ISSUE - 1

IAP BENGALURU

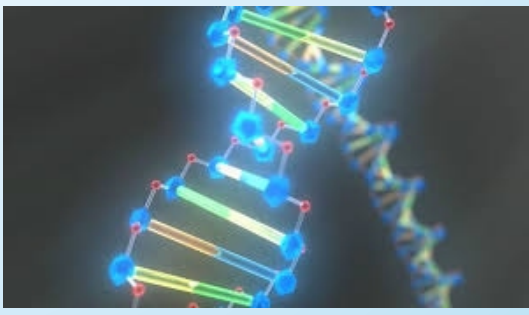
PEDISCAN

MONTHLY NEWSLETTER OF

IAP BENGALURU







JANUARY - 2023

Contents

Message from The President

Dear IAP members and friends,

First of all, New year greetings to you from IAP Bengaluru!!

Congratulations to all the team members of PEDISCAN for bringing the monthly colourful e-magazine.

It is indeed a great pleasure to be a life member of IAP. All of us know that our branch has grown like a banyan tree with many esteemed members, multiple subspecialities and chapters. The credit for this goes to all our previous office-bearers and executive members along with IAP members in Bangalore who, since its inception, have made invaluable contributions.

As we start our year's activities, we, as pediatricians, should reach out to the parents and public regarding the many issues that arise in the upbringing of children. We must discuss promoting health and prevention of illness or harm to children and not be reduced to reacting after a problem occurs. This awareness campaign must involve medical & nursing students in colleges, trainees in other teaching institutions as well as all our practising pediatricians, where we come across different sections of people. Once this happens, gradually, parents will feel that pediatricians are transparent while managing the issue.

This year we move towards "Our Priority: Child & Adolescent Safety" which includes safety at school, in transport, at home, on the roads, in public places like malls etc. This is in view of the fact that many innocent children and adolescents are being negatively affected by modern life in both urban and rural areas. We intend to bring SOPs for the same. I request all our members to give us inputs about safety issues and how to tackle the same.

This year is a great time for all of us to further enhance public education through e-portals like facebook, youtube videos, twitter, etc. I request all our members to take this up as a priority, to increase our presence in all sections of the society, apart from our routine work at our establishments.

We will be following all the National Health Programmes for children and adolescents and Guidelines set out by Central IAP under the leadership of CIAP President Dr Upendra Kinjawadekar, Honorary Secretary General Dr Vineet Saxena and CIAP Team 2023.

This month we have taken up the physical PG teaching programme on Sunday the 22nd of January starting with Genetics in Pediatric disorders and a scientific session in the evening including the genetics and vaccine topics. Also, we have conducted a health camp at RR nagar on 8th, Sunday morning.

I heartily congratulate our immediate past president of IAP BPS, Dr G V Basavaraj, who was elected as President-Elect 2023 for CIAP. We will be seeing and experiencing vibrant activities under him at the National level. I congratulate our EB members Dr E Adarsh, Dr Basavaraj Patil, Dr Karunakara B P, Dr Rajendra Salagare and Dr Sumitha Nayak for being elected from Karnataka for this year 2023. I also congratulate Dr Priya Shivalli, elected as Vice President of IAP Karnataka for 2023 who will be coordinating IAP activities.

This year we are having the Bangalore Pedicon from 9th to 11th, June 2023 (2nd Saturday & Sunday) at NIMHANS Auditorium with many workshops, scientific sessions with expert discussions and cultural programmes. Kindly register for the same at earliest with EARLY BIRD OFFER which closes on 22-01-2023.

I congratulate the entire team members of PEDISCAN under Dr Nagalatha and Dr Gowri Somayaji for accepting the responsibility as Editor and Executive editor of the e-magazine which will be rolling out every month this year. I request our members to contribute articles in their field of interest to make it more informative as well for the interesting non-scientific contents like poems, arts, hobbies and healthy habits.

We have a wonderful team of IAP Bengaluru 2023 with enthusiastic office bearers and executive members advised by seniors like Dr Shanthraj, Dr Gnanamurthy, Dr Geetha Patil and historian Dr Kishore Baindur. I congratulate Dr Ravishankara M and Dr Gunda Srinivas who are with us for all our digital programs. I personally thank Dr Bhaskar Shenoy and Dr Sharath Chand for helping us with our financial management. Dr Nandish is with us for clarifications in legal matters.

I am fortunate enough to have Dr Chidananda N K as Secretary and Dr Harilal Naik as Treasurer with me for the intense work of IAP Bengaluru.

President-Elect Dr Somashekar will support mewith the decision-making during the year.

Our Vice presidents Dr Rajashekarmurthy G R and Dr E Adarsh will be closely associated with all the programmes. Our Joint Secretary Dr Nithyananda S K brings his experience to our team. All our esteemed EC members Dr Chandrashekar M A, Dr Dhananjaya P, Dr Khader Farooq, Dr Mallesh K, Dr Maaz Ahmed, Dr Padma Prakash, Dr Prashanth M V, Dr C P Ravikumar, Dr Sahana Devadasand DrSowjanay G Thave already volunteered for the many tasks ahead of us.

We are happy to inform you that Chairmen and convenors of various committees have volunteered to guide us & conduct the numerous activities of IAP Bengaluru.

Hope IAP BPS Team 2023 will support and be supported by all IAP members in fruitfully carrying out activities throughout the year.

Dr. S M Prasad
President, IAP Bengaluru 2023

Message from The Secretary

Dear Bengaluru IAPians,

I wish you all a very happy and prosperous new year 2023!

It gives me immense pleasure, as Secretary of the branch, that our inaugural edition of the monthly newsletter Pediscan is being released with excellent scientific articles.

I sincerely thank our Pediscan team headed by Dr Nagalatha and Dr Gowri Somayaji and the team members Dr Poornima, Dr Shalini Sharma, Dr Sushma, Dr Naresh and Dr Jeffrey. With their excellent efforts, I am pretty sure that every member of IAP Bengaluru will not only eagerly wait for the next edition but also will archive the editions for future reference.

I also request each one of you to contribute whole-heartedly by providing academic and non academic articles both in English and Kannada as the team keeps bringing to us the updates about the organisation's activities.

Jai IAP!

Dr. Chidananda N K
Secretary, IAP Bengaluru 2023 - 2024

Message from The Treasurer

Warm greetings for Happy New Year 2023 and Makara Sankranti from the New IAP Bengaluru Team 2023 led by The President Dr. S.M. Prasad.

It gives me immense pleasure and happiness to communicate through this flagship e-magazine PEDISCAN.

I wish all the best to the new team of PEDISCAN led by Chief Editor Dr. Nagalatha and Managing Editor Dr. Gowri Somayaji, who will carry forward the legacy and surpass the benchmarks set by their predecessors.

I was fortunate to work under the most dynamic and popular President of IAP BPS Dr. G.V. Basavaraj as a Treasurer for the year 2022, who has led the previous team successfully and carried the academic and non-academic activities to great heights. Myself and my team of 2023 led by Dr. S.M. Prasad will look forward to carry forward the legacy and achieve high standards set out by previous teams of IAP BPS.

I look forward to carry forward the financial discipline and management done by our previous Treasurers to higher levels in the changing scenarios, challenges, and in line with the laws of the land with cooperation and support from all mentors, seniors, members of IAP Bengaluru and the enthusiastic new team of 2023.

Looking forward for all your support, suggestions, and contributions throughout the year.

Warm Regards,

Dr. Harilal Naik M L
Treasurer, IAP Bengaluru 2023 - 2024

Message from The Past President of IAP BPS 2022

Namaskara to all at IAP Bengaluru!

Let me begin with congratulations to the new team of IAP Bengaluru led by my friend, the dynamic Dr Prasad. This is a young, energetic, passionate team of office bearers and executive committee members who will take our branch to higher glories

IAP BPS has always made its presence felt in Bengaluru and at the national level. Our Stalwarts have headed the CIAP and its various sub-speciality chapters over the years. We have won awards, including best branch, multiple times. In Bengaluru, our regular academic sessions have been a point of pride where we share both knowledge and experience with each other. I have had a very successful tenure heading IAP BPS and now move on, secure in the knowledge that Dr Prasad, Dr Chidanand and Dr Harilal will take it to greater heights.

Even as I travel all over the country fulfilling my obligation to the Central IAP, Bengaluru will remain my home and I hope to be part of its activities as often as possible. Wishing the IAP Bengaluru team 2023 and the Pediscan team all success!!

Dr. G V Basavaraja
President - Elect CIAP 2023
Past President of IAP BPS 2022

Message from The President, IAP Karnataka

HAPPY NEW YEAR 2023

Warm greetings from myself & my Team 2023 of IAP Karnataka. Its great pleasure to address you through this newsletter PEDISCAN.

Our team 2023 is mixture of youth & experienced. With all your wishes & support we could select our team unanimously. Looking forward to work in liaison with central IAP & all district branch. Looking forward the co-operation from all experienced seniors & past office bearers in IAP. Academic excellence & improvement in skill remain the utmost top priority of our team . We encourage all the district branch to conduct CME'S , Health days & other centrally sponsored programmes. We suggest all members to participate in health camps to reach the Unreach& Special need childrens

I take this opportunity to thank all those who helped us to from a team & also invite you all to KARPEDICON 23 on oct 13,14 & 15th 2023 at Kalaburagi.

Congratulation Team 2023 for bringing out inaugural issue & best wishes for the successful year ahead.

Your's in Academy

Dr. Sharanagouda Patil
President IAP Karnataka 2023

Message from The IAP KTK HSG

Greetings from IAP KSB Kalaburagi

Wishing you all a Happy and Prosperous New year 2023.

With all your blessings and support we could make the team for this year 2023. We seek your continuous co-operation in bringing a change for the better management of sick and needy children. We will be in communication with you all regarding the programs that are to be chalked out both from central office and ourself. we encourage fully for the local activities. Looking forward to your valuable support towards academics, skill improvement and any other activities.

With warm regards

Dr. Rohit Bhandar
HON SEC GEN
IAP KSB

Message from The VP, IAP KSB

Dear IAPians,

Wishing you all a Happy New Year 2023!

Best wishes to IAP Bangalore Team 2023 headed by Dr. Prasad SM for this new beginning and all future endeavours. Best wishes to Dr. Nagalatha and editorial team of Pediscan.

Warm regards,

Dr. Priya Shivalli
Vice President, IAP KSB
Bangalore Zone
Hon Secretary,
IAP Bangalore 2021-2022

From the Editor's Desk

Greetings from the editorial team

"The magic in new beginnings is truly the most powerful of them all" - Josiyah Martin

It's a new year, new verse, a new chapter... A new team taking over...

The year that passed, 2022, has put IAP BPS in the lime light at National level with our Beloved Dr. Basavaraja winning the coveted post of central IAP President-Elect. We not only realised our dream of Mission Basav but in the process, we saw a united IAP Bengaluru... a united IAP Karnataka.

As Sir leaves the post of President of IAP BPS to enter the National arena, he has set high standards for the upcoming team headed by Drs. Prasad and Chidananda.Team 2023 has both stalwarts of IAP BPSand a young and dynamic executive board and we are sure that IAP Bengaluru will continue to scale new heights.

ಪೊಸ ಚಿಗುರು ಹಳೆ ಬೇರು ಕೂಡಿರಲು ಮರಸೊಬಗು ।
ಪೊಸಯುಕ್ತಿ ಹಳೆತತ್ತ್ವದೊಡಗೂಡೆ ಧರ್ಮ ॥
ಋಷಿವಾಕ್ಯದೊಡನೆ ವಿಜ್ಞಾನ ಕಲೆ ಮೇಳವಿಸೆ ।
ಜಸವು ಜನಜೀವನಕೆ – ಮಂಕುತಿಮ್ಮ ॥

Pediscan also has a new editorial team and we will do our best to match the high standards set by Drs Kishore Baindur, Shubha Badami et al.

Pediscan will be a showcase of achievements and activities of the Bengaluru branch, the sub-chapters, as well as its individual members. So we appeal to all the members to please keep us posted about the academic activities, school health programs, the community service and the charitable activities that we are very sure so many of you participate in. Let us share and publish these events and encourage more colleagues to take up such activities. All information regarding group or individualactivities are welcome.

The last few years have also taught us the value of community. We, the members of IAP Bengaluru, are a collective with similar academic and professional interests but we are also friends and colleagues. We would like to offer Pediscan as a platform for members to also share their personal achievements, interests and passions outside of pediatrics too, and encourage the sense of community.

We cannot build a future without acknowledging the past. As suggested by our historian, Dr Kishore Baindur, we plan to include a section'DOWN MEMORY LANE' in future editions.

We will continue to share academic pearls, but aim to keep the articles simple, applicable to all, with tips for practicing pediatricians and postgraduate students. This month's focus is on Genetics as relevant to pediatricians, with an overview of Karyotyping by Dr Surya G Krishnan and a guide to the diagnostic approach to genetic disorders by Dr Sanjeeva GN.

A Letter to the Editor section will be included in later editions, so members can share their view points

A small quiz is planned for every edition, and people who answer it correctly, quickly and consistently will be aptly rewarded at Bangalore Pedicon.

Here's hoping for a very interactive year

Dr. Nagalatha S, Chief Editor
Dr. Gowri Somayaji, Managing Editor
& Team Pediscan 2023

Message from The Historian

“History (derived from Ancient Greek ἱστορία (historía) 'inquiry; knowledge acquired by investigation') is the systematic study and the documentation of the human activity. The time period of events before the invention of writing systems is considered prehistory. “History" is an umbrella term comprising past events as well as the memory, discovery, collection, organization, presentation, and interpretation of these events. Historians seek knowledge of the past using historical sources such as written documents, oral accounts, art and material artifacts, and ecological markers. History is not complete and still has debatable mysteries.”

Thus is explained about history in the WIKIPEDIA. Considering the nearing of a half century since the establishment of our revered organisation, the IAP, BPS, it is but natural for many of to wonder about the journey of this glorious body through the decades. Documentation and recording it for posterity would be our endeavour in the days to come.

Growing form a small seed sown in 1976 under the banner of the Bangalore Pediatric Society, our organisation has reached the present giant proportions with a membership of a thousand and more. Illustrious leaders have adorned the portals of this august body and toiled to bring it to this stature.

Many a laurel has come along during this long journey including “Best Branch” awards, etc. Our leaders have reached the pinnacle of National level IAP. Many others have lead sub speciality chapters and held many important posts at national and regional levels.

The past was worth remembering and the future is bright. It is for us members to contribute to make our IAP, Bengaluru more vibrant and be the leader as always.

Capsules of details “DOWN MEMORY LANE” would be presented in the monthly meetings henceforth as a regular feature. It is but next to impossible to recollect all events and happenings that have by gone. We request your kind cooperation and inputs to make our efforts fruitful and authentic.

Dr. Kishore Baindur
Historian, IAP Bengaluru

In the year 1988, a major devastating road traffic accident with multiple major fractures all over the body brought me to Bangalore and introduced me to BPS. I participated in the National conference held at Ashoka hotel and began my journey in BPS. BPS has educated me, refined me, to become what I am today and has given me every opportunity to achieve all my milestones. My association with BPS, which in due course became IAP Bengaluru, has given me a lot of friends who have shared my views and helped me to achieve my goals. I am eagerly waiting for someone to break the records of “Team 2015” in respect of number of registrations for Bangalore Pedicon and the total contribution to IAP Bengaluru. Today I feel very proud to say I am a humble member of IAP Bengaluru.

Dr. Shantharaj
Past-President IAP BPS

Dr. Surya G Krishnan
Consultant Pediatrician
and Pediatric Geneticist

KARYOTYPING

Karyotyping is a medical term which is used frequently in Pediatric practice. Let us have some detailed information about this basic yet very important genetic test.

WHAT IS KARYOTYPING?

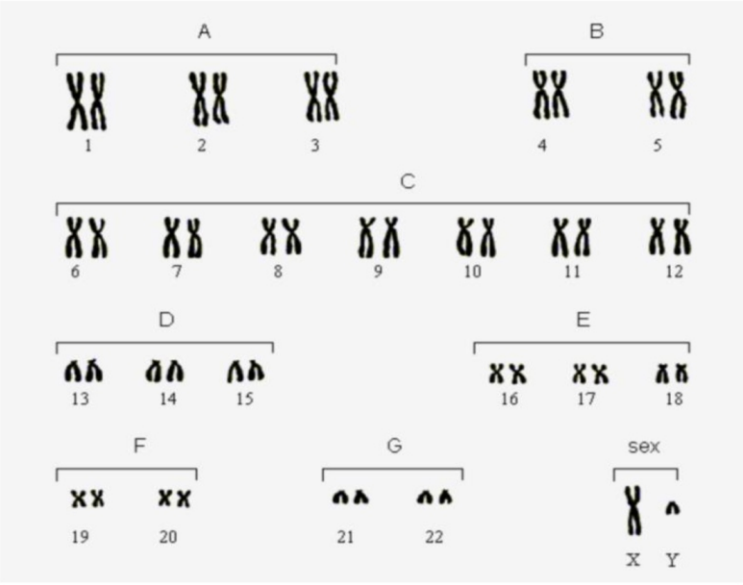
Karyotyping is a cytogenetic technique comprising the process of arranging, pairing and organizing the chromosomes in a specific manner followed by analysing each pair to find any numerical or structural chromosomal variations.

In 1956, Tijo JH and Levan A counted the human chromosomes for the first time and confirmed the total number to be 46. Ever since then and even before that various techniques have been developed for visualization of chromosomes.

TYPES OF CHROMOSOMES:

TYPE	POSITION OF THE CENTROMERE	p ARM AND q ARM
Metacentric	in the exact middle of the chromosome	both arms are of the same length
Submetacentric	closer to the middle region	both arms of different lengths (p arm smaller than q arm)
Acrocentric	closer to the end	very small p-arm
Telocentric	at the end	no p-arm

Chromosomes have been classified based on their size and position of centromere into 7 groups from A to G.



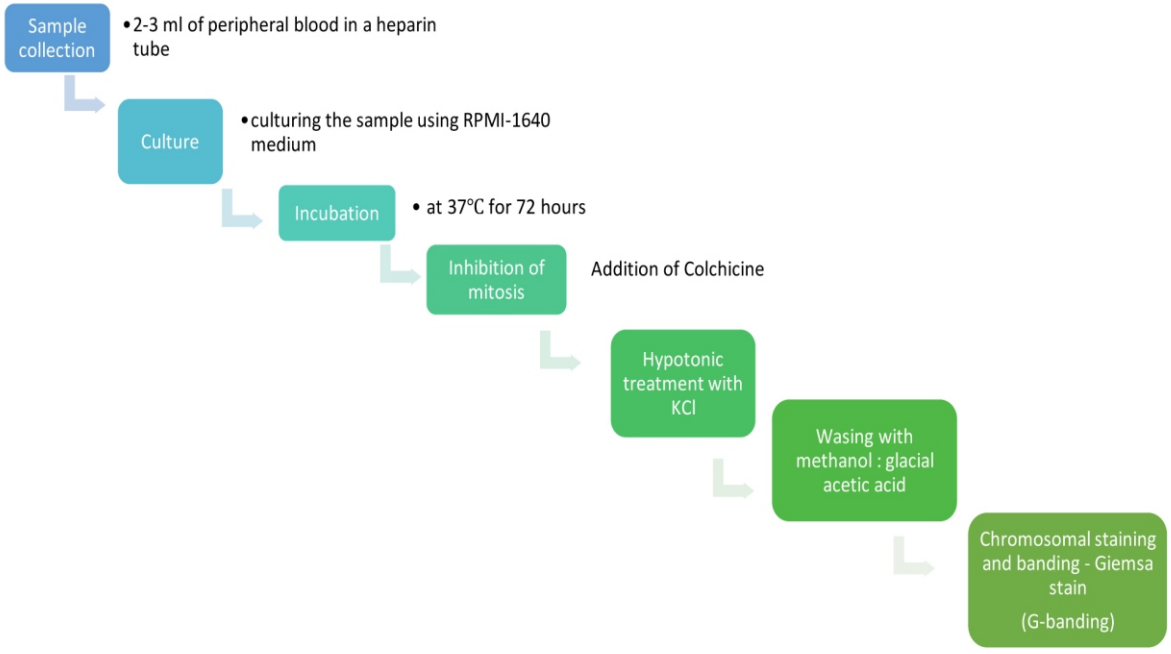
Chromosomes and Karyotype (web-books.com)

GROUP	CHROMOSOMES	DESCRIPTION
A	1,2,3	Large, metacentric
B	4,5	Large, submetacentric
C	6,7,8,9,10,11,12, X	Medium-sized, submetacentric
D	13,14,15	Medium-sized, acrocentric with satellites
E	16,17,18	Medium - short metacentric or submetacentric
F	19,20	Short metacentric or submetacentric
G	21,22, Y	Short acrocentric with satellites (No satellite for Y chromosome)

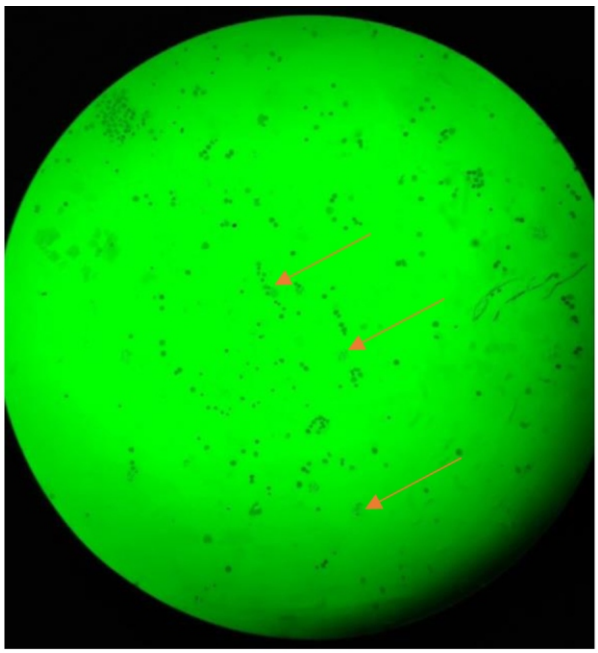
SAMPLES USED FOR KARYOTYPING:

1. Peripheral Blood Leucocytes: most used sample, 2-3 ml blood in heparin sample
2. Chorionic villous sample
3. Fetal cells from amniotic fluid by amniocentesis
4. Skin biopsy (fibroblast culture)

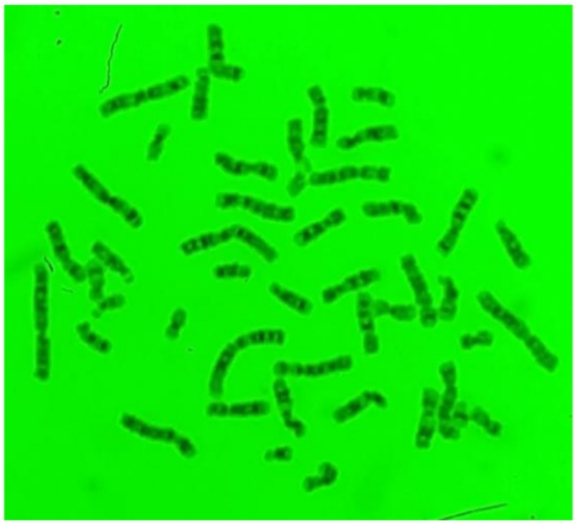
TECHNIQUE OF KARYOTYPING:



G- banding (treating the metaphase chromosomes with trypsin for partial digestion followed by Giemsa staining) results in unique alternate dark and light bands on each homologous pair that helps in identifying each chromosome. The euchromatin regions rich in Guanine and Cytosine nucleotides (GC rich) are transcriptionally more active, have less condensed chromatin and incorporate less Giemsa stain hence appear light where as the heterochromatin regions rich in Adenine and Thymine (AT rich) which are relatively gene poor and have more condensed chromatin, incorporate more stain and appear dark.



: Indicates metaphases spread out under light microscopy at 10X resolution



Single metaphase with individual chromosomes at 100X resolution under oil immersion

COMMON CHROMOSOMAL ABNORMALITIES DETECTED BY KARYOTYPING

1. Numerical abnormalities (aneuploidy) 2. Structural abnormalities

NUMERICAL	STRUCTURAL
Monosomy	Deletion
Trisomy	Duplication
Tetrasomy	Translocation
	Inversion
	Ring
	Breakage (Fanconi anemia)

WHEN TO ORDER KARYOTYPING?

1. Child with syndromic features (e.g., Down Syndrome, Trisomy 13, Trisomy 18, Turner Syndrome, Klinefelter syndrome)
2. Child with multiple congenital anomalies
3. Child with short stature +/- primary amenorrhoea in girls
4. Child with ambiguous external genitalia
5. Child with suspected chromosomal breakage syndromes (e.g. Fanconi anemia)
6. Couple with recurrent pregnancy loss
7. Couple with infertility, primary or secondary
8. Cancer genetics e.g., Chronic Myeloid Leukemia
9. Parents of the index child showing a chromosomal abnormality, especially with translocation, deletion, duplication etc.

HOW TO READ A KARYOTYPING REPORT?

A pictorial representation of the chromosomes arranged in pairs, in the order of their decreasing size is called a karyogram, which is the report generated after karyotyping.

The final report consists of details of the person in whom the test is done, indication for testing, details about the sample and technique used for testing, pictorial representation of chromosomes, interpretation, and recommendation if any further testing is needed.

Always cross-check the details of the person in whom the test is done, including age/date of birth, gender, the sample collected and indication for testing.

Let us try to interpret a few reports, starting with the one of the most common scenarios we come across. There are three basic steps while reading a karyogram. Each set of information is separated by a comma.

1. The total number of chromosomes: Is the total number of chromosomes 46 or is there any aneuploidy?
2. Sex chromosomes: XX for the female and XY for the male gender
3. Other information: gain (e.g., trisomy) or loss (monosomy), deletion or duplication involving any chromosome

For example:

a) 47,XY,+21

Here 47 represents the total number of chromosomes, which has 1 extra chromosome apart from the normal number of 46. Hence this is aneuploidy.

XY denote the sex chromosomes, meaning this is a male child/adult.

+21 denotes the presence of an extra chromosome 21. There are 3 copies of chromosome 21 causing trisomy 21, commonly known as Down syndrome.

b) 46,XY,t(9;22)(q34;q11.2)

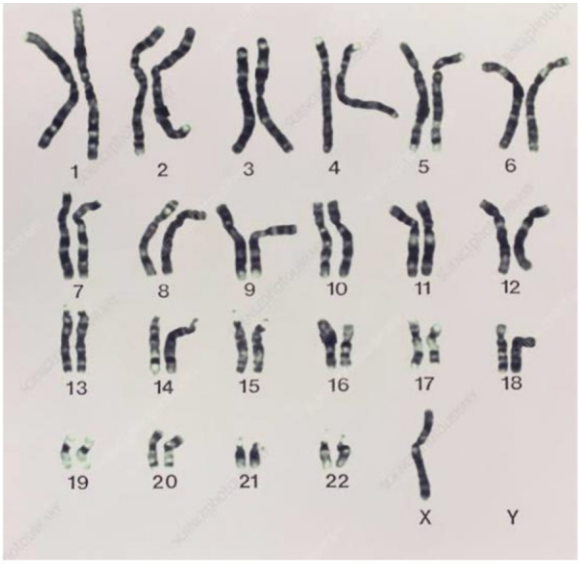
Chromosome number is 46, there is no aneuploidy.

XY denote male sex chromosomes.

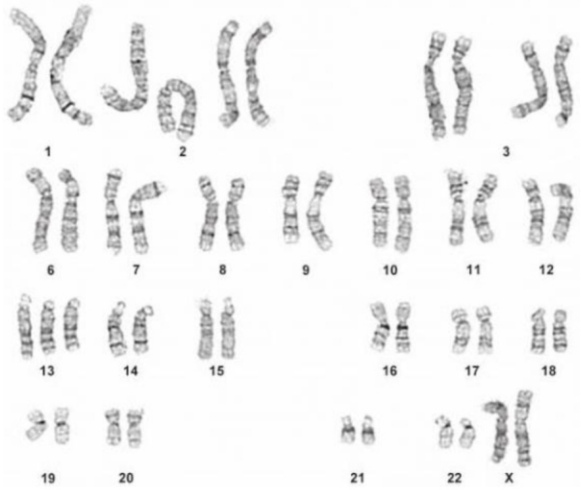
t(9;22) mentions translocation involving chromosomes 9 and 22.

q(34;11.2) mentions that the translocation involves the long arms, band 34 of chromosome 9 and band 11.2 of chromosome 22.

The proto-oncogene Abelson murine leukemia (ABL1) gene, located on the band q34 on chromosome 9, translocates to the band q11 on chromosome 22 where the breakpoint cluster gene (BCR) is. The translocation t(9;22)(q34;q11) results in the formation of a fusion gene called BCR-ABL1 and this represents the common cytogenetic abnormality noted in chronic myeloid leukemia (CML), more commonly known as the Philadelphia chromosome.



Karyogram showing chromosomes arranged in homologous pairs. Let us count the total number, it is 45 (46-1). One chromosome is missing. Now start checking each pair to see which is not paired. There is only one X chromosome. The other X chromosome or a Y chromosome is missing. This represents 45,XO which is Monosomy X commonly known as Turner Syndrome.



Let us count the total number first. It shows 47 (46+1). There is one extra chromosome. Now start checking from chromosome 1. Chromosome 13 has 3 copies. Next check the sex chromosomes. Two X chromosomes are present, showing female gender. So this is 47,XX,+13 which represents the karyogram of Trisomy 13 in a girl.

Chromosome Substitution Strains: A New Way to Study Genetically Complex Traits, Annie E. Hill, Eric S. Lander, and Joseph H. Nadeau

ADVANCES IN CYTOGENETICS BEYOND CONVENTIONAL KARYOTYPING: Molecular cytogenetics

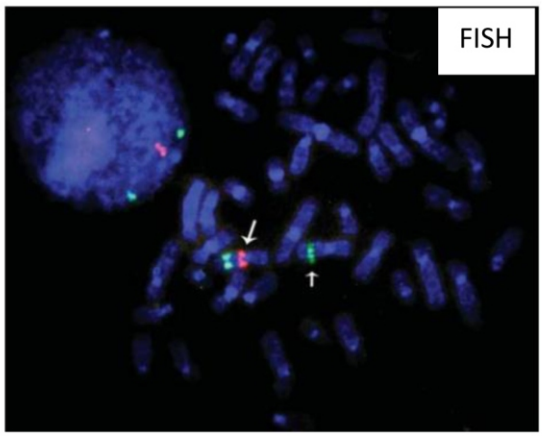
a. **FISH**: Fluorescence In-Situ Hybridization
   The basic elements of FISH are a flourescent DNA probe (purified DNA tagged with flourescent dye) and a target sequence. The interphase or metaphase chromosomes are exposed to a probe which binds to the matching sequence within the chromosomes followed by in-situ hybridization technique to localize the tagged DNA sequences on chromosomes. This technique helps in the diagnosis of common aneuploidies, deletion & duplication syndromes, telomeric rearrangements, and in cancer genetics- for identification of translocations like Philadelphia chromosome.

b. **Spectral Karyotyping** (SKY): painting each pair of chromosomes with different colours.
SKY was introduced in 1996. This is a multi-fluorochrome FISH in which all the chromosome pairs are simultaneously visualized in different colours in a single hybridization. It works on the principle of FISH but uses metaphase chromosomes. This technique helps in the identification of numerous marker and derivative chromosomes. SKY in combination with FISH can increase the accuracy of karyotype interpretation.

c. **Spectral Colour Banding** (SCAN): painting each band of a chromosome with a different colour.
This multicolour banding technique analyses a single chromosome allowing simultaneous visualization of all the bands on a chromosome in different colours in a single hybridization.

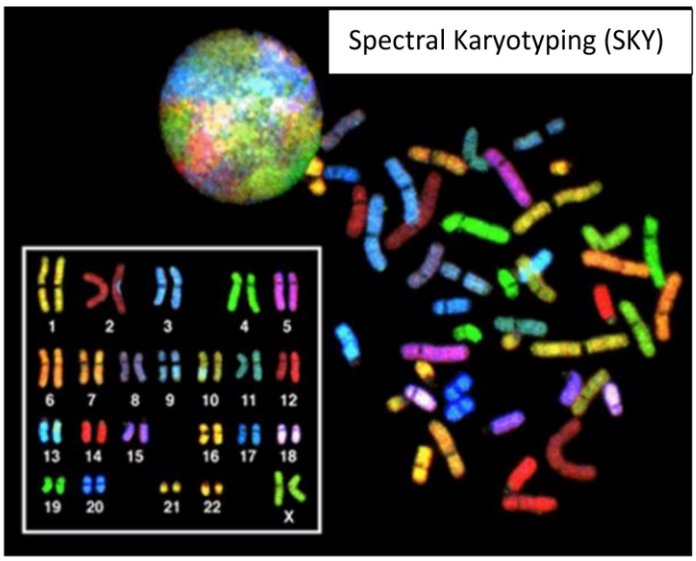
d. **Array Comparative Genomic Hybridization (CGH)/ Microarray**:
This technique is also named molecular karyotyping which is a high-resolution genome-wide screening of copy number variants. Array CGH was introduced in 1998. This genetic test is preferred in the evaluation of children with developmental delay, autism spectrum disorder etc. This also helps in getting detailed information on the extent of deletion or duplication on a particular chromosome as detected in karyotyping.

<u>Pictorial representation of the reports of FISH, SKY and SCAN.</u>

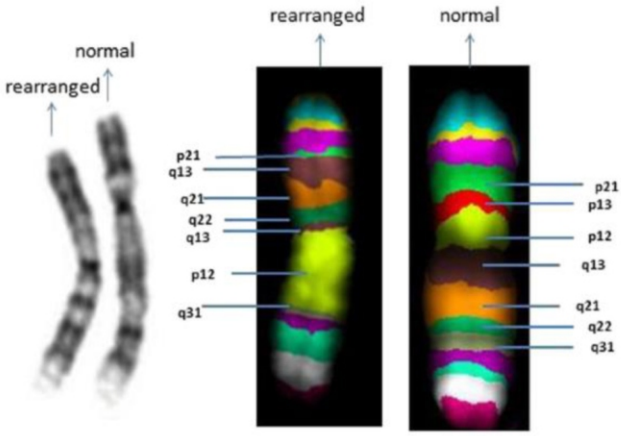


Metaphase after fluorescent in situ hybridization (FISH) with VYSIS-R William Syndrome region. The normal chromosome (long arrow) shows a pink signal at 7q11.23 and a green signal at the control segment. On the abnormal chromosome 7(small arrow), only the control green signal is observed, indicating a deletion at 7q11.23

Souza et al, Genetics and Molecular Biology, 30, 1, 17-20 (2007)



http://genomicsinitiative.com



Spectral Colour Banding

G-banding image of rearranged chromosome 9 (on the left) and of the normal chromosome 9 (on the right) and Multicolour banging image of the rearranged chromosome (on the left) compared to the normal chromosome 9 (on the right) shows bands involved in the rearrangement.

https://www.researchgate.net/publication/287310958_Constitutional_chromothripsis_involving_the_critical_region_of_9q2113_microdeletion_syndrome

TURN-AROUND TIME (TAT):

When will the report be ready? This is one of the most frequently asked questions about genetic testing. The TAT and sample details of the common cytogenetic tests are mentioned below.

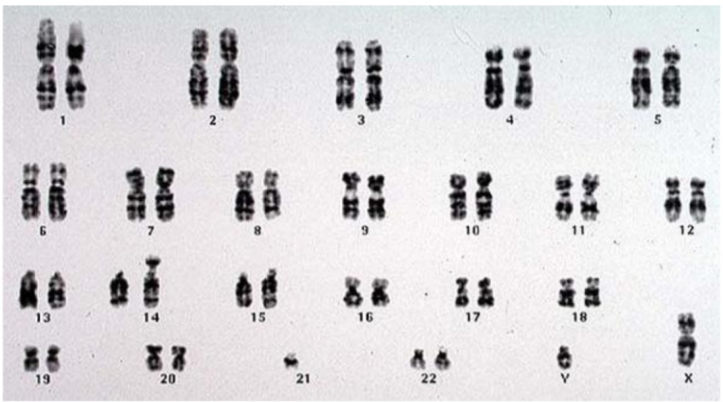
TEST	TAT	SAMPLE REQUIRED
Conventional karyotyping	2-3 weeks	2-3 ml blood in Heparin tube
FISH	5-7 days	Sample from CVS/ Amniocentesis / Skin biopsy
Array CGH	2-4 weeks	2-3 ml blood in EDTA tube

ARE THERE ANY LIMITATIONS OF KARYOTYPING?

Yes, there are limitations.

1. Resolution of karyotyping is limited to around 5Mb. That means chromosomal abnormalities, say deletion or duplication more than 5 Mb (5 million base pairs) size can only be detected by karyotyping. Microarray can be used for detecting smaller deletions/duplications.
2. An actively growing source of cells is needed. Fresh blood sample and immediate processing are needed or else the culture failure may happen.
3. Other limitations:
    - Karyotyping is time-consuming.
    - Requires fresh sample that must be processed immediately on arrival.
    - Not to forget, karyotyping requires a lot of expertise starting from cell culture, harvesting, staining, banding through analysis of the metaphases to reporting.

To conclude, let me suggest that when we come across a child with facial dysmorphism or congenital anomalies, it is better to start the genetic evaluation with karyotyping. If any chromosomal abnormality is detected in the index child, then the karyotyping must be done in the parents also to know if one of them or either carries the same abnormality or is it occurring for the first time in the child; especially with translocations. This is very important for genetic counselling and to assess the risk of recurrence in subsequent pregnancies. Conventional Karyotyping is still a very important test particularly when it comes to the numerical and structural abnormalities of chromosomes even amidst all the advancements in the field of genetics.



Try to interpret this karyogram and mail it to me!

Just to make you go through the numbers and alphabets!

Dr Surya G Krishnan
Consultant in Pediatrics and Pediatric Genetics
Child Central Clinic,
Koramangala 3rd Block, Bangalore - 560034
Email id: suryagk@gmail.com

A Diagnostic Approach to Genetic Disorders

Dr. Sanjeeva G N
Associate Prof. of Pediatrics
Fellow in Clinical Genetics
Indira Gandhi Institute of Child Health, Bangalore

INTRODUCTION

The clinical features of a genetic disorder may manifest at any age, with the majority of them typically presenting in childhood. The estimated prevalence of a genetic disease is 53/1,000 children and young adults. In one large pediatric hospital in the United States, 96% of chronic disorders leading to admission were found having an obvious genetic component or being influenced by genetic susceptibility.

The factors which will help in choosing appropriate genetic tests include category of the genetic disorder suspected, knowledge of underlying molecular abnormality and availability of specific test. This short review gives a brief summary of the diagnostic approach to genetic disorders. This will be useful for the pediatric postgraduate students as well as practicing pediatricians. To keep the discussion precise and simple, this article restricts its discussion to common disorders of mendalian inheritance. Hence discussion on testing of disorders of non-mendelian inheritance, like imprinting disorders, are beyond the scope of this chapter.

Major categories of genetic disorders include single-gene, genomic, chromosomal, and multifactorial conditions.

Chromosomal Disorders

Deletions, duplications, and chromosomal rearrangements that affect whole chromosomes, or large portions of a chromosome, are typically referred to as **chromosomal disorders.** These disorders can be associated with either numerical abnormality or structural abnormalitiesof chromosomes.

Numerical abnormalities involve the loss or gain of one or morechromosomes, referred to as **aneuploidy**, or the addition of oneor more complete haploid complements, known as polyploidy.Loss of a single chromosome results in **monosomy.** Gain of oneor two homologous chromosomes is referred to as **trisomy or tetrasomy,** respectively.One of the most common disorders associated with chromosomal numerical abnormality is Down syndrome, where most common cytogenetic abnormality is presence of an extra copy **(trisomy)** of an entire chromosome 21. A disorder like turner syndrome when all or a part of a chromosome is missing is referred to as **monosomy.**

Structural chromosomal rearrangement in which a genomic region from one chromosome is transferred to a different location on the same chromosome or on a different (nonhomologous) chromosome is referred as **chromosomal translocation.** Translocations can be balanced, meaning that no genetic material has been lost or gained, or they can be unbalanced, in which some genetic material has been deleted or duplicated.

Genomic disorders

Genomic disorders result from the loss or gain of chromosomal/DNA material (copy number variations [CNVs]). There are a number of well-delineated genomic disorders including **deletions** (copy number loss), **duplications** (copy number gain), **inversions** (altered orientation of a genomic region), and **chromosomal rearrangements** (altered location of a genomic region). Some of these genomic disorders may manifest with clinically recognizable distinctive phenotypes, many others do not have a

distinctive pattern of phenotypes but can cause developmental delay, cognitive impairment, structural birth defects, abnormal growth patterns, and changes in physical appearance.

When the deletion that affects 2 or more genes are located near each other on a chromosome, it is called as **Contiguous gene disorders or microdeletion syndromes.** (Eg. DiGeorge syndrome or 22q microdeletion syndrome - deletions of genes located on chromosome 22q11).

Single gene or Monogenic disorders

More than 10,000 single-gene, or monogenic, traits and disorders are known. Most are individually rare, but together they affect between 1% and 2% of the general population at any one time. The diagnosis, investigation, and family management of these disorders represent the major workload challenge in clinical genetics.

The hallmark of a single-gene disorder is that the phenotype is determined by underlying abnormality in an individual gene. There may be individual variation **(variable expressivity)** in the phenotypes associated with single-gene disorders. These phenotypic variations may be due to the severity of the variation affecting the gene and additional modifications caused by genetic, environmental, and stochastic factors. Some identifiable syndromes and diseases can be caused by more than one gene (e.g., Noonan syndrome). This is called **genotypic heterogeneity.** In contrast, mutations affecting a single gene may produce different phenotypes (e.g., FBN1 gene and Marfan syndrome, Acromicric dysplasia, Familial ectopia lentis, Weil Marchesani syndrome type 2, Stiff skin syndrome etc). This is called **phenotypic heterogeneity.**

These monogenic disorders can be confirmed either by directly sequencing that gene **(Sanger sequencing)** or by multiplex ligand dependent probe amplification (MLPA) when looking for intragenesmall deletions and/or duplications. In case of genetic heterogeneity,screening all the disease-causing genes using a next-generation sequencing technology based on disease-specific panel is more efficient and cost-effective than to screen genes individually. When such panels are not available, or when the diagnosis is in not certain, whole exome sequencing (WES) where the protein-coding regions of all genes are screened using next-generation sequencing technology.

When an individual harbors more than one distinct cell populations is referred as **mosaicism.** These cell lines may have a single-gene defect, a genomic disorder, or a chromosomal defect. The natural history of such defects may differ depending on the proportion of each lines.

When to suspect a genetic disease? RED FLAGS

These red flags should prompt the busy clinician to consider a genetic cause. A red flag suggests further action which may be testing, intervention, counseling, follow-up, or referral to a medical geneticist.

Family history

- Multiple affected siblings or individuals in multiple generations.
- Unexplained early neonatal/infant/childhood deaths in the family.
- Advanced parental age (Maternal age $\geqslant$ 35 yr or Paternal age $\geqslant$ 40 yr).
- Repeated pregnancy loss or infertility.

Don't forget that lack of a family history does not rule out genetic causes.

Groups of congenital anomalies. Two or more anomalies (cluster of anomalies) including dysmorphic facies are much more likely to indicate the presence of a syndrome with genetic implications.

- A rule of thumb in genetics is that a patient with two or more minor congenital anomalies warrants additional evaluation to look for a major anomaly that might not be immediately obvious, such as a structural heart or kidney defect.
- Another rule of thumb is that the presence of any other anomaly in an individual with a common, multifactorial malformation (eg, spina bifida, cleft lip, congenital heart disease) should prompt consideration of a genetic syndrome

· **Extreme or exceptional presentation of common conditions.** Early onset of disease or unusually severe reaction to infectious or metabolic stress, for example.

· **Neurodevelopmental delay or degeneration.** Developmental delay, developmental regression in children, or early onset neurologic deterioration in adults.

· **Extreme or exceptional pathology.** Rare tumors or other pathology or multiple primary cancers in one or different tissues, for example.

· **Surprising laboratory values.** Abnormal laboratory values in an otherwise healthy individual; extreme laboratory values for a typical clinical situation.

· **Pregnancy screening abnormality**

- Maternal serum α-fetoprotein
- Maternal 1st-trimester screen
  - § Maternal triple or quad screen or variant of this test
- Fetal ultrasonography
- Noninvasive prenatal testing (NIPT)
- Fetal karyotype

DIAGNOSTIC TESTS

The choice of diagnostic test depends on the category of genetic disorder suspected, underlying molecular abnormality and availability of specific test.

Karyotype

When a genetic disease as a result of chromosomal abnormality (Down syndrome) is suspected, a cyto-genetic technique of chromosomal analysis referred to as **Karyotype** is a simple and cost-effective diagnostic test. Any tissue with living nucleated cells that undergo division can be used for studying human chromosomes. Most commonly, circulating lymphocytes from peripheral blood are used, although samples for chromosomal analysis can be prepared relatively easily using skin, bone marrow, chorionic villi, or cells from amniotic fluid (amniocytes). A karyotype can detect chromosomal aneuploidies, and relatively large chromosomal deletions and duplications (>5Mbp). It is also helpful in identifying inversions and chromosomal translocations. Some of the limitations of karyotype analysis include its requirement of a sample containing fresh viable cells, low sensitivity for the detection of subtle abnormalities and though mosaicism may be detected by routine karyotyping,they can never be 100% excluded.

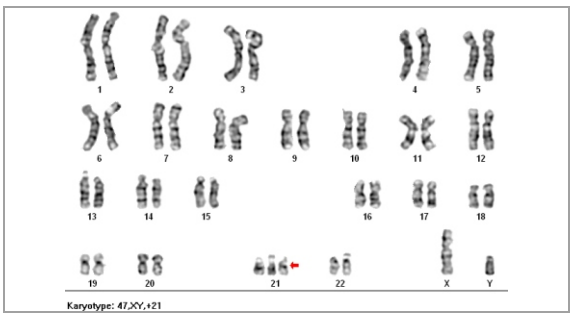


Fig 1: Karyotype of a boy with Down syndrome (47,XY,+21)

Fluorescent in situ hybridization (FISH)

Fluorescent in-situ hybridization (FISH) can provide information about the copy number and location of a specific genomic region. This diagnostic tool combines conventional cytogenetics with molecular genetic technology. It is based on the unique ability of a portion of single-stranded DNA (i.e., a probe) to anneal with its complementary target sequence on a metaphase chromosome, interphase nucleus or extended chromatin fiber. In FISH, the specific DNA probe is labeled with a fluorochrome which, after hybridization with the patient's sample, allows the region where hybridization has occurred to be visualized using a fluorescence microscope. FISH has been widely used for clinical diagnostic purposes during the past 20 years and there are a number of different types of probes that may be employed.

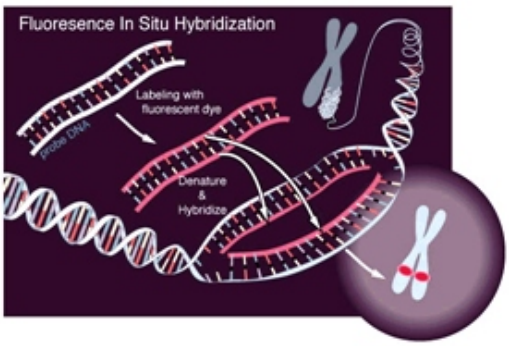


Fig 2: Fluorescent in situ hybridization (FISH)

Centromeric Probes are used for rapid diagnosis of the common aneuploidy syndromes (trisomies 13, 18, 21) from a prenatal diagnostic sample of chorionic villi.It can also be used to screen mosaicism. **Chromosome-Specific Unique-Sequence Probes** are specific for a particular single locus which can be used to identify submicroscopic deletions and duplications causing microdeletion syndromes. **Whole-Chromosome Paint Probes** consist of a cocktail of probes when used together in a single hybridization, the entire relevant chromosome fluoresces (i.e., is 'painted'). Chromosome painting is useful for characterizing complex rearrangements, such as subtle translocations, and for identifying the origin of additional chromosome material, such as small supernumerary markers or rings.

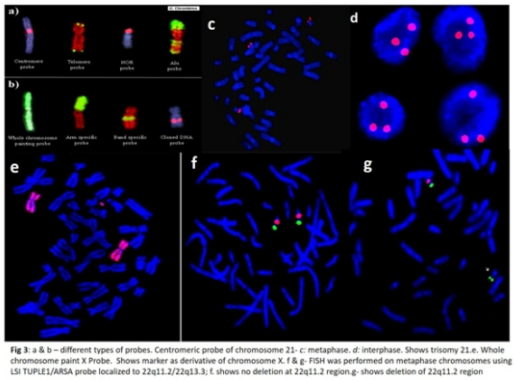


Fig 3: a & b – different types of probes. Centromeric probe of chromosome 21- *c:* metaphase. *d:* interphase. Shows trisomy 21.e. Whole chromosome paint X Probe. Shows marker as derivative of chromosome X. f & g- FISH was performed on metaphase chromosomes using LSI TUPLE1/ARSA probe localized to 22q11.2/22q13.3; f. shows no deletion at 22q11.2 region.g- shows deletion of 22q11.2 region

Sequencing

Direct DNA-based mutation testing detects the specific gene mutation (i.e., sequence change) in single gene disorders. The specific approach used is customized to the biology of the genebeing tested. The target DNA fragment is amplified using PCR and sequenced using the dideoxy chain termination method called as **Sanger sequencing.** This is a gold standard method of mutation screening when a few distinct mutations occur inall affected individuals (eg. Thalassemia).

The major limitations of this method is thatonly DNA that is amplified is sequenced and screening of conditionswhere many possible mutations occur in different individuals, is challenging. Gene sequencing techniques may not be able to identify diseases caused by triplet repeat sequences. Molecular diagnostic tests for such diseases caused by triplet repeat sequences include southern blot and PCR based assays.

Multiplex ligand dependent probe amplification (MLPA)

In single gene disorders, sequencing cannot be used to screen for mutation when intra-gene deletion or duplications are the predominant molecular abnormalities (Duchene Muscular Dystrophy). In such situations, MLPA is the most cost effective molecular diagnostic methods used, especially when knowledge about the gene and disease is available.

Comparative Genomic Hybridization Microarray testing (array CGH)

Array-based copy number detection assays can be used to screen for chromosomal deletions (large and small) and duplications across the genome, but do not provide information about the orientation or location of genomic regions. It can also be used to detectlarge, multigene deletions or duplications (copy number variations). Inaddition, with increasing resolution, single-gene or smaller intragenic deletions or duplications can be detected by aCGH. This approach can detect deletion and duplication mutations as small as several kilobases within 1 gene that would be missed by either chromosome analysis or direct mutation testing by DNA sequencing.

In this molecular-based technique,differentially fluorescent dye-labeled patient's DNA (green fluorophore) and a normal reference DNA (red fluorophore) are mixedin equal amounts and the green to red fluorescence ratio is measured along each tested area.

Interpretation

- Regions of amplification (duplications) - excess of green fluorescence,
- Regions of loss (deletions) - excess red fluorescence.
- Normal - appear yellow.

Advantages of aCGH

- Can test all critical disease-causing regions in the genome at once.
- Can detect duplications and deletions not currently recognized as recurrent disease-causing regions probed by FISH, and detect single-gene and contiguous gene deletion syndromes.
- Does not always require cell culture to generate sufficient DNA

Limitations of aCGH

- Does not detect balanced translocations or inversions
- May not detect low levels of chromosomal mosaicism.

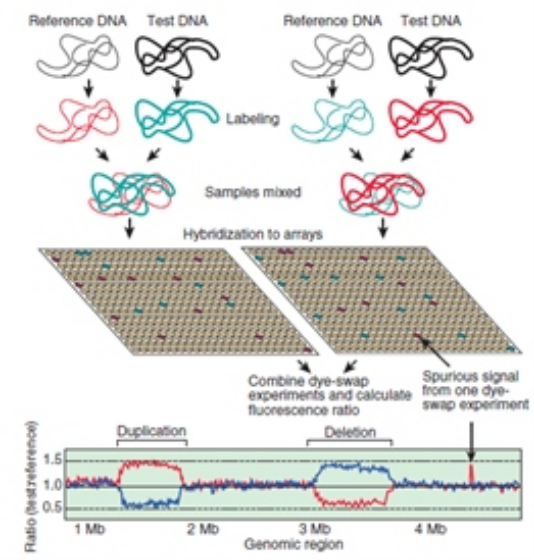


Fig 4: Array comparative genomic hybridization

Exome Sequencing

Among the 3.2 billion bases in the human genome, 1.9% (approximately 60,000,000 bases) are in the coding regions (exons) of almost all the human genes. Recent advances in the technology has allowed for massive, inexpensive DNA sequencing. Hence, now it is feasible to sequence these coding regions. In clinical exome sequencing, >80% of known genes and >90% of genes that have been associated with human disease are sequenced. By comparing with the reference sequence, the variations (an average 35,000 variants or 0.06%) can be detected. Most of these variants include some laboratory and computational errors and are inconsequential polymorphisms and minor polynucleotide repeats near intron/ exon boundaries. But among these 35,000 variants, determining a potential disease-causing variant is challenging.

Certain rules are followed to determining the disease causing variants

1. **Familial segregation** - needs detailed pedigree and clinical phenotype of individual family members
2. **Pathogenicity assessment** - computational estimate of likelihood of a given DNA sequence variant resulting in biological consequences
    a. Nucleotide conservation
    b. Differences in coded amino acids
    c. Filtered using available database
3. **Functional studies** - inferring that the variant alters the function of the gene product.

The major limitations of exome sequencing include inability to detect gene deletions or rearrangements (i.e. structural variants), and also will not detect mutations found within noncoding sequences such as introns or the promoter.

Among the important **ethical considerations,** reporting an incidental finding is a major one. Exome sequencing may identify many medically amenable or not amenable conditions in a patient. Though international guidelines are evolving, currently, a pretest counselling for patients and obtaining consent to report such condition is practiced.

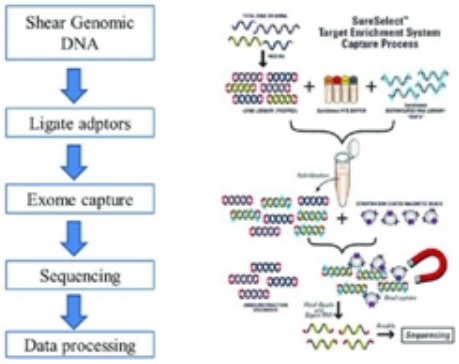


Fig 5 : Basic steps required for exome sequencing

Conclusion

Most frequently asked questions by the families with suspected genetic diseases are

- What does my child have? (diagnosis)
- Why did it happen? (etiology/inheritance)
- What will happen in the future? (natural history; prognosis)
- Is there a treatment? (therapy)
- Could the same thing happen to other family members? (recurrence risk)

To answer all these questions, an accurate diagnosis is important. Newer advances in the technologies has made this task of establishing accurate diagnosis a reality. However, complete evaluation of the child as well as family members, categorization of the disease and appropriate choice of tests are important elements required for confirmation of the clinical condition.

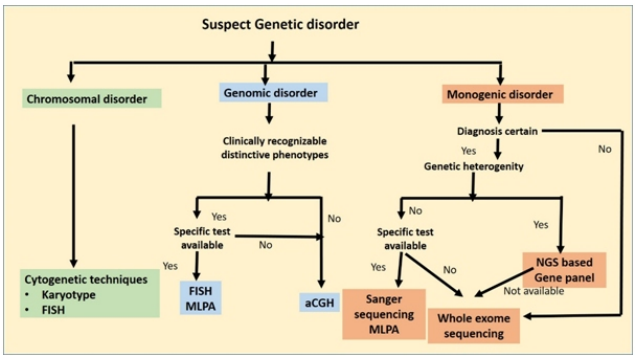


Fig 6 : A diagnostic approach to a child with Suspected genetic disorder

References :

1. Kliegman et al. Nelson textbook of Pediatrics.Edition21.vol1.chapter IX.Philadelphia,2020.
2. Turnpenny P, Ellard S. Emery's elements of Medical Genetics.Edition15.China.2017
3. Whelan A J et al. Genetic red flags: Clues to thinking genetically in primary care practice. Prim Care Clin Office Pract.31 (2004) 497–508
4. Elias Elinati. Human male infertility. 2012 available at https://www.researchgate.net/publication/278643608_Human_genetics_of_male_infertilityyyyyyyyy

Genetic Poetry - Dr. Jeffery Aaron

You’re not a Chromo-Some… You’re Chromo SO-ME

Dating always has a 1st trimester
The view that gets your heart go bolster
Fingers crossed for a womb of love to foster
But look out for the aneuploidic disaster

Well here we are at the marriage trimester
Anamolies rise if you go against her
Make sure you don’t go DOWN that roadster
Because Termination is the ultimate disaster

A kid comes out sooner or later
Tiny throughout in most of the chapter
A perfect smile with a face so tender
Baby you’re not a chromosome you’re my ChromoSoMe ever after
#some

ಅಭಿಸಾರಿಕೆಯ ಅಂತರಂಗ

ಕಾಯುವಿಕೆಯ ರೋಮಾಂಚನದಲು
ಕಣ್ಣಂಚಿನ ಅಶ್ರು ಮೊಳಕೆ
ರಾತ್ರಿ ಪೂರ ಒಂಟಿತನದ ಚಡಪಡಿಕೆ
ಅರ್ಥವಾದೀತೆ ಉಕ್ಕಿಬರುವ ಬಿಕ್ಕಳಿಕೆ

ತಡವಾಗಿ ಬರುವ ಬಿರುಗಾಳಿ ನೀನು
ಬೀಸಿ ಬಂದು ಭರ್ರನೆ ಸುಳಿದು ಮಾಯ
ತಡಕಿ ಹುಡುಕೆ ಒಡಲೆಲ್ಲ ಗಾಯ
ತಾಳೆನಾ ಯಾತನೆ ನೀನೇನು ಅತಿಥಿಯೇನು

ಅವಿಸಿಟ್ಟ ಸಿಟ್ಟು ಸೆಡವು ನಯ ನಾಜೂಕ
ಒಟ್ಟೊಟ್ಟಿ ಕಟ್ಟಿ ಒಗೆದು ಬಿಡುತ್ತೀಯೆ
ಸ್ವರ್ಗ ಸೀಮೆಯ ಸೆರಗ ಮೀರಿ ನಾಕ
ಅದ್ದದ್ದಿ ತೆಗೆದು ಹುಚ್ಚು ಹಿಡಿಸುತ್ತೀಯೆ

ಅರ್ಥವಾದರು ಅರ್ಥವಾಗದ ನಿಗೂಢ ನೀನು
ನಿನ್ನೊಡನೆ ಜಿದ್ದಾಜಿದ್ದಿಯೆ ಮೂಢಮತಿ ನಾನು
ಈ ಗಳಿಗೆ ಗಂಧ ಮಧುರಾನುಭೂತಿ ನಿರಂತರ ಅಮರ
ಇದ್ದೂ ಇಲ್ಲದ ಉಳಿದೂ ಉಳಿಯದ ಅಂತರಾಂತರ

ಬದುಕ ತಿಮಿರದ ಕೊನೆಯ ಬೆಳಕು ನೀನು
ಭಯ ವಿಹ್ವಲ ಅಸಹಾಯ ಅಳುಕು ನಾನು
ಬಾಳ ಬಟ್ಟೆಯಲಿ ತಿರುವುಗಳು ವಿರಳವೇನು
ತೀರದಂಚಿನವರೆಗು ಕೈ ಹಿಡಿದೆನ್ನ ಕರೆದೊಯ್ವೆಯೇನು

ಡಾ- ನಿತ್ಯಾನಂದ ಸುಂಡವಾಳು

IAP BENGALURU TEAM 2023

Dr. S M Prasad
President
IAP Bengaluru 2023

Dr. Sharanagouda S Patil
IAP KSB President

Dr. Rohit Bhandar
IAP KSB Secretary

Dr. Priya Shivalli
IAP KSB VP - BLR

CENTRAL EB MEMBERS

Dr. Adarsh E

Dr. Basavaraja Patil

Dr. Rajendra Salagere

Dr. Karunakara B P

Dr. Sumitha Nayak

Dr. Somashekar A R
President-Elect 2023

Dr. G V Basavaraja
Immediate Past President

Know Your Team

ADVISORS

Dr. Shantharaj A

Dr. Gnanamurthy N

Dr. Geeta Patil

Dr. Chidananda
Secretary

Dr. Harilal Naik M L
Treasurer

Dr. Adarsh E
VP

Dr. Rajashekarmurthy G R
VP

Dr. Nityananda S K
Joint Secretary

EXECUTIVE COMMITTEE

Dr. Chandrshekar M A

Dr. Dhananjaya P

Dr. K Farooq

Dr. Mallesh K

Dr. Maaz Ahmed

Dr. Padma Prakash

Dr. Prashanth M V

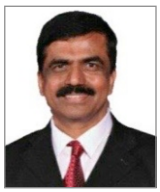
Dr. Ravikumar C P

Dr. Sahana Devdas

Dr. Sowjanya G T

Dr. Kishore Baindur
Historian

Dr. Ravishankar Marpalli
Digital Head



Dr. Bhaskar Shenoy
Accounts Head

Dr. Nandeesh B
Legal Head

Dr. Gunda Srinivas
Digital Co-head

Dr. Sharath Chand
Accounts Co-head

EDITORIAL TEAM

Dr. Nagalatha S
Chief Editor

Dr. Gowri Somayaji
Managing Editor

Dr. Jeffery Aaron

Dr. Naresh Kumar Y

Dr. Poornima

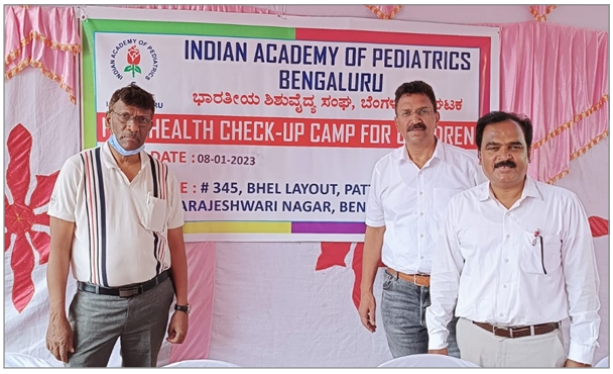
Dr. Shalini Sharma

Dr. Sushma

IAP Bengaluru Activities Report

Dr. Basavaraj delivering a talk on the very first day as President Elect 2023 at installation of office bearers of IAP Telangana twin cities, Hyderabad

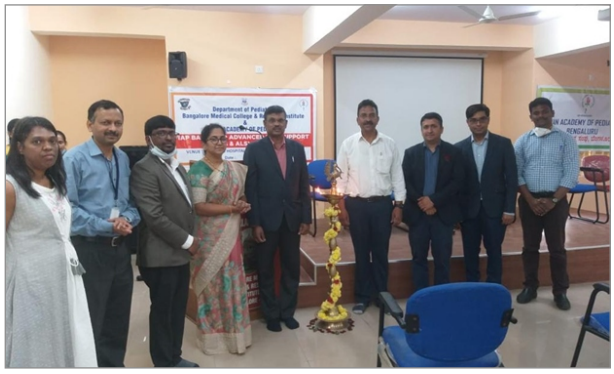
Paramesh Sir delivering talk on Air Pollution
Its implications on Planet at KBNIMS, Kalaburagi



First Health Camp at RR Nagar

BNRP @ RMH 11/01/2023
Basic NRP was conducted at Rangadore Memorial Hospital
Lead Instructor - Dr. Janaki S R (RMH)
Course Co-ordinator - Dr. Naresh Kumar Y (RMH)
Faculty - Dr. Raksha (RMH) and Dr. Veeraraja (IGICH)

Dr. S M Prasad attending National Annual Medical Conference 2022 received the IMA National President Best Adjudged State Secretary Award 2022 Year



Inauguration of PALS at
Department of Pediatrics BMCRI

Excellent Oration by Dr. Nagabhushana for State IAP Installation Ceremony

Upcoming Events

February 2023

1. Tentative scientific Program - Paediatric Nephrology
   Clinical Approach to Enuresis Dr. Saumil Gour, Rainbow Children Hospital, Maruthalli

2. Panel discussion on Common office issues:
   Panel moderator : Prof. Sunil Kumar Msrmth
   Experts : Prof. Premalata, Pediatric Nephrologist, MVJ Medical College, Hoskote
   Dr. Saumil Gour
   Dr. Bhavana, Associate Prof. of Microbiology, IGICH
3. Glimpses from the Past-IAP BPS - Dr. Kishore Baindur
4. Recent updates in Pediatric nephrology Dr. Jagadish Somanna