Bharath
INSTITUTE OF HIGHER EDUCATION AND RESEARCH
(Declared as deemed to be university under section 3 of UGC Act 1956, vide notification No.F.9-5/2000-U.3)

20.01.2020

From
Head of the Department,
Department of Genetic Engineering,
Bharath Institute of Higher Education and Research,
Chennai

To
The Dean Engineering,
Bharath Institute of Higher Education and Research,
Chennai

Respected Sir,

Sub: Requesting for Industrial visit- Reg

I would like to request you to permit our 2019 Batch Genetic Engineering students for Industrial visit. I have enclosed the Student name list for your reference. We request you to approve the Industrial visit in the ARMATS Biotech Labs Pvt.Ltd, Chennai and do the necessary transportation facility for the benefit of students and staffs.

Thanking you

Yours sincerely

Dr. P.B Ramesh babu
E mail- rameshbabu.bi@bharathuniv.ac.in
Phone- 9841788001

Bharath Institute of Higher Education & Research (Declared as Deemed to be University u/s 3 of UGC Act, 1956) DEPT. OF GENETIC ENGG.

Dr. P.B. RAMESH BABU, Ph.D.,
Professor and Head
Department of Genetic Engineering,
Bharath Institute of Higher Education & Research
(Declared as Deemed to be University U/S 3 of UGC Act, 1956)
Chennai-600 073. INDIA

ARMATS BIOTEK
TRAINING AND RESEARCH INSTITUTE (ABTRI)

To

The Head of the Department,
Department of Genetic engineering,
BIHER.

Sir,
Sub: Req for Industrial visit – Reg

Regarding Industrial visit we permit your students for Industrial visit at Maduvankarai, Guindy, Chennai 600032 on 25.01.2020 referred to letter dated 20.01.2020

Regards,
Dr.Arumugam

Register office:
New 14/18, Mettu street/Link road
Maduvankarai, Guindy
Chennai 600032

Students Industrial Visit Report - Armats Biotek Training and Research Institute

The B.Tech II year students of Genetic engineering, BIHER had visited the Institute and known about following processes:

- Electrophoresis – SDS PAGE
- Polymerase Chain Reaction
- ELISA – Direct ELISA

Detailed report of the Industrial visit

Polymerase Chain Reaction:

PCR theoretically amplifies DNA exponentially, doubling the number of target molecules with each amplification cycle. The objectives of this experiment involved to Isolate the DNA genome ,amplify a specific gene and to understand the principles of the polymerase chain reaction. The DNA strands were unwound (denatured) by enzyme to form two single strands. A RNA polymerase binds and synthesizes a short complementary piece of RNA on the DNA strand at the initiation site of replication. Followed by the DNA hetero duplex acts as a priming site for the DNA polymerase that binds and produces the complementary strand. Totally three steps of the PCR was performed which includes majorly, First, the template DNA or genetic material was denatured; the strands of its helix were unwound and separated-by heating to 90-96°C. The second step is hybridization or annealing. The Taq polymerase was added to initiate DNA replication, with a few length RNA primers. In the PCR reaction, short complimentary double stranded oligos primers which were complimentary to the gene sequence were added that bind the denatured DNA and act as origins of replications. Following denaturation, the reaction mixture was rapidly cooled to a temperature below the melting point of the specific primers (~55°C). In the third step, the temperature of the reaction was gradually raised to the optimal temperature for the polymerase (68-72°C). The polymerase synthesized new DNA, starting from the primer, the polymerase reads a template strand and generated complementary nucleotides very quickly. The result was two new helices in place of the first, each composed of one of the original strands plus its newly assembled complementary strand.

Electrophoresis – SDS PAGE

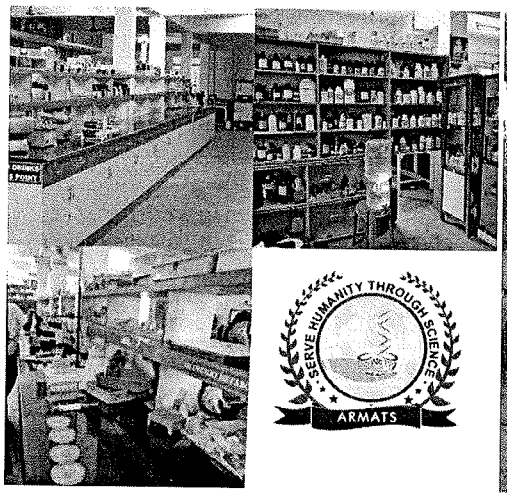
The different buffers required for SDS-PAGE were prepared and the Casting gels for SDS-PAGE were prepared. The surface of the balance with a tissue paper was cleaned and a butter paper was placed on the balance and tare the weight of paper before weighing. 0.5 M Tris-HCl pH 6.8 and 1.5 M Tris-HCl pH 8.8 were prepared according to the protocol. tris-base was dissolved in minimum volume of water and proceeded for adjusting the pH of the buffer to the required value. The pH of 0.5 M tris base solution was adjusted to 6.8 using concentrated HCl. The pH of 1.5 M tris base solution was gradually adjusted to 8.8 using concentrated HCl. 144.1g of glycine, 30.3g of tris base and 10g SDS was weighed and dissolved in distilled water. For the Buffer preparation, the components on a magnetic stirrer were dissolved. For the Gel casting 10% SDS and 10% APS was prepared by weighing the required amount and dissolving it in water. 12.5% resolving gel solution was prepared ,consisting of 3ml acrylamide and bis-acrylamide, 1.5ml of 1.5M Tris–HCl pH 8.8, 60µl of 10% SDS and 60ul of 10% APS, 2.11µl TEMED and 1.38ml water. 12.5% resolving gel prepared earlier slowly was poured along the sides of the glass plates. Stacking gel was prepared consisting of 1 ml acrylamide and bis-acrylamide 1.8ml 1.5M Tris–HCl pH- 6.8, 75ul of 10% SDS and 75ul of 10% APS, 4.5ul TEMED and 1.6ml water.A comb was inserted inside the stacking gel immediately after its transfer into the assembly and the gel was allowed to polymerize for 25 minutes. The combs were removed and the samples were loaded into the wells and buffer was loaded and the electrophoretic run was proceeded. The power-pack was switched OFF ,the lid was removed and the glass plates were taken out of the electrophoresis unit. Finally the gel was carefully removed and place it in a tank containing water for further processing.

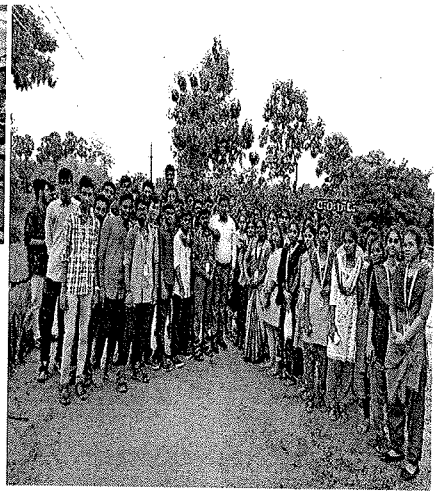
ELISA – Enzyme linked Immuno sorbent assay

Direct ELISA :

Antigen Coating was the first procedure in which the purified antigens solution was diluted to a final concentration of 1-10 µg/ml in bicarbonate/carbonate antigen-coating buffer (100 mM $NaHCO_3$ in deionized water; pH adjusted to 9.6). 100 µL of diluted antigen was added to each well of a microtiter plate. The plate was covered with adhesive plastic and incubated at 4°C overnight (or 37°C for 30 min)and the coating solution was removed and the plate was washed 3X with 200 µL PBS (Phosphate Buffered Saline) buffer(10 mM Na_2HPO_4 and 1.8 mM NaH_2PO_4 in deionized water with 0.2% Tween 20; pH Adjusted to 7.4) with for 5 minutes each time. The second procedure involved Blocking in which 200 µL blocking buffer (5% w/v non-fat dry milk in PBS buffer) was pipetted per well to block residual protein-binding sites. The plate was covered with adhesive plastic and incubated for 1-2 hour(s) at 37°C .The blocking solution was removed and and the plate

was washed 2X with 200 µL of PBS for 5 min each time. The plate was flicked and pat dry. The third step was Reagent Preparation,the diluted standard solutions were prepared followed by the Primary Antibody Incubation Serially diluted the conjugated primary antibody with blocking buffer immediately before use. 100 µL of diluted secondary antibody solution was pipette into each well and the plates were covered with adhesive plastic and incubated for 2 hours at room temperature and finally the content in the wells was removed and washed them 3X with 200 µL PBS buffer for 5 min each time. For the Substrate Preparation , the substrate solution was prepared immediately .The enzymes used for signal detection were horse radish peroxidase (HRP) and alkaline phosphatase (AP), and their corresponding substrates, stopping solutions, detection absorbance wavelengths and colour developed . For the Signal Detection step, 90 µL of substrate solution was pipette in to the wells with the control and standard solutions and the Incubation of the plate at 37°C in the dark was done. Shades of blue was observed in the wells with the most concentrated solutions. Other wells showed no obvious color. 100µL of stopping solution to the wells was pipetted. The sample concentration from the standard curve was interpreted.





STUDENTS NAME LIST

S.No	Roll. No	Name
1	U18BR001	Varsha Vijayakumar
2	U18BR002	Saravanan M
3	U18BR003	Priyadharshini N
4	U18BR004	Reema R
5	U18BR005	Kanakavalli Ramanujam
6	U18BR006	Dyuthi Sai
7	U18BR007	Harini K
8	U18BR008	Athmaja V Christophy
9	U18BR009	Dharanidevi K. S.
10	U18BR011	Magadalene Shelshia. D
11	U18BR012	S Preethi
12	U18BR013	Anupriya
13	U18BR014	Sathya Narayanan G. Y.
14	U18BR015	Giridharan M
15	U18BR016	R Janani Rubasree
16	U18BR017	V Harshavardhan
17	U18BR018	Eralla Divya
18	U18BR019	Bhavitha Sree Pedamalla
19	U18BR020	Angelin Nancy N
20	U18BR021	Putta Neeraja
21	U18BR022	Battula Neha
22	U18BR023	Ahamadulla A
23	U18BR024	Kanna Yamini Durga
24	U18BR025	Aremanda Rajesh Raman

25	U18BR026	Mandapalli Sridhar
26	U18BR027	Akash K
27	U18BR028	Samyuktha R
28	U18BR029	Samyuktha S
29	U18BR030	Mannela Maruthi Sai Ram
30	U18BR031	Mulakala Megha Varsha
31	U18BR032	Vadnam Naviteja
32	U18BR033	Balasubramaniyam. P
33	U18BR034	Pooja Girish Patil
34	U18BR035	Tanaya Prasanna Dehadrai
35	U18BR036	Haritha D
36	U18BR037	Nakka Ganesh
37	U18BR038	Harsha Vardhini G
38	U18BR039	Kolipaka Usha Sree
39	U18BR040	Lavanya V
40	U18BR041	Nandhan P
41	U18BR042	Jangam Laxmikanth Reddy
42	U18BR043	Abhiraami K
43	U18BR045	Neminla Keerthi
44	U18BR046	Vanjivakam Yashwanth Kumar
45	U18BR047	Sharmila Dharshini A
46	U18BR048	Duravalla Soma Sekhar
47	U18BR049	Siddavarapu Harika
48	U18BR050	S Fiza Sultana
49	U18BR051	Monika Kumari K
50	U18BR052	Bhagyalakhsmi M
51	U18BR053	Karunakaran Greeshma

52	U18BR055	Sonali V
53	U18BR057	Melingi Geetanjali
54	U18BR058	Masapogu Surendra Babu
55	U18BR061	Baereddi Sushmitha
56	U18BR062	Potta Swaraj
57	U18BR064	Deeraj Mani Kumar
58	U18BR065	Dadi Sushma
59	U18BR066	Yoghit M

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