates are the most abundant class of biological molecules. In plants they are produced by photosynthesis. In animal cells, carbohydrates in the form of glucose and glycogen serve as the chief source of energy for vital activities of the body. Their caloric value is 4 K.cal/g for all organisms .Some carbohydrates have highly specific functions, e.g., ribose in the nucleoproteins, galactose in certain lipids and lactose in milk. Some carbohydrates perform structural role such as cellulose, chitin etc.. Carbohydrates play a key role in the metabolism of amino acids and fatty acids. Starch is a storage polysaccharide in plants. It is a homo polysaccharide, containing -glucopyranose as its repeating units. It is water insoluble amorphous substance. It is not sweet in taste. It is a non reducing sugar.

B:TESTS FOR PROTEINS:-

Reagents required:

- 1. 1% CuSO₄
- 2. 40% NaOH
- 3. 1% Ninhydrin solution (freshly prepared)
- 4. Conc. HNO3

TEST SOLUTION: 5% solution of Egg albumin

General principle: Proteins are essential, as nutrients for all organisms. Proteins serve as structural and functional components in the living body. They are composed of about 20 different amino acids. Specificity of a protein depends upon the sequence and number of amino acid units. Amino acids are joined in protein molecules by peptide bonds. The tests mentioned here are colour reactions of proteins. These colour reactions are due to the reaction between one or more constituent radicals or groups of the complex protein molecules and reagent/ or reagents used in any given test.

1. The Ninhydrin Reaction: Ninhydrin (triketohydrindene hydrate), a powerful oxidizing agent, reacts with all amino acids between pH4 and 8 to give a purple coloured compound. This reaction is very sensitive.



2. The Xanthoproteic Reaction: Amino acids which contains an aromatic nucleus form yellow nitro derivatives on heating with conc. HNO₃. On addition of alkali salts are formed which are are orange in colour.

3. The Biuret Test: Alkaline copper sulphate reacts with compounds containing two or more peptide bonds to give a violet colored complex. The depth of the color obtained is a measure of the number of peptide bonds present in the protein. The name of the test comes from the compound <u>Biuret</u> which gives a typical positive reaction.

Biuret CONH₂

NH

 CONH_2

Conclusion: After systematic analysis the test solution was found to contain a protein.

Discussion:

The proteins are the most abundant intracellular macromolecules constituting about 50% of the dry weight of an organism.Some proteins function as structural elements(collagen , hair, elastin, wool etc.),some as hormones(insulin, oxytocin, glucagon etc.), some as oxygen carriers(haemoglobin, myoglobin),some as carrier molecules in blood plasma(albumin ,globulin etc.), some as defense molecules(antibodies),and as **enzymes** in all the biochemical reactions of the body.Fibrinogen is a soluble protein of blood which takes part in blood clotting .

Proteins from animal source are complete proteins containing all essential amino acids needed during early development, pregnancy ,lactation and injuries. These proteins are readily absorbed and utilized by the body for eg. meat ,poultry ,cheese ,milk and one plant source protein, soy bean. Plant source proteins like cereals ,pulses,beans,dry fruits etc. ,are incomplete proteins lacking some essential amino acids. Proteins also can be used as source of energy. Each gram . of protein contains

Sl No	Experiments	Observations	Inference
1	Ninhydrin Test: 1 ml of test solution + 5 drops of 1% Ninhydrin solution and boiled for 2 mins.	A purple colour appears in the solution	Amino acids are present .May be a protein
2	Xanthoproteic Test: 1 ml of test solution + 1 ml of conc. HNO ₃ .Heat on a direct flame.Cool and add sufficient 40% NaOH to make the solution strongly alkaline.	A yellow colour appears in the acidic solution which changes to bright orange with alkali.	Proteins are present
3	Biuret Test 1 ml. of 1% copper sulphate + 2 ml of the test solution + 2 ml of 40% NaOH and mixed thoroughly.	A violet colour appears in the solution	Proteins are present

C: TEST FOR FATS/LIPIDS/OILS

Reagents required:

- 1. 10% KOH in absolute alcohol.
- 2. Potassium bisulphate
- 3. Dehydrated alcohol, Acetone, Chloroform
- 4. Water
- 5. Sodium Chloride.

TEST SOLUTION: Olive Oil

General principle: The lipids are heterogenous group of organic substances present in plant and animal kingdom and chemically they are various types of esters of different alcohols. Lipids are insoluble in water but soluble in fat solvents such as ether ,choloroform, benzene, acetone etc. They are stored as a concentrated food reserve in adipose tissues of animals. Lipid serve as the structural constituents of cell membrane and cell wall. When fats and oils are heated with alkali (hydrolysis), fatty acids and glycerol are liberated, in a process called saponification .

Solubility Test: Solubility of the fats/ oils is tested by mixing 2ml. Test solution with four different solvents- Water, Dehydrated Alcohol, Acetone and Chloroform, in separate test tubes and shaken vigorously.Fats are completely soluble in Acetone and Chloroform solvents, partially soluble in dehydrated Alcohol and insoluble in water

Saponification Test: When fats/ oils are heated with alkali, the excess alkali

(10% KOH) present reacts with the liberated fatty acids to form the sodium or potassium salts which gives the solution a characteristic soapy appearance.

Soaps are soluble in water but are precipitated on the addition of excess sodium chloride(NaCl).

Sl No	Experiment	Observation	Inference
1	Solubility Test : Four test tubes are taken and marked A, B, C & D and following tests are performed. Test Tube A: - 1 ml test solution & 5 ml of water is mixed with vigorous shaking	Insoluble	fat/oil may be present
	Test Tube B: 1 ml of test solution is mixed with 5 ml of Chloroform	Soluble(miscible)	fat/oil may be present
	Test Tube C: 1 ml of test solution is mixed with 5 ml of dehydrated alcohol.	Partially Soluble	fat/oil may be present
	Test Tube D: 1 ml of test solution is mixed with 5 ml of acetone.	Soluble(miscible)	fat/oil confirmed in the Test solution

	Acrolein Test:		
2	In a test tube. 5cm deep layer of	A characteristic	Glycerol is
	KHSO ₄ (potassium bisulphate) is	pungent odour of	present in the
	taken. Now about 5 drops of the test	acrolein is produced.	test solution.
	solution is added & further covered		May be an oil or
	with KHSO4 and heated slowly		fat
2			
3	Saponification Test:		
	1 ml test solution + 5ml. alcoholic		
	KOH and boil carefully for 1min. A		
	soapy solution is formed. Cool the		
	solution and add excess of NaCl,	A precipitate (soap) is	
	boiled and cooled.	formed	An oil or fat is present
	I		

Acrolein Test: When glycerin is heated with potassium bisulphate(KHSO4), dehydration occurs and the aldehyde acrolein is formed, which has a characteristic/ pungent odour. This test is given by glycerol which is either free or combined as an ester.



Conclusion: After systematic analysis the test solution was found to be an oil or fat.

Discussion: The lipids include fats ,oil, waxes and related compounds. In the body lipids serve as an efficient and quick source of energy in times of starvation and fasting .Caloric value of

lipid is 9K.cal/gm. It serves as an insulating material in the subcutaneous tissues and around certain organs like brain and nervous tissue. Mono and poly unsaturated fats present in seed oils, olive oil and nuts provide essential fatty acids (EFA) like

Standard Operating Procedure

Department of Zoology

St. Mary's College, Shillong

Cytology, Genetics and Molecular Biology

List of practicals:

1. Preparation of mitosis slides from Onion root tips.

2. Preparation of meiosis slides from grasshopper testis.

3. Study of sex chromatin(Barr body) from human buccal epithelial cells.

4. Study of Polytene chromosomes from the salivary glands of Chironemus larvae.

5.Determination and study of multiple alleles(ABO blood groups) and Rh factor in humans.

6. Calculation of chiasma frequency and coefficient of terminalization from slides prepared from grasshopper testis.

7. Preparation of human karyotype of normal male and female and Klinefelter/Turner's/Down's syndrome.

8. Study of phenotypic variation in natural population.

9. Study of chromosome types from slides/photographs.

1. Aim of Experiment: <u>Preparation of mitosis slide from Onion root tips(2n=16)</u>

Theory: Mitosis is an equational cell division observed in somatic cells. Mitosis ensures that each

daughter cell receives one copy of each of the replicated chromosomes. During the process of mitosis,

the chromosomes pass through several stages known as prophase, metaphase, anaphase and

telophase. The actual division of the cytoplasm is called cytokinesis and occurs during telophase.

The meristematic cell at the root tips of onion are actively dividing and many cells are in the stages of mitosis. Therefore, the root tips makes an excellent tissue to study the stages of mitotic cell division.

Materials required: Onions rooted in water, glass slide, cover glass, acetocarmine stain, blotting paper, watch glass, scissor, forceps, and compound microscope.

Procedure:

1. Obtain an onion bulb that has been rooted in water.

2. Cut two or three root tips of about 2mm in size from the base of the bulb.

3. Place the root tips in a watch glass. Add a few drops of 1M HCl and let the root tips stand in the solution for 10 minutes.

4. Remove one root tip from the HCl solution and place on a slide by using forcep.

5. Add two drops of acetocarmine stain and leave for 10 minutes.

6. Apply a cover glass on the root tip and remove excess stain with blotting paper.

7. Squash by a firm vertical thumb pressure.

8. Mount the slide on the compound microscope.

9. Use the low power objective (10X) to scan for the dividing cells and then use the 40X power objective to study the mitotic stages in individual cells.

10. Identify the different mitotic stages with reasons.

2. Aim of Experiment : <u>Preparation of meiosis slide from Grasshopper testis(2n=21)</u>

Theory: Meiosis is a cell division that occurs in the germ cells of sexually reproducing eukaryotic organisms. Meiosis reduces the chromosome number by half producing haploid daughter cells which function as gametes. Meiosis is completed in two stages, meiosis I and meiosis II. Meiosis I is reductional while meiosis II is equational. At the end of meiosis four haploid daughter cells are produced.

Material required:Follicles of Grasshopper testis, glass slide, cover glass, acetocarmine stain, blotting paper, watch glass, forceps, and compound microscope.

Procedure:

1. Place a small drop of acetocarmine stain in the middle of a clean glass slide.

2. Take three or four grasshopper testis follicles from the fixative and place them in the stain. Leave for 10 minutes.

3. Cover the slide with a clean cover glass.

4. Heat the slide gently over the flame of a spirit lamp to flatten and spread the chromosomes. The stain must not boil.

5. Blot out excess stain with blotting paper.

6. Squash by a firm vertical pressure.

7. Identify the different meiotic stages with reasons.

3. Aim of Experiment: <u>Study of sex chromatin (Barr body) from human buccal epithelial</u> <u>cells</u>

Theory: Murray L.Barr and Edward G Bertram in 1949 first observed a darkly stained body lying against the nuclear envelop of interphase cells in the buccal epithelial cells of human females. This body is named as sex chromatin or barr body. In human, normal females have two X chromosomes while normal males have one X chromosome and one Y chromosome. In comparison to the X chromosomes, the Y chromosome contain very few genes. In this respect, the females thus have two sets of genes in the X chromosomes in contrast to the one set in males. Consequently a dosage compensation mechanism exists to bring about an equality in the quantity of the X coded gene products in the two sexes. This mechanism is achieved by inactivating one of the X chromosome in females by heterochromatinization. Heterochromatinization of the X chromosome. Germ cells lack barr body. Therefore, the number of barr body is always one less than the number of X chromosome. There is no barr body in normal males (XY) and in Turner syndrome (45XO), one is present in Klinefelter syndrome (47,XXY), one in normal females(XX), two in metafemales (47,XXX) and so on.

Materials required: Sterilized spatula, acetocarmine stain, glass slide, cover glass, glycerine and compound microscope.

Procedure:

- 1. Wash the mouth with water.
- 2. Take a blunt sterilized spatula and scrape the inner side of the cheek.
- 3. Make a thin smear of the scraped material on a clean glass slide.
- 4. Allow to dry at room temperature.
- 5. Add few drops of acetocarmine stain and leave for 5 minutes.
- 6. Mount the material in glycerine.
- 7. Study the slide under a compound microscope.

4. Aim of Experiment: <u>Study of Polytene chromosomes from the salivary glands of</u> <u>Chironemus larvae(2n=4)</u>

Theory: Polytene chromosomes are giant chromosomes present in the salivary glands of dipteran flies. These giant chromosomes were first observed by E.G.Balbiani. Each polytene chromosome reveals a linear series of alternating dark and light bands. These chromosomes also show diffuse areas called chromosome puffs or balbiani rings. Puffs are regions of active transcription as evident from the presence of messenger RNAs. The unusual characteristics of polytene chromosomes are its large size, they show pairing despite their presence in somatic cells and the paired homologs undergo may rounds of replication but without apparent strand separation, a process known as endomitosis.

Materials required: *Chironemus* larvae, glass slide, cover glass, acetocarmine stain, forceps, blotting paper and compound microscope.

Procedure:

- 1. The larva is placed on a slide and its salivary glands are removed.
- 2. Clean water is dropped to remove all debris and remains of the larva.
- 3. Add a few drops of acetocarmine stain on the salivary glands.
- 4. Leave to stand for 10 minutes.
- 5. Place a cover glass and blot out excess stain using blotting paper.
- 6. Squash by a firm vertical pressure.
- 7. Study the preparation under compound microscope.

5. Aim of Experiment: <u>Calculation of chiasma frequency and terminalization coefficient</u> from meiotic slides of grasshopper testis

Theory: meiosis is a type of cell division that reduces the chromosome number of a parent germ cell by half to produce four haploid gamete cells. The process of meiosis is divided into meiosis I and meiosis II and both meiotic divisions have multiple phases. Meiosis I or the first meiotic division begins with prophase I followed by metaphase I, anaphase I and telophase I. Prophase I is again divided into five substages namely, leptotene, zygotene, pachtene, diplotene and diakinesis. In zygotene the chromosomes appear as long slender threads. Homologous chromosomes then begin pairing or synapsis during pachytene. Each chromosome of a homologous pair (bivalent) is made up of two chromatids. At diplotene the homologues start separating but appear to be joined at chiasmata which are the pints of crossing over between non-sister chromatids. A chiasma is a point where the non-sister chromatids of a homologous pair exchange of genetic material during crossing over. The movement of chiasmata to the ends of a bivalent is known as terminalization. It begind at diplotene and ends at diakinesis.

Chaisma counting has been regarded as the most straightforward method of scoring the total number of crossing over events in the genome. The calculation of chaisma frequency is considered as a good estimate of the level of genetic recombination occurring in the genome of an organism. Terminalization coefficient shows the degree of terminalization of chiasmata. At diakinesis the chiasmata are fully terminalized and remain together by their extreme terminal chiasma.

Tabulation

Sl	Stage	Total no. of	Total	Chiasma	Total	Terminalization
No.		bivalents (x)	Chiasma (y)	frequency	terminalized	coefficient
				(y/x)	chiasma (a)	(a/y)

6. Aim of Experiment: <u>Determination and study of multiple alleles (ABO blood groups)</u> and Rh factor.

Apparatus and glassware: Glass slides, lancet, cotton wool, pins.

Reagents required : Anti-A, Anti-B, Anti-D sera, alcohol.

Material: Blood

General Principle: Sera containing antibodies (antisera) are used to determine the blood type of persons. The blood is tested for the agglutinogen (antigen) with known Anti-A, Anti-B and Anti-

D sera that contains antibodies which would cause red cells to agglutinate. Antiserum A cause cell type A to clump and Antiserum B cause cell type B to clump. If the blood does not agglutinate upon adding either Antisera A or Antisera B, the individual is said to belong to O because their red blood cells lack the agglutinogen A and B. Such individuals are known as "Universal donors". Individuals of blood type AB have both antigen A as well as antigen B on their red blood cells. When Antiserum A and Antiserum B is added to the blood of such persons, clumping is seen to occur in both cases. Individuals of group AB are called "Universal recipients" because their sera lack the agglutinins (antibodies a and b) and they can receive blood of all other groups.

Anti D sera contains antibodies against a particular factor known as Rh factor. This factor was first discovered in Rhesus macaque. Clumping of red cell blood upon adding anti D shows that the individual is Rh positive. No clumping indicates that the individual is Rh negative, which means that the individual lacks the Rh factor.

Experimental Procedure:

1. A clean glass slide is taken and divided into three spaces using a glass marking pencil.

2. A new sterilized lancet is taken and the finger previously cleaned with alcohol is pricked by the lancet.

3. Blood is dropped in the centre of each of the three marked spaces of the slide.

4. One drop each of anti-A, anti-B and anti-D is added respectively to each drop of blood.

5. The serum and the blood is mixed with the help of separate pins.

6. The slide is placed undisturbed at room temperature for 30 minutes and then examined for agglutination.

Anti-A	Anti-B	Anti-D	Blood group
No agglutination	No agglutination	Agglutination	O positive
Agglutination	No agglutination	Agglutination	A positive
No agglutination	Agglutination	Agglutination	B positive
Agglutination	Agglutination	Agglutination	AB positive

Results/ Observations :

Anti-A	Anti-B	Anti-D	Blood group

Discussion: The ABO blood group system discovered by Karl Landsteiner in 1900 is a classic example of multiple allele in human beings. Landsteiner recognized the A,B, AB and O blood group in man, on the basis of the presence or absence of A and B antigens which are located on the surface of the RBCs. The ABO system is remarkable because antibodies can be present without prior exposure to the antigen. Such antibodies are present in the blood plasma.

Thus people with a particular ABO antigen on their RBCs will have in the serum, the antibody against the other antigen. Serum is the blood plasma where the clotting factors such as fibrinogen have removed. Type A persons have antigen A on their RBCs and anti-b antibody in their serum, type B have antigen B on their RBCs and anti-a antibody in their serum. Type O persons do not have either antigen A or antigen B but have both antibodies, anti-a and anti-b in their serum. Type AB has both antigens A and B but lack either antibody in their serum.

The four blood type A, B, AB and O phenotype are produced by an I gene that can occur in three allelic forms. The designation I stand for isoagglutinogen which is another term for antigen. I^A and I^B alleles are responsible for the production of respective A and B antigens, 'I ' is an allele that does not produce neither of the antigens. The 'i 'allele and its phenotype are recessive. The presence of the I ^A and I^B allele provides a good example of co-dominance as the heterozygotes (I^AI^B) exhibits the phenotype of both the alleles

Blood groups	Genoytype	Antigen on	Antibodies in	Can donate	Can receive
		RBC	plasma	blood to	blood from
				groups	groups
А	I ^A I ^A or I ^A i	А	Anti- b	A, AB	A,O
В	I ^B I ^B or I ^B i	В	Anti- a	B, AB	B, O
AB	$I^A I^B$	A, B	None	AB	A,B, AB, O
					(universal
					recipient)
0	Ii	None	Anti-a, Anti-	A,B, AB, O	O (universal
			b.		donor)

The knowledge of blood groups paved the way for blood transfusion to be carried out safely. Blood transfusion between incompatible blood groups can have fatal consequence as mixing of blood lead to blood clumping or agglutination which can result in the blockage of the capillaries.

7. Aim of Experiment: Preparation of human karyotype of normal male and female

Theory: Karyotype is the phenotypic appearance of the entire chromosome complement of a species. It represents all the the chromosome types based on their morphology in the total complement. The number and the morphology of each chromosome pair is normally constant and characteristic for a species. A karyogram is the actual representation of the karyotype performed from a microphotograph. The criteria needed to prepare karyotype are: (i) the total number of chromosomes in the cell (ii) measurement of the length of each chromosome including the individual length of short arm and long arm (iii) localization of the position of the primary constriction that is the centromere and (iv) localization of the secondary constriction.

Procedure:

1. A well scattered microphotograph of metaphase plate is selected.

2. The length of short arm of each chromosome is measured.

3. The length of long arm of each chromosome is also determined.

4. The whole length of the chromosomes is the calculated by adding the short arm length and long arm length

5. Centromeric index i.e. value is calculated:

'i' = short arm length/whole length x 100

6. Chromosomes are next classified into the following groups on the basis of the location of the centromere:

On the basis of centromeric index, chromosomes are classified into

(i).Metacentric chromosomes: When the location of the centromere is near the the two arms are almost equal.

(ii). Submetacentric. Chromosomes: They have noticeably unequal arm lengths.

(iii).Acrocentric chromosome: They have one long arm and one very short arm.

(iv).Telocentric chromosomes: They have their centromere adjacent to one telomere.

'i' values	Location of centromere	Nomenclature
> 47.5 - 50	Median	Μ
> 37.5 - 47.5	Median region	Μ
> 25.0 - 37.5	Submedian region	Sm
> 12.5 - 25.0	Subterminal region	St
> 2.5 - 12.5	Terminal region	Т
> 0.0 - 2.5	Terminal	Т
Karytotype table

Chromosom e type	Chromosom e pair	Chromosom e number on the plate	Short arm lengt h	Long arm lengt h	Total lengt h	Centromeri c index (i value)	Type of constrictio n

8. Aim of Experiment: <u>Study of phenotypic variation in natural population.</u>

Principle:

No Two individuals are exactly alike except for identical twins. A phenotype is any observable characteristic or trait of an organism such as its morphology, development, biochemical or physiological properties and behavior. Phenotype result from the expression of an organism's genes as well as the influence of environmental factor^s and the interaction between the two. Individuals within population show variation in traits. These heritable differences among the individual is because of the cumulative effect of the genetic information passed on from parents to offspring (via molecules of DNA) as well as by the influence of the environmental conditions. This study, therefore, aims to observe the listed phenotypic traits among the student of the class.

Method:The students of the class are taken a natural population. Traits are listed and asurvey ismade and recorded.

Observation: The following traits have been studied:

Name, Age, Community, Blood Group, Ear lobe, eye colour, Finger print, Height, Hair etc.

<u>Discussion:</u> The ultimate factor determining the phenotypic characteristic of the individual is the genotype. The term variation describes the difference in characteristics shown by organisms belonging to the same natural population or species. Variation which produce individuals showing clear-cut differences with no intermediates between them, such as blood groups in humans are known as discontinuous variations. Height, weight, eye colour, ear lobe, complexion,

etc which show a complete graduation from one extreme to the other without any break are known as continuous variations.

Studies have shown that both heredity (nature) and environmental factors (nurture) interact to varying degrees in different individuals to influence the continuous phenotypic variation.

Phenotypic variation is a fundamental pre-requisite for evolution by natural selection. It is the living organism as a whole that contributes (or not) to the next generation, so natural selection affects the genetic structure of a population indirectly via the contribution of phenotypes. Without phenotypic variation there would be no evolution by natural selection.

9. Aim of Experiment: Study of chromosome types from slides/ Photographs

Introduction:

A chromosome is an organized structure of DNA and protein. The word' chromosome' comes from the Greek word 'chroma = colour' and 'soma=body', due to their property of being strongly stained by particular dyes.

Chromosomes vary widely between different organisms. Typically, eukaryotic cells have large linear chromosomes and prokaryotic cells have smaller circular chromosomes. In eukaryotes, nuclear chromosomes are packaged by proteins into a condensed structure called chromatin. Each chromosome can be longitudinally split into two chromatids, each containing a highly coiled, fibre like chromonema. Centromeres and telomeres are two essential feature of all eukaryotic chromosomes.

Centromeres are those condensed regions within the chromosome that are responsible for the accurate segregation of the chromosome during mitosis and meiosis. Telomeres are the region of the DNA at the end of the linear eukaryotic chromosome and provide terminal stability to the chromosome and ensure its survival.

Types of chromosomes based on the position of the centromere:

Each chromosome has a constriction point called the centromere, which divides the chromosomeinto two sections or 'arms'. The short arm of the chromosome is labeled the 'p arm'. The long arm of the chromosome is labeled the 'q arm'. The location of the centromere on each chromosome gives the chromosome its characteristic shape and can be use to help describe the location of specific genes.

Metaphase chromosomes differ from one another in size and shape. However, the relative position of the centromere is constant, which means that the ratio of the length of the two arms is constant for each chromosome. This ratio is an important parameter for chromosome

identification. Also, the ratio of length of the two arms allows classification of chromosomes into several morphologic types.

(i). Metacentric: If the centromere is near the middle of the chromosome, the two arms of the chromosome are nearly equal. The chromosome appears V-shaped during anaphasic movement. Such a chromosome is called metecentric.

(ii).Sub-metacentric: If the centromere is situated at some distance away from the middle, one arm will be shorter than the other. Such a chromosome appear L-shaped during anaphasic movement and is called sub-metacentric.

(iii).Acrocentric: When the centromere is situated near the end of the chromosome (subterminal) it appears rod-shaped and is called acrocentric.

(iv). Telocentric: If the centromere is truly terminal i.e. situated at the tip of the chromosome, the chromosome is said to be telocentric. Telocentric is very rare.

Standard Operating Procedures

Department of Zoology, St. Mary's College, Shillong

PHYSIOLOGY

List of Practicals:

- 1. Preparation of haemin crystals from human blood.
- 2. Determination of clotting time of human blood.
- 3. Estimation of haemoglobin content of human blood.
- 4. To enumerate the RBC count of human blood.
- 5. To enumerate the total WBC count of human blood.
- 6. To study the effect of pH on the activity of salivary amylase on starch.
- 7. Estimation of oxygen consumption in fish by Winkler's method.

1. Aim: Preparation of Haemin crystals

Apparatus and Glassware: Microscope; test tube; slide and cover-slips.

Reagents: 1. Glacial Acetic acid

2.Rectified Spirit

Principle:

The red cells make up about 45% by volume of the blood. Haemoglobin ,a respiratory pigment found in the RBC , imparts red colour to the blood , is about 14.7 gm. per 100 ml. of the whole blood. It is made up of two parts ,a non- protein part called **haem** and a protein component called **globin**. Haemoglobin is readily separated into its protein and non-

protein components by treatment with acetic acid. The globin is usually denatured in the process, and the iron containing portion i.e., haem reacts with acetic acid to form **acid haematin** which on heating forms reddish brown microscopic, prismatic insoluble crystalline compound known as **haemin or haemin crystals**.Haemin does not combine with oxygen. The formation of haemin crystals is used as a test for blood.The shape and size of the haemin crystals differ from species to species.

Procedure:

1. The middle finger is sterilized with rectified spirit.

2. The finger is then pricked with a lancet and a drop of blood is taken on a clean slide and air dried.

3.Add 2-3 drops of glacial acetic acid to the dried blood and put a cover slip over it. Gently heat the slide over a low flame until the material is dried up. Cool the slide in air.The slide is observed under the microscope. The haemin crystals were noticed as shining prism-like or star shaped bodies.

Observation/Result:

Numerous shining needle –like,star or prism shaped haemin crystals are observed. Smaller crystals aggregate to form larger crystals of varied shapes.

Precautions:

The lancet/needle should be sterilized before using.
 The slide should not be over-heated.

2.Aim: Determination of clotting time of blood.

Apparatus and Glassware: Stop watch; capillary tube, lancet/ needle.

Material : Fresh blood ,rectified spirit, cotton.

Principle:

When the blood is exposed to air it undergoes coagulation or clotting. The blood coagulation or clotting is a phenomenon when blood looses its fluidity and sets into a semisolid jelly. The blood plasma contains a soluble protein **fibrinogen** which is converted to insoluble **fibrin** with the help of an enzyme called **thrombin**. Thrombin exists in blood as inactive prothrombin, which is activated to **thrombin** by **thromboplastin**, a blood clotting factor released from platelets. The fibrin forms a network of long threads, into the meshes of which the red and white blood cells get entangled. The clot gradually retracts and serum separates out. This phenomenon is aided by calcium ions. The clotting time is the time required for blood to clot in a test tube at 37*C. Variations in clotting time are possible which are due to various factors. Clotting time may vary from the blood of one individual to another. If the anticoagulant heparin is present in the blood, clotting may not take place.

Thromboplastin+ prothrombin + Ca⁺⁺ — Thrombin(active) (inactive)

Thrombin + fibrinogen _____ Fibrin (clot)

Procedure:

Capillary glass-tube method-

1. The finger is cleaned with rectified spirit and pricked with a sterilized needle/lancet. The first drop of blood is wiped away. When the second drop is formed, one end of the capillary tube was held close to it, to allow the blood to flow into capillary tube.

2.As soon as the capillary tube is filled with blood, The stop watch was started.

3. The capillary tube is held in the palm to keep it warm.

4.After 2 minutes, a small piece of capillary tube is broken from one end to check the status of blood.If the blood has not solidified ,more time is allowed.

5. The capillary tube should be about 15 cms. Or $6^{"}$ long.

6.A small piece of capillary tube is carefully broken off every one minute and checked until clotting has taken place. The clotting time was noted immediately.

Result:

The clotting time of my blood is found to be **X** minutes.

Discussion:

3. AIM: Estimation of haemoglobin content of one's own blood.

PRINCIPLE:

The haemoglobin content can be measured in a haemoglobinometer or haemometer which measures the grams of haemoglobin in 100 ml of whole blood and provides an estimate of the oxygen carrying capacity of the RBCs. For clinical analysis, Sahli's haemoglobinometer is widely used. The principle of estimation is based on treatment of the blood with diluted acid (N/10 HCl) to produce a brown coloured acid haematin, followed by gradual dilution with N/10 HCl until the colour matches the standard colour in the non-fading glass tubes.

REQUIREMENTS:

1) **Sahli's haemometer**: The apparatus consists of two vertical sealed tubes carrying standard brown colour. Between the two tubes is a compartment to hold a graduated tube to be used to place the blood sample for dilution and matching with the standard glass tubes. On one side of the tube the

graduations are in gram percentage and on the other side in percentage only. The instrument is supplied with a pasteur pipette which is graduated up to the 20 μ L mark.

- 2) Sterilised needle/lancet
- 3) Rectified spirit
- 4) N/10 HCl



PROCEDURE:

- 1) The graduated tube and pipette are cleaned properly.
- 2) N/10 HCl is taken in the graduated tube up to the 2 mark on the gram percentage side.
- 3) The middle finger is sterilised with rectified spirit and pricked with a sterilised needle/lancet.
- 4) Blood is then drawn into the Pasteur pipette up to the 20 µL mark.
- 5) The blood is released into the graduated tube and mixed thoroughly with N/10 HCl with the help of a glass rod.
- 6) N/10 HCl is then added drop wise to the blood in the graduated tube and stirred properly with the glass rod till the colour of the solution matches that in the standard glass tubes.
- 7) Finally the haemoglobin percentage is noted against a white background.
- 8) The process is repeated two more times and the average is taken.

OBSERVATION:

Individual:

Sl. No.	Haemoglobin content (in G %)	Mean Hb %
1.		
2.		
3.		

Class:

Sl. No.	Students' Names	Haemoglobin content (in G %)

RESULTS: The haemoglobin content is found to be ______ G%

DISCUSSION:



4. AIM : To enumerate the RBC count of one's own blood

PRINCIPLE:

The counting of RBCs is done under the high power of the microscope objective. The RBCs are counted in the central ABCD square of the Neubaur slide. In the central ABCD square, the RBCs are counted in five median squares i.e. 80 smallest squares(four corner squares and the central square). Hayem's diluting fluid is used for dilution of blood; it prevents haemolysis, rouleaux formation, coagulation and bacterial growth. Since Hayem's diluting fluid is used for diluting fluid is used for diluting fluid is used for dilution fluid is used for diluting fluid is used for diluting fluid.

Number of RBCs per cubic mm of blood = <u>Number of cells counted X dilution factor X 400 X 10</u> Number of small squares counted =<u>Number of cells counted X 200 X 400 X 10</u>

80

= Number of cells counted X 10,000

Where, dilution factor = 200, total no. of small squares in the central ABCD square =400 and height of the chamber = 0.1mm.

The number of median squares in which RBCs are counted is 5. Each of these squares have 16 small squares. Therefore, the number of small squares counted is 80. However, the central ABCD chamber has 25 squares, each of which is made up of 16 small squares. Therefore, the total number of cells that constitute an area of 1 mm^2 in the central ABCD square is 400.

All these factors are taken into account while determining the number of RBCs.

In normal adults, the RBC count is about million per cubic mm of blood in males and million per cubic mm of blood in females.

REQUIREMENTS:

- 1) Haemocytometer
- 2) RBC pipette
- 3) Sterilised needle/ blood lancet & rectified spirit
- 4) Hayem's diluting fluid
- 5) Compound microscope

Description of apparatus:

- 1) **RBC pipette**: It is a capillary tube graduated lengthwise and opening into a bulb which contains a red pellet. The graduation shows three figures-0.5 the lowest one, 1 just below the bulb and 101 just at the top of the bulb. The capillary tube continues a little behind the bulb and is connected to a rubber tube with a plastic edge for sucking.
- 2) **Haemocytometer chamber**: It is a specialised slide that has a counting chamber with a known volume of liquid.
- The haemocytometer consists of a heavy glass slide (3" X 1.5") with two counting chambers, each of which is divided into nine large 1mm squares on an etched surface separated by an H-shaped trough.
- A cover slip sits on top of the raised supports of the H-shaped troughs enclosing both chambers. There is a notch at either end where the cell suspension is loaded into the haemocytometer. When loaded with the cell suspension it contains a defined volume of fluid.
- The engraved grid on the surface of the counting chamber ensures that the number of particles in a defined volume of liquid is counted.
- Each counting chamber has its nine 1 mm X 1 mm separated from one another by triple lines. Area of each is 1mm².
- Within each of the larger corner 1mm² squares are 16 smaller squares. These are meant for counting WBCs.
- The central 1mm² area is divided into 25 small squares, each of which is marked into a further 16 smallest squares. The RBCs are counted in five median squares i.e. 80 smallest squares(four corner squares and the central square) of the central area.



Standard Haemocytometer Chamber



PROCEDURE:

- 1) The Neubaur slide is fixed on the microscope and viewed under the 40X objective so as to visualise the central ABCD square; a cover slip is then placed over the slide.
- 2) The RBC pipette is cleaned and dried.
- 3) The middle finger and the puncturing needle/lancet are sterilised with a pad of cotton wool dipped in rectified spirit.
- 4) The middle finger is then pricked with the puncturing needle/lancet so that blood flows freely without squeezing the finger.
- 5) The first few drops of blood are wiped away, the flowing blood is then sucked into the RBC pipette up to the 0.5 mark slowly and carefully.
- 6) The blood in the RBC pipette is then immediately mixed with Hayem's diluting fluid which is sucked into the pipette up to the 101 mark.
- 7) The pipette is rotated so as to allow the blood to mix with the diluting fluid.
- 8) The diluted blood is then run into the grooves on the slide so that it spreads over the central platform of counting squares under the cover slip. Once an optimum drop has been placed ,the cells should then allowed to settle for one minute and then counting should be done.
- 9) The counting squares are brought into focus under the 40X objective of the microscope.
- 10) Under the microscope the RBCs are counted in five median squares- four corner squares and the square in the centre. Any corpuscle lying on lines should be moved upwards or to the right side of the square.

OBSERVATIONS:









CALCULATIONS:

Number of RBCs per cubic mm of blood = $\underline{\text{Number of cells counted X dilution factor X 400 X 10}}$ Number of small squares counted

Suppose total number of RBCs in 5 small squares= A+B+C+D+E

Or, total number of RBCs in 80 smallest squares=A+B+C+D+E

Therefore, number of RBCs in one smallest square= $\frac{A+B+C+D+E}{80}$

Therefore, number of RBCs in 400 smallest squares= A+B+C+D+E X40080

But height of chamber=0.1mm and dilution of blood=200 times

Therefore, 1 mm³ of blood will contain = $\frac{A+B+C+D+E X400X200}{80X0.1}$ = A+B+C+D+E X 10,000

RESULT: After performing the experiment, the number of RBCs counted is found to be_____ million per cubic mm of blood.

PRECAUTIONS:

- 1) The pipette should be rinsed properly with the diluting fluid before the experiment.
- 2) Blood should not clot in the pipette.
- 3) Blood should be accurately pipette upto the 0.5 mark and diluting fluid to the 101 mark.
- 4) The slide and cover glass should be cleaned before the experiment.
- 5) The needle/lancet should be sterilised before pricking.
- 6) RBCs are counted in the central squares of the haemocytometer chamber.

DISCUSSION:

5. AIM : To enumerate the total WBC count of one's own blood

PRINCIPLE:

The counting of WBCs is similar to that of RBCs i.e., counting is done in a haemocytometer with the help of a microscope. The WBCs are counted in the four corner squares of the ruled area of the counting slide. The blood is diluted 20 times before counting. The chemical composition of the WBC diluting fluid is 3% glacial acetic acid stained with 1% gentian violet. The diluting fluid makes RBCs invisible and it stains the WBCs.

Number of WBCs per cubic mm of blood= <u>Number of cells counted X Dilution factor</u> No. of 1 mm² counted X depth of chamber

Where, dilution factor= 20 depth of the chamber= 1/10mm

Since one corner square is 1mm², four such squares will be 4 mm²

Therefore, Number of WBCs per cubic mm of blood= Number of cells counted X 20 X 10 4 = Number of cells counted X 50

The average total number of WBCs is 6000 to 8000 per cubic mm, the normal range being 4000 to 11000 per cubic mm of blood.

REQUIREMENTS:

- 1) Haemocytometer
- 2) WBC pipette
- 3) Sterilised needle/ blood lancet & rectified spirit

- 4) WBC diluting fluid
- 5) Compound microscope

Description of apparatus:

- 3) **WBC pipette**: It is a capillary tube graduated lengthwise and opening into a bulb which contains a white pellet. The graduation shows three figures-0.5, the lowest on;, 1, just below the bulb; and, 11 just at the top of the bulb. The capillary tube continues a little behind the bulb and is connected to a rubber tube with a white plastic edge for sucking.
- 4) **Haemocytometer chamber**: It is a specialised slide that has a counting chamber with a known volume of liquid.
- The haemocytometer consists of a heavy glass slide (3" X 1.5") with two counting chambers, each of which is divided into nine large 1mm squares on an etched surface separated by an H-shaped trough.
- A cover slip sits on top of the raised supports of the H-shaped troughs enclosing both chambers. There is a notch at either end where the cell suspension is loaded into the haemocytometer. When loaded with the cell suspension it contains a defined volume of fluid.
- The engraved grid on the surface of the counting chamber ensures that the number of particles in a defined volume of liquid is counted.
- Each counting chamber has its nine 1 mm X 1 mm separated from one another by triple lines. Area of each is 1mm².
- Within each of the larger corner 1mm² squares are 16 smaller squares. These are meant for counting WBCs.
- The central 1mm² area is divided into 25 small squares, each of which is marked into a further 16 smallest squares. The RBCs are counted in five median squares i.e. 80 smallest squares(four corner squares and the central square) of the central area.





Standard Haemocytometer Chamber



PROCEDURE:

- 1) The Neubaur slide is fixed on the microscope and viewed under the 10X objective so as to visualise the four corner squares; a cover slip is then placed over the slide.
- 2) The WBC pipette is cleaned and dried.
- 3) The middle finger and the puncturing needle/lancet are sterilised with a pad of cotton wool dipped in rectified spirit.
- 4) The middle finger is then pricked with the puncturing needle/lancet so that blood flows freely without squeezing the finger.
- 5) The first few drops of blood are wiped away, the flowing blood is then sucked into the WBC pipette up to the 0.5 mark slowly and carefully.
- 6) The blood in the WBC pipette is then immediately mixed with WBC diluting fluid which is sucked into the pipette up to the 11 mark. The pipette is rotated so as to allow the blood to mix with the diluting fluid.
- 7) The diluted blood is then run into the grooves on the slide so that it spreads over the central platform of counting squares under the cover slip. Once an optimum drop has been placed ,the cells should then allowed to settle for one minute and then counting should be done.
- 8) The counting squares are brought into focus under the 10X objective of the microscope.
- 9) Under the microscope the WBCs are recognised by their retractile appearance and by the colour given to them by dilution. The counting is done in the four corner squares of 1mm².

OBSERVATIONS:









CALCULATIONS:

Number of WBCs per cubic mm of blood= <u>Number of cells counted X Dilution factor</u> No. of 1 mm² counted X depth of chamber

Where, dilution factor= 20 depth of the chamber= 1/10mm

Since one corner square is 1mm², four such squares will be 4 mm²

Therefore, Number of WBCs per cubic mm of blood= <u>Number of cells counted X 20 X 10</u>

= Number of cells counted X 50

4

RESULT: After performing the experiment, the number of WBCs counted is found to be_____per cubic mm of blood.

PRECAUTIONS:

- 1) The pipette should be rinsed properly with the diluting fluid before the experiment.
- 2) Blood should not clot in the pipette.
- 3) Blood should be accurately pipette up to the 0.5 mark and diluting fluid to the 11 mark.
- 4) The slide and cover glass should be cleaned before the experiment.
- 5) The needle/lancet should be sterilised before pricking.
- 6) WBCs are counted in the four corner squares of the haemocytometer chamber.

DISCUSSION:

6. AIM: To study the effect of pH on the activity of salivary amylase on starch

PRINCIPLE:

Salivary amylase present in saliva, acts on starch, a polysaccharide and converts it to maltose, a disaccharide . Starch gives blue colour with iodine. When salivary amylase breaks down starch, the colour of the solution when tested against iodine solution changes from blue to blue-violet to red-violet to red –brown to light brown and finally to yellow after which there is no more change in colour of the solution.

Since enzymes are proteins, they are activated by extremes of pH (proteins are denatured by strong alkalis and acids). Apart from this, pH can produce reversible inhibition of enzymes. When certain amount of enzyme and substrate with buffer at different pH values are set up and all of them incubated at the same temperature at the same time for the same length of time, the amount of chemical change varies from one pH to another. At the optimum pH, the amount of chemical change is maximum. On either side of the optimum pH, the amount of chemical change or the reaction velocity falls off, indicating inactivation of the enzyme.

REQUIREMENTS:

- 1) Test tubes, test tube stands and holders
- 2) Porcelain cavity blocks
- 3) Funnel & cotton
- 4) Pipettes & measuring cylinders
- 5) Water solution

CHEMICALS REQUIREMENTS:

- 1) **1% starch solution:** Prepared by dissolving 1 g of starch in 100 ml of distilled water. The solution is then boiled and cooled.
- 2) **2% iodine solution:** Prepared by dissolving 2 g of KI in 100 ml of alcohol and adding one crystal of iodine.
- 3) Buffer solutions of pH 4.0, 6.8 & 9.2
- 4) Distilled water

Collection of saliva:

A thin film of cotton is made and dipped in water. The water is drained off the cotton and it is spread over the mouth of the funnel in such a way that it acts as a filter. The funnel is kept over a clean glass tube. Apiece of rubber is chewed and the incoming saliva is poured into the funnel. The filtered saliva is collected in the test tube. 1ml of saliva is taken and to it 9 ml of distilled water is added to get 1:9 dilution of the enzyme.

PROCEDURE:

- 1) Six test tubes are taken. Three of them are labelled as experimental tubes and three as control tubes.
- In one experimental and one control tube, 3ml of starch solution and 2ml of buffer solution of pH
 4.0 are taken. 1 ml of diluted enzyme is added to the solution in the experimental tube and the two test tubes are then placed in a water bath set at 37°C.
- 3) Immediately one drop each is taken from the two test tubes with the help of separate droppers and added to iodine solutions in different cavities of the porcelain block. The colour of the solutions is noted and the time of adding is taken as zero minute. This is repeated after and interval of 1 minute for about 5- 8 minutes.
- 4) Steps 2 & 3 are repeated for the second and third experimental and control tubes, the difference being only in the addition of buffer pH 6.8 to the second set of tubes and buffer pH 9.2 to the third set.

OBSERVATION:

TABLE: Effect of pH on the activity of salivary amylase in digesting 3 ml of 1% starch solution

i) pH 4.0

	Reaction with iodine			
Time (in minutes)	Experimental tube	Control tube		

ii) pH 6.8

	Reaction with iodine			
Time (in minutes)	Experimental tube	Control tube		

iii) pH 9.2

	Reaction with iodine			
Time (in minutes)	Experimental tube	Control tube		

RESULT:

The achromic point is reached i.e. a stage arises when starch is digested (yellow colour). The time taken to reach this point is _____ minutes. At pH 4.0 and pH 9.2, the colour of iodine when mixed with the solution from the experimental tube was blue at the beginning and the colour did not change when tested for every minute for a period of about 6 minutes. As for the control tube, the colour of iodine

when mixed with its solution gave blue colour and did not change for every one minute for a period of 6 minutes.

DISCUSSION:

7.AIM: DEMONSTRATION OF RADIAL IMMUNODIFFUSION

Principle:

The interaction between an antibody and an antigen is the main reaction in an immune response. When a soluble antigen is bound by an antibody, the antigen-antibody complex forms a precipitate and the reaction is called precipitation.

Radial Immunodiffusion, also called Mancini method, is a technique by which we can determine the interaction between an antigen and an antibody as well as find out the quantity of an antigen in a given sample. In this test, antigens are placed in a small well made in an agarose gel that contains antibodies. The antigens diffuse out radially from the wells into the agarose gel. When the diffused antigens meet the antibodies in the gel they will interact with each other. The interaction of antigens and antibodies is however defined by the specificity or ability of the antibody to identify the antigen. The antigen- antibody complex formed by the interaction can be observed as a precipitin ring around the well.

<u>Requirement</u>:

Agarose, 1X assay buffer, template, gel punch syringe, antigen samples, antiserum, clean glass slides, petri dish, cotton wool, micropipette, micropipette tips, etc.

PROCEDURE:

- 8. Weight 0.1 g of agarose powder and completely dissolve it in 10 ml of 1X assay buffer by heating the buffer. This will give a 1% agarose.
- 9. The molten agarose is allowed to cool to about 55°C.
- 10. 200µl of the antiserum is added to the agarose solution and mixed well by stirring.
- 11. The agarose solution containing the antiserum is poured onto a clean glass slides placed on a horizontal surface and then left undisturbed to allow the agarose solution to solidify into a gel.
- 12. Wells are cut in the agarose gel using a gel puncher.
- 13. 20µl of the antigen samples are added into the wells.

14. The gel is then placed in a petri dish containing some wet cotton wool and left undisturbed overnight at room temperature.

OSERVATION:

A precipitin ring is seen around the wells in which the antigen samples have been added.

DISCUSSION.

8. AIM: Estimation of oxygen consumption in fish by Winkler's method

INTRODUCTION:

Oxygen is one of the most important chemical components of life processes in any aquatic system. Almost all plants and animals use oxygen for respiration. The main sources of dissolved oxygen are the atmosphere and the photosynthetic processes of green plants. Oxygen from the air is absorbed by direct diffusion and agitation of surface water by wind action and turbulence. The amount of water dissolved in oxygen depends on the area of the water surface, temperature and salinity. The amount of oxygen derived from green plants depends on the density of plants and the duration and intensity of effective light. In undisturbed water with thick vegetation, the photosynthetic activity of plants produce a distinct rise in the amount of dissolved oxygen which reaches a maximum in the late afternoon and falls again at night because of removal by respiration of both plants and animals. The decrease of dissolved oxygen in natural waters is brought about by respiration of biota, decomposition of organic matter, inflow of oxygen results when a combination of these factors acts simultaneously. Aquatic plants and animals show a wide variety of adaptations to obtain the necessary oxygen and to survive the critical period of oxygen deficiency.

Dissolved oxygen is an important factor in assessing water quality; when organic pollution is higher, water has very little oxygen dissolved in it.

THEORY:

Winkler's method is based on an oxidation-reduction reaction which is widely used as a standard means to based on the fact that potassium hydroxide reacts with manganese sulphate to give a white precipitate.

 $MnSO_4 + 2KOH \rightarrow Mn(OH)_2 + K_2SO_4$

$$2Mn(OH)_2 + O_2 \rightarrow 2 [MnO(OH)_2]$$

In the presence of oxygen in highly alkaline solutions, the white precipitate of manganous hydroxide is oxidised to a brown coloured manganese oxyhydrate $[MnO(OH)_2]$. This occurs in direct

proportion to the amount of oxygen present. Thus, the approximate amount of oxygen may be judged from the intensity of the brown colour of the precipitate. In strongly acidic medium, the $Mn(SO_4)_2$ formed by the acid converts the iodide ions back into iodine, itself being reduced back to manganese ions in an an acidic medium.

 $[MnO(OH)_2] + KHSO_4 + 2KI \rightarrow I_2 + MnSO_4 + K_2SO_4 + 2H_2O$

The amount of free iodine is equivalent to the amount of oxygen in the sampleThe amount of iodine can be determined by titration with sodium thiosulphate.

REQUIREMENTS: Burette, pipettes, measuring cylinders, conical flasks, BOD bottle, beakers, glass troughs or jars to keep the fish, fish, weighing pan

CHEMICALS REQUIRED:

- 1) **MnSO₄.H₂O**: Prepared by dissolving 36.4 g of MnSO₄.H₂O in 100 ml of distilled water, followed by stirring with a magnetic stirrer and filtering. The filtrate is Winkler's solution.
- 2) Alkaline iodide solution: Prepared by dissolving 50 g NaOH or 70 g KOH and 13.5 g of NaI or 15 g of KI in 100 ml of distilled water.
- 3) Concentrated H₂SO₄
- 4) $0.025 \text{ N Na}_2\text{S}_2\text{O}_3.5\text{H}_2\text{O}$: Dissolve 12.415 g of $\text{Na}_2\text{S}_2\text{O}_3.5\text{H}_2\text{O}$ in 500 ml of distilled water.
- 5) **1% starch solution**: Prepared by dissolving 1 g of starch in 100 ml of distilled water. The solution is then boiled and cooled.

PROCEDURE:

- 1) The weight of the fish is taken.
- 2) The volume of the glass jars/troughs is measured by filling it with water measured with a measuring cylinder.
- **3**) The water sample is collected from the glass jars/troughs in a suitable BOD bottle taking care that no air bubbles are introduced and closed by a stopper under water.
- **4)** The stopper is then carefully removed and 1 ml of MnSO₄ is added followed by 1 ml of alkaline iodide solution. The stoppers are replaced taking care that no air bubbles enter. Separate pipettes are to be used for each reagent.
- 5) A white then brownish precipitate is formed on introduction of the reagents. The intensity of the brown colour is proportional to the amount of oxygen present. The bottles are shaken vigorously to ensure the mixing of reagents with the water sample.
- 6) The precipitate is allowed to settle down to about half the volume of the bottle. 1 ml of concentrated H₂SO₄ is run along the neck of the bottle. The tip of the pipette should be just under the water surface. The stopper is added without trapping air bubbles. The bottle is shaken vigorously until the precipitate dissolves. A few small air bubbles may be developed due to the presence of acid.
- 7) 25 ml of the treated sample is taken out in a measuring cylinder and introduced in a conical flask with a wide surface.
- 8) 2-3 drops of starch solution is added. The solution develops a blue colour.

- **9)** The solution is titrated against 0.025 N Na₂S₂O₃ filled in the burette. Till it becomes colourless and volume of titrant is noted down. Steps 7-9 are repeated for two more redings.
- **10**) After taking the water sample before respiration, the fish is placed in water.
- **11**) The water sample is again collected one hour after introduction of fish in the water and steps 3-9 are repeated. Estimation should be done immediately after collection of the sample.

OBSERVATION:

i) Before introduction of fish (control)

Number of Volume of sample taken		Burette reading		Volume of	Mean volume
	(ml)	Initial Reading (ml)	Final Reading (ml)	$Na_2S_2O_3$ titrant used ($Na_2S_2O_3$ titrant used (
1.	25			· · · · · · · · · · · · · · · · · · ·	· · · · · · · · · · · · · · · · · · ·
2.	25				
3.	25				

ii) After introduction of fish (experimental)

Number of	Volume of	Burette reading		Volume of	Mean volume
observations	(ml)	Initial Reading	Final Reading	0.025 N $\text{Na}_2\text{S}_2\text{O}_3$	of 0.025 N Na ₂ S ₂ O ₃
		(()	titrant used (ml)	titrant used (ml)
1.	25				
2.	25				
3.	25				

CALCULATIONS:Dissolved oxygen (D.O.) mg/L = $56 \times ml \text{ of } Na_2S_2O_3$ x $N \text{ of } Na_2S_2O_3$ Vol. of sample- vol. of reagents0.01

Volume of BOD bottle= 125 ml

Volume of experimental jar= A ml

Volume of sample taken= 25 ml

Volume of $Na_2S_2O_3$ used for control sample= **B** ml

Volume of $Na_2S_2O_3$ used for experimental sample= C ml

Volume of reagents= 3 ml

Weight of the fish= \mathbf{D} g

- For control sample:
 - D. O. = $\frac{56 \times \mathbf{B}}{25-3} \times \frac{0.025}{0.01}$ = $\frac{56 \times \mathbf{B} \times 2.5}{22}$ = $\mathbf{E} \text{ mg/L}$

1 litre of water contains E mg of dissolved oxygen

Therefore, A ml of water will contain $\mathbf{E} \ge \mathbf{A} / 1000 = \mathbf{F} \mod \mathbf{O}$ dissolved oxygen

• For experimental sample:

D. O. =
$$\frac{56 \times C}{25-3}$$
 x $\frac{0.025}{0.01}$
= $\frac{56 \times C \times 2.5}{22}$
= G mg/L

1 litre of water contains **G** mg of dissolved oxygen Therefore, A ml of water will contain $\mathbf{G} \ge \mathbf{A}/1000 = \mathbf{H}$ mg of dissolved oxygen

Difference in D.O. between control and experimental samples= $\mathbf{F} - \mathbf{H} = \mathbf{J} \text{ mg/ hour}$ Volume of D.O. consumed by the fish = $\mathbf{J} \text{ mg/ hour}$ Weight of the fish = \mathbf{D} g D g of the fish consumed \mathbf{J} mg of dissolved oxygen/hour 1 g of the fish consumed $\mathbf{J} / \mathbf{D} = \mathbf{K}$ mg of dissolved oxygen/hour

Rate of oxygen consumed by the fish= **K** mg of dissolved oxygen/ g of fish/hour **RESULT: DISCUSSION:**