THE GUT MICROBIAL GENOMIC STUDY AMONG THE PVTGs OF INDIA

RESEARCH PROJECT BY

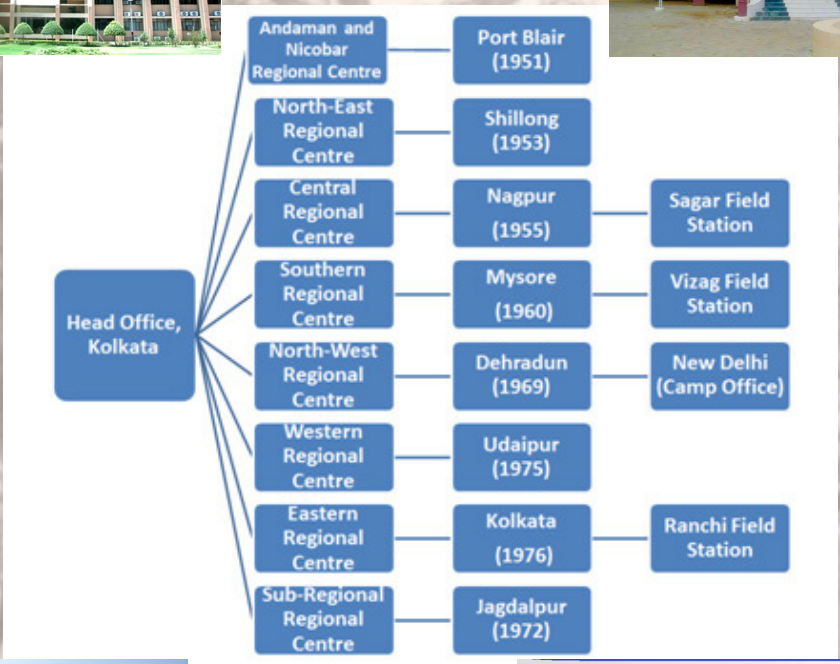
ANTHROPOLOGICAL SURVEY OF INDIA

Ministry of Culture, Government of India

In Collaboration With

Birbal Sahni Institute for Palaeosciences, (An autonomous institute under Science and Technology, Government of India), Lucknow.

DBT- Centre for Microbial Informatics, School of Life Science, University of Hyderabad; (Institute of Eminence), Hyderabad.


Head Office, Kolkata
Andaman and Nicobar Regional Centre
Port Blair (1951)
North-East Regional Centre
Shillong (1953)
Central Regional Centre
Nagpur (1955)
Sagar Field Station
Southern Regional Centre
Mysore (1960)
Vizag Field Station
North-West Regional Centre
Dehradun (1969)
New Delhi (Camp Office)
Western Regional Centre
Udaipur (1975)
Eastern Regional Centre
Kolkata (1976)
Ranchi Field Station
Sub-Regional Regional Centre
Jagdalpur (1972)

ANTHROPOLOGICAL SURVEY OF INDIA'S MANDATE FOR BIOLOGICAL ANTHROPOLOGY RESEARCH

The Anthropological Survey of India (AnSI) is a government organization under the Ministry of Culture, Government of India. It is the only research organisation to pursue anthropological research in all branches of anthropology in a Governmental setup in India. The AnSI's research mandate in the field of biological anthropology includes:

Human Biological Diversity: Research to study the biological diversity among different populations and communities in India. This includes examining variations in physical traits, genetics, and health-related aspects among different groups.

Human Evolution: Research related to human evolution, including the study of fossil evidence, archaeological findings, and genetic studies to understand the evolutionary history of human populations in the Indian subcontinent.

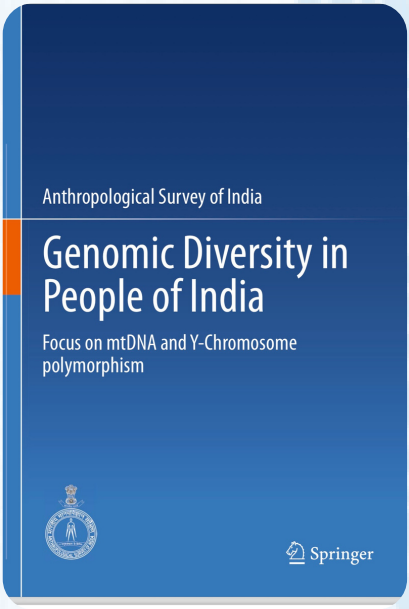
Population Genetics: Research on the genetic structure of different Indian populations and how they have evolved. This includes the study of genetic markers and their distribution in various ethnic groups.

Health and Disease: Research on health-related aspects, such as the prevalence of diseases and nutritional status.

Biocultural Adaptations: Research on how human populations have adapted to their environmental and cultural contexts.

The research in the field of biological anthropology is both to contribute to basic research and help policymakers to make informed decisions related to public health, genetic conservation, and identification of disease and population-specific markers and candidate genes among the populations of India.

BIOLOGICAL RESEARCH IN THE RECENT PAST BY ANSI



The major research projects which have been recently completed under the National Projects by AnSI are:

- Community Genetics extension programme.
- DNA polymorphism in the contemporary Indian population and ancient skeletal remains.
- Anthropometric study among the Denotified, Nomadic and Semi Nomadic communities of India.
- Paleoanthropology

For Community Genetics Extension Programme, Beta Thalassemia Sickle Cell Anemia was considered. The project's objective was to create a massive awareness regarding Hemoglobinopathies and Thalassemia, and facilitate mass screening among the high-risk target groups. Besides that, the project aimed to understand the spectrum of mutations and genetic variation of hemoglobinopathies genes in the study areas and to create a database on the molecular heterogeneity and the frequency distribution of haemoglobinopathies for future reference and monitoring.

More than 12000 samples were collected and screened for the Beta Globin gene, and certain new hemoglobin variants were reported based on the analysis of this data.

In the “DNA Polymorphism in Contemporary Indian Population and Ancient Skeletal Remains” study, the DNA material of Indian populations, including endangered tribal populations of the Andaman Islands, was studied. The survey began to develop a resource of cell lines and DNA samples that can be used to study DNA sequence polymorphisms in contemporary Indian populations with the following objectives:

To study Genetic diversity – mtDNA, Y Chromosome markers;
To understand the phylogenetic architecture of the Indian populations,
To generate a DNA database of Indian tribal populations;
To know the candidate gene association with various diseases;
To identify new candidate genes through genome-wide studies.

PALEOANTHROPOLOGY AND SHIWALIK EXCAVATION STUDY

The study "Palaeoanthropology and Shiwalik Excavation study" was undertaken as a rich ancient human skeletal repository with a wide-ranging collection of about thirty human skeletal series representing a vast horizon of culture from the late Stone Age to the historical period including the Harappan sites was available with AnSI. The project was undertaken with the following objectives:

To recover paleoanthropological and archaeological remains to understand the evolution and cultural adaptation of the hominoids.
To know whether the Siwalik palaeolithic culture was transmitted from Soanian to Acheulian or represents an independent innovation of (Acheulian) technology.

Under this project, fieldwork was conducted in the middle Siwalik region of the Gumarwin area of Bilaspur district and the Upper Siwalik region of the Saketi area of Sirmaur district of Himachal Pradesh.

THE CURRENT PROJECT

GUT MICROBIAL GENOMIC STUDY AMONG THE PARTICULARLY VULNERABLE TRIBAL GROUPS (PVTG) OF INDIA

The present study, "Gut Microbial Genomics Among India's PVTGs", will be the first major study in India on the gut microbial abundance in vulnerable tribal communities (henceforth PVTGs) across India, living in diverse geographical settings while maintaining traditional subsistence practices. PVTGs often have unique dietary practices, and their food sources differ from the other population groups. Thus, the proposed research can help explore the genetic diversity of PVTGs, and compare gut microbiota in different contexts. Additionally, it may shed light on antibiotic-resistant microbes and their association with diet and exposure to biomedicine. Furthermore, the study will inform how dietary shifts impact the gut microbiome and thus may provide insights into the biomedical consequences of changing dietary habits in diverse populations. Enhancing our understanding of the gut microbiome allows us to observe the direct evolutionary effects of dietary shifts and their impact on nutrition and health. The study will also be of special interest to understand the health issues of the PVTGs and recommend strategies for improving their health status.

Brief technical details of the proposed project

Coverage and Sampling:

The study will cover most of India's PVTGs. Out of the 74 listed PVTGs only those which are similar in genetic compositions, dietary practices, and criteria of geographic isolation with some others may be excluded. Faecal samples will be collected from 40 healthy individuals. Various health measurements (height, weight, waist, and hip circumference), body composition (RMR, Visceral fat, Skeletal Muscles, Fat Mass, Percentage of Body Fat), Blood pressure, Blood sugar, and haemoglobin levels will be taken to assess their current health status.

Inclusion and Exclusion Criteria:

Inclusion	Exclusion
BMI *(Asia Pacific, WHO, 2000)*	
Within Normal Range (18.5-22.9)	Underweight, Overweight and Obese
WHR (WHO, 1989)	
Within Normal Range: (Male - <0.95, Female - <0.85)	(Male - >0.95, Female - >0.85)
Haemoglobin	
Normal Range Female – 12.1 – 15.1gm/dl); Male – 13.8 – 17.2 gm/dl)	Anaemic
Blood Pressure (JNC VII)	
Normal Prescribed Values (SBP-≤120mmHg; DBP - ≤80mmHg)	Hypertensive and Hypotensive
Medications	
No biomedicines (chemical drugs) in the last 90 days	Use of biomedicine in the last 90 days
Diseases (30 days reporting)	
No diseases suffered	Some illness or the other suffered
Other Physiological conditions for exclusion	
• Pregnant and Lactating Women • Women who have experienced miscarriage or foetal wastage in the last three months. • Individuals experiencing Constipation and diarrhoea.	

The population covered for faecal samples will also be surveyed for socio-demographic information, health and hygiene behaviours, morbidity and preferred treatment practices, etc., in addition to dietary data. Dietary information will be obtained through Food Frequency (FFQ) method, and nutritional values will be calculated using established guidelines.

SAMPLE COLLECTION AND ANALYSIS

Faecal samples will be obtained from selected participants with their informed consent and following the scientific protocols in this regard to ensure the quality of research. All faecal samples will be transferred to the nearest AnSI laboratory within 24 hours and stored at -200C.

DNA Extraction:

DNA will be extracted from the stool sample using the DNA Stool Mini Kit as per the manufacturer's protocol. DNA will be re-suspended as per the protocol of the Kit.

DNA quantification and quality check:

The extracted DNA will be quantitated at a 260/280 ratio using UV-visible spectrophotometer by absorption technique. The quality of the extracted DNA will be checked by agarose gel electrophoresis

Identification of Gut Microbes and DNA Sequencing:

Illumina deep metagenomic sequencing is a powerful technology for studying the gut microbiome, providing insights into the genetic composition and functional potential of the microorganisms residing in the gastrointestinal tract. The extracted DNA is processed to create a metagenomic library. This involves fragmenting the DNA into smaller, manageable pieces and attaching adapters for sequencing. The goal is to generate a library that represents the genetic diversity of the microbiome. The prepared metagenomic library is then subjected to deep sequencing using Illumina sequencing platforms. This technology can generate vast amounts of short DNA sequences, known as "reads," in parallel. The generated sequencing data will be subjected to extensive bioinformatic analysis to make sense of the genetic information. This process will include:

Read Quality Control: Ensuring that the sequencing reads are of high quality and free from errors.

Taxonomic Profiling: Identifying the microbial species present in the sample by comparing sequences to known microbial genomes (reference databases).

Functional Analysis: Predicting the functional capabilities of the microbiome by identifying genes and pathways present in the sample.

Metagenomic Assembly (Optional): In some cases, metagenomic assembly may be performed to reconstruct microbial genomes from the sequencing data, allowing for a more in-depth analysis.

EXPECTED OUTCOME

The present study among the PVTGs who are still in their primitive mode of subsistence, i.e., essentially omnivorous and dependent on forest produce, having minimum or no direct agricultural practice, is expected to provide base data for comparisons and serve as a reference to compare and reveal the commonalities and variations that have been induced in the gut as a result of the socio-economic lifestyle patterns and the occurrence of different chronic and non-chronic diseases.

The gut microbial profiles of the present research are expected to represent an unadulterated gut that has not been influenced by fast food and preservative-dependent commercial products. Further, these guts are still shielded from the overuse of medicines and antibiotics; thus, antibiotic-resistant microbes are expected to be a rarity in the studied profiles.

The consequences of our behaviours affect not only the external environment but also the internal one. Thus, integrating mechanistically based investigations and microbial ecology studies using sequencing will provide insights into how best to reshape host-microbial interactions to promote health in India.

Research Partners

- Birbal Sahni Institute for Palaeosciences, (An autonomous institute under Science and Technology, Government of India); Lucknow.
- DBT- Centre for Microbial Informatics, School of Life Science, University of Hyderabad; (Institute of Eminence), Hyderabad.

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- Prof. Ch. Venkataramana (Senior Professor, Department of Plant Sciences, Co-Principal Investigator, DBT- Centre for Microbial Informatics, University of Hyderabad)
- Dr. Niraj Rai, Scientist C, Department of Palaeobotany, Birbal Sahni Institute of Palaeobotany, Lucknow
- Dr. Bidyut Roy, Former Professor, Indian Statistical Institute and Presently, Rabindranath Tagore Fellow, Anthropological Survey of India

IN-HOUSE RESEARCH TEAMS:

Principal Investigators

1. Dr. Harasawaradhana, Deputy Director (P), Southern Regional Centre, AnSI, Mysore: Study design, Data & Sample Collection; Training of staff

2. Dr. B P Urade, Deputy Director (P), Head Office – Coordination with State Government Officials for Fieldwork and overall administration of the project

3. Dr. Mithun Sikdar, Superintendent Anthropologist (P), Southern Regional Centre, AnSI, Mysore: Laboratory Analyses and Data Analyses

The above three principal investigators in the team will work with the Director of the Anthropological Survey of India and Principal Coordinators, orchestrating the research and administrative activities.

Principal Coordinators

Laboratory/Laboratories: Dr. VenuGopal P.N (Assistant Anthropologist, Southern Regional Centre).

Fieldwork & Sample collection: Dr. Abhishikta Ghosh Roy (Anthropologist, Central Regional Centre)

Administration and Collaborations: Dr. Shiv Kumar Patel (Assistant Anthropologist, Head Office)

OTHER MEMBERS OF THE RESEARCH TEAM

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44.	Ms. Soma Bhowmik, JRF
45.	Ms. Arna Chatterjee

THE PROJECT IS APPROVED BY INSTITUTIONAL ETHICAL COMMITTEE

Approved Login registration on NECRBHR, DHR Portal.
Reference No. EC/NEW/INST/2023/3910 Dated 10.08.2023

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Lay Person
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Social WorkerFormer, Project Officer,State Public Health Cell, West Bengal State Rural Development, Panchayats And Rural Development Depart, Govt. Of West Bengal

PVTGs IN INDIA

State-wise list of Particularly Vulnerable Tribal Groups (PVTGs)						
Name of States/Union Territory	**S.No**	**Name of the Particularly Vulnerable Tribal Group**		**Name of States/Union Territory**	**S.No**	**Name of the Particularly Vulnerable Tribal Group**
Andhra Pradesh (including Telangana)	1	Chenchu		Maharashtra	41	Katkaria /kathodi
	2	Bodo Gadaba			42	Kolam
	3	Gutob Gadaba			43	Maria Gond
	4	Dongaria Khond		Manipur	44	Maram Naga
	5	Kutia Kondha		Orissa	45	Chukutia Bhunjia
	6	Kolam			46	Birhore
	7	Konda Reddi			47	Bondo
	8	Kondasavara			48	Didayi
	9	Bondo Porja			49	Dongaria Khond
	10	Khond Porja			50	Juang
	11	Parengi Porja			51	Kharia
	12	Tothi			52	Kutia Kondha
Bihar (including Jharkhand)	13	Asur			53	Lanjia Saura
	14	Birhor			54	Lodha
	15	Birjia			55	Mankirdia
	16	Hill Kharia			56	Paudi Bhuiya
	17	Korwa			57	Saura
	18	Mal Paharia		Rajasthan	58	Saharia
	19	Parhaiya		Tamil Nadu	59	Irular
	20	Sauria Paharia			60	Kattunayakan
	21	Savara			61	Kota
Gujarat	22	Kolgha			62	Korumba
	23	Kathodi			63	Paniyan
	24	Kotwalia			64	Toda
	25	Padhar		Tripura	65	Raing
	26	Siddi		Uttar Pradesh (including Uttrakhand)	66	Buksa
Karnataka	27	Jenu Kuruba			67	Raji
	28	Koraga		West Bengal	68	Birhor
Kerala	29	Cholanaikayan			69	Lodha
	30	Kadar			70	Totos
	31	Kattunayakan		Andaman & Nicobar island	71	Great Andamanies
	32	Koraga			72	Jarawa
	33	Kurumbas			73	Onge
Madhya Pradesh (including Chhattisgarh)	34	Abujh Maria			74	Sentinelese
	35	Baiga			75	Shom Pen
	36	Bharia				
	37	Birhor				
	38	Hill Korba				
	39	Kamar				
	40	Sahariya