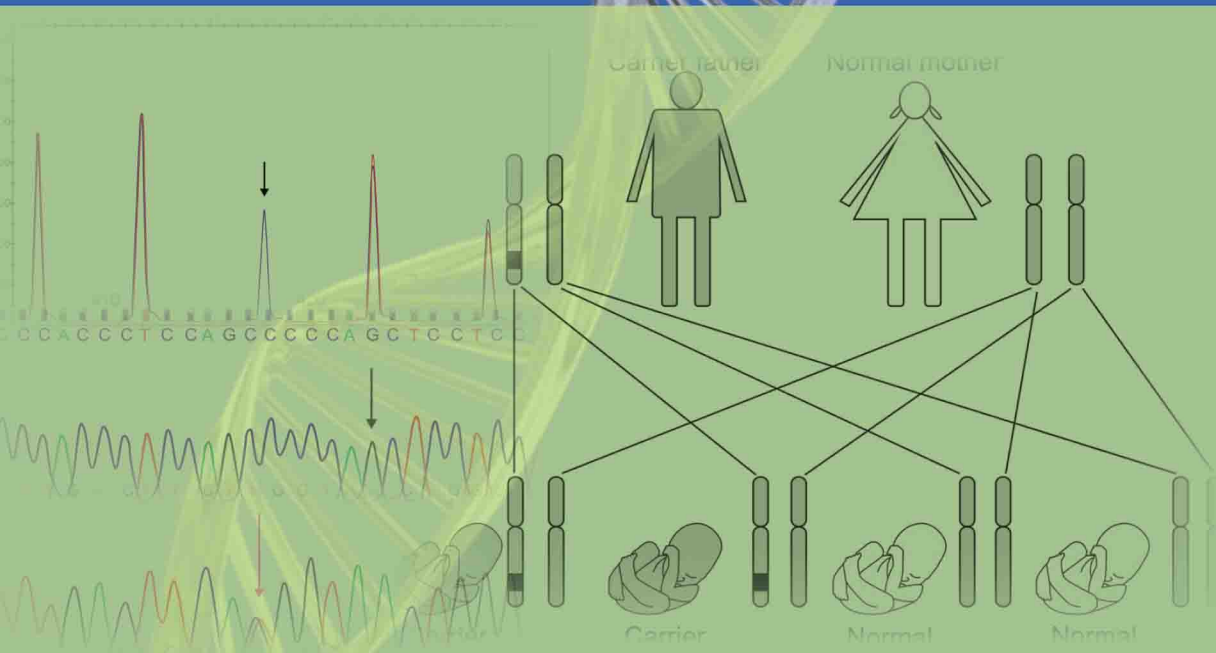
# LABORATORY MANUAL FOR MOLECULAR GENETIC TESTS

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*Foreword*
Ishwar Chandra Verma

JAYPEE

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# 3 Sample Collection

For DNA based tests, peripheral blood sample (9–10 mL) is collected in an EDTA vacutainer. Smaller volumes may be collected from neonates and sick babies. DNA extraction must be started on the same day of sampling whenever possible. Blood samples can be stored in the refrigerator for up to one week without any significant loss in quality or yield of DNA. Storage for longer periods must be avoided and when unavoidable, must be done in a deep freezer. Frozen samples have to be processed immediately after thawing.

For prenatal diagnosis, either chorionic villus (CV) sampling, which is usually a first trimester or early second trimester procedure (after 10 weeks of pregnancy) or amniotic fluid (AF) sampling, which is a second trimester procedure (after 16 weeks of pregnancy) is done.

CV tissue is collected in special media (see annexure) in a sterile tube and transported from the operation theater to the diagnostic lab. Once it reaches the laboratory, the tissue is cleaned under dissecting microscope to remove blood, mucus and maternal tissue. The tissue should be cleaned carefully to avoid maternal contamination.

Amniotic fluid samples are normally received in a sterile centrifuge tube and any blood staining should be recorded. About 15–20 mL of fluid can be easily aspirated, which is usually sufficient for either molecular or cytogenetic analysis.

All samples should be carefully labelled with patient's name, age, registration number and date of collection of sample.

Strictest quality control procedures need to be adhered when handling prenatal samples. It may be a good idea to handle one sample a time whenever the workload permits.

DNA should be stored at 2–8 °C for short duration and at –20 °C to –70 °C for long-term storage. Frequent freeze-thawing should be avoided and preferably DNA should be aliquoted in small volumes.

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LABORATORY MANUAL FOR

# MOLECULAR GENETIC TESTS

This book is a compilation of molecular techniques that are routinely performed for diagnosis of common single gene disorders. It provides a brief summary about the disease, etiology and inheritance with illustrations before giving the details of the actual laboratory procedures. This is an easy-to-follow manual and will be very useful and handy for the aspiring diagnostic laboratories, young scientists and clinical geneticists.

**Madhumita Roy Chowdhury** MSc PhD is working as a Senior Scientist in Genetics Unit, Department of Pediatrics, All India Institute of Medical Sciences, New Delhi, India. She has almost 16 years of research experience in the field of molecular genetics and cytogenetics, primarily in diagnostics. Her main research area has been X-linked recessive disorders, such as Hemophilias, Fragile X and Duchenne Muscular Dystrophy; autosomal dominant disorder such as Tuberous Sclerosis Complex, and skeletal dysplasias such as Achondroplasia and Hypochondroplasia to name a few. She has been instrumental in starting many new techniques in the molecular laboratory. She has many publications in national and international journals and chapters in books to her credit.

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