10.5005/jp-journals-10065-0031

REVIEW ARTICLE

Fanconi Anemia

[1]Richa Saxena, [2]Mampi Debnath, [3]Sudha Chamoli

ABSTRACT

Fanconi anemia (FA) is an infrequently occurring autosomal recessive disorder that is genetically and phenotypically heterogeneous. The main features include myriad of congenital malformations, progressive pancytopenia, and predisposition to both hematologic malignancies as well as solid tumors. Here, in this article, topics such as the association of FA with other syndromes, FA-associated genes and cancer susceptibility, researches in FA, gynecological concerns, management, and diagnostic strategies have been discussed elaborately.

Keywords: Acute myeloid leukemia, Fanconi anemia, Diepoxybutane, Human papillomavirus, Hematopoietic stem cell transplantation, Mitomycin C, Myelodysplastic syndrome.

How to cite this article: Saxena R, Debnath M, Chamoli S. Fanconi Anemia. World J Anemia 2018;2(2):55-60.

Source of support: Nil

Conflict of interest: None

INTRODUCTION

Fanconi anemia (FA) is an infrequently or rarely occurring autosomal recessive disorder that is genetically heterogeneous and is characterized by various kinds of congenital malformations, hematological problems as well as proneness to malignancies that are characterized with multiple genetic mutations and abnormalities, the involvement of numerous organs and numerous forms of cancer risks. It is a lethal disease that is commonly encountered in children (who are around 5-year-old).

It presents with an increased risk of bone marrow failure, congenital anomalies, hematologic [e.g., myelodysplastic syndrome (MDS) and acute myeloid leukemia (AML)] and non-hematologic malignancies (e.g., squamous cell carcinoma).[1-3]

[1]Obstetrician Gynecologist, [2]Postgraduate Student, [3]BDS Graduate

[1]Jaypee Brothers Medical Publishers, New Delhi, India

[2]The MS University of Baroda, Vadodara, Gujrat India

[3]Uttaranchal Dental and Medical Research Institute, Dehradun, Uttarakhand, India

Corresponding Author: Richa Saxena, Obstetrician Gynecologist, Jaypee Brothers Medical Publishers, New Delhi, India, e-mail: synapse94@hotmail.com

CLINICAL FEATURES OF THE PATIENTS WITH FANCONI ANEMIA[4-7]

The characteristics or the clinical features associated with FA have been described Table 1.

Relation with Other Syndromes

Fanconi anemia is classified into the following group of syndromes:[8-10]

- *Chromosomal instability syndromes (by its genetic pathological characteristics):* It includes Ataxia telangiectasia, Nijmegen breakage syndrome, Bloom syndrome, and Werner syndrome. It occurs due to the defects associated with deoxyribonucleic acid (DNA) repair mechanism, elevated cancer risk, and few other changes phenotypically.
- *Inherited bone marrow failure syndromes:* The syndromes related to bone marrow failure are termed as the unsuccessful hematopoietic function of the bone marrow, and they include the following:
  - Shwachman-Diamond syndrome
  - Amegakaryocytic thrombocytopenia
  - Pearson syndrome
  - Dyskeratosis congenita (DKC)
  - Diamond-Blackfan anemia
  - Severe congenital neutropenia.

Genes Involved in Fanconi Anemia

The genome homeostasis is maintained by the FA proteins along with some other proteins. They are involved in DNA repair processes and cell division. Germ-line mutation of any one of the genes that encode FA proteins causes FA.

The mutated genes found in a population with FA, are known as fine needle aspiration cytology FANC genes.

Table 1: Clinical features of Fanconi anemia

• Physical and mental illness
• Failure of bone marrow or pancytopenia
• Elevated risk of malignancies
• Alteration in the functioning of the endocrine hormones
• Diabetes
• Osteoporosis or osteopenia
• Multiple systems or organ dysfunction
• Infertility (in 50% of women with FA, rare in males)
• Complications associated with pregnancy (preeclampsia, premature labor, marrow failure, etc.).

FANCA
FANCB
FANCC
FANCD1 (also known as *BRCA2*)
FANCD2
FANCE
FANCF
FANCG
FANCI
FANCJ
FANCL
FANCM
FANCN
FANCO
FANCP
FANCQ

The genes involved are as shown above.

All the *FANC* genes are autosomal recessive except *FANCB*, as this gene is localized on the X chromosome.

The most frequently occurring mutations FA patients worldwide include *FANCA*, *FANCC*, *FANCG* and *FANCD*.[2] Amongst all, *FANCA* gene abnormalities account for around 60–70% of FA patients and currently more than 100 types of mutations are in the knowledge that are found throughout the *FANCA* gene. Most of these mutations may lead to premature termination or intragenic large deletions, apparently lacking protein expression (null-mutations) and altered proteins showing single amino acid substitution.[11-15]

The major 15 complementation groups of FA and their different corresponding genes have been elaborated along with their chromosomal location in Table 2.[16]

Table 2: Major genes identified for Fanconi anemia

Complementation group	*Gene symbol*	*Proportion of FA attributable to mutations in this gene (%)*	*Chromosomal location*
FA-A	*FANCA*	~60–70	16q24.3
FA-B	*FANCB*	~2	Xp22.31
FA-C	*FANCC*	~14	9p22.3
FA-D1	*BRCA2*	~3	13q12.3
FA-D2	*FANCD2*	~3	3p25.3
FA-E	*FANCE*	~3	6p21.3
FA-F	*FANCF*	~2	11p15
FA-G	*FANCG*	~10	9p13
FA-I	*FANCI*	~1	15q26.1
FA-J	*BRIP1*	~2	17q22
FA-L	*FANCL*	~0.2	2p16.1
FA-M	*FANCM*	~0.2	14q21.3
FA-N	*PALB2*	~0.7	16p12
FA-O	*RAD51C*	~0.2	17q25.1
FA-P	*SLX4*	~0.2	16p13.3

FANCA: A HYPERMUTABLE GENE

An essential protein that is being encoded by the *FANCA* gene is involved in the FA pathway, an important cell process. The pathway of FA plays a distinct role in the specific type of DNA damage which is termed as inter-strand cross-links or ICLs. The product of the *FANCA* gene, i.e., *FANCA* protein, is a constituent of the FA core complex. A process called monoubiquitination leads to activation of *FANCD2* and *FANCI* proteins. The process is carried out by the FA core complex. It then leads to the formation of the *FANCD2-FANCI* (ID) protein complex. This complex then further causes colocalization of the DNA repair proteins to the DNA damage site.

Consequently, rectification of the error occurs and DNA replication further continues.[17]

For genomic stability, the *FANCA-FANCG* interaction is of utmost importance. Maintenance of genomic integrity is done by *FANCA*. It helps in the protection of humans from bone marrow failure and malignancies.

The chief location of *FANCA* is in the nucleus and it shows interaction with cytoplasmic as well as nuclear proteins. There are some research data that suggest that *FANCA* possesses specific cytoplasmic function apart from possessing a nuclear function.

The northern blotting technique has helped recognition of multiple *FANCA* transcripts that proposes the involvement of the *FANCA* gene in the regulation of *FANCA* expression. This is carried out through the mechanism of alternative splicing.

MUTATIONS IN FANCA

FANCA stands out to be the most polymorphic and very heterogeneous FA gene. It differs from FANCC and *FANCG*. FA-A is the most frequently encountered complementation group. Mutation analysis for this can be carried out on genomic DNA as well as on cDNA.

The main reason behind the hypermutability of *FANCA* is the dispersion of a large number of repetitive elements throughout this gene. Repetitive elements comprise Alu-repeats, short direct repeats, CpG-islands mononucleotide tracts, and several hotspot-motifs, for, e.g., CCTG/CAGG. This gene possesses an array of mutations. The most frequently occurring mutation includes large deletions (spanning 1–31 exons). In various reports, there have been discussions regarding the deletions that are spanning entire *FANCA* genes. Some of the splice site

mutations, missense, and nonsense mutations have also been reported. In most of the cases, *FANCA* mutations are termed as "private mutations" because the patients carrying the same mutations and homozygosity are found rarely (except few of the common mutations).

CANCER AND ITS ASSOCIATION WITH FANCONI ANEMIA

Cells those are insufficient in the FA pathway shows characteristics such as impairment in the DNA damage repair system, defective replication of the DNA and missegregation in the chromosome that altogether leads to genomic instability. Finally, it causes a halt in p53-dependent cell cycle and causes bone marrow failure or failure of cell cycle checkpoints. This form of instability may lead to the propagation of mutations and cancer.

Fanconi anemia is considered to be a cancer-prone condition. There is a high risk of vulnerability to both nonhematologic as well as hematologic malignancies in the patients who show FA.

The most frequently occurring clonal abnormalities include the following:

- Duplications and triplications of the long arm chromosome 1
- Addition of portions of the long arm of chromosome 3
- Deletions of long arm of chromosome 5
- Rearrangement of the short arm of chromosome 6
- Monosomy 7 or deletion of portions from long arm of chromosome 7
- Deletions of long arm of chromosome 11
- Gain of chromosome 8 and 21.

Some more studies have reported the existence of a few new cryptic translocations, deletion, and mutations. It included:

- Amplification of NRAS, MLL-PTD, FLT3-ITD, ERG
- Translocation of ZFP36L2-PRDM[16]

But no mutations were encountered for CBL, CEBPα, TP53, TET2, and NPM.11 This disclosed the association of FA and a distinct pattern of genomic abnormalities in FA-related leukemia and MDS.[18]

More studies have identified the presence of four main FA genes namely, FA-D1, FA-J, FA-N and FA-O that are found in case of breast cancer (biallelic or monoallelic).[19-21]

RESEARCHES IN FANCONI ANEMIA

Following are some of the recent researches led by various researchers and scientists:

New Gene Involved in Fanconi Anemia[22]

- Researchers have identified a new gene involved in FA, a rare genetic disease. They studied and discovered specific mutations in the *RFWD3* gene, related to DNA repair, which is involved in the development of this disorder.
- During the study, the use of next-generation massive sequencing technology is done by researchers.
- The author of the research noticed mutations in the *RFWD3* gene in a child with FA and confirmed the relationship of mutation and disorder with functional studies in cell and animal models.
- Earlier, there was involvement of 21 genes in FA which is directed by a study that gave a way to the discovery of another of the genes causing this disorder, the *FANCQ*.
- The discovery of new genes is necessary for genetic diagnosis and advice, but also for the development of new therapies, e.g., gene therapies.
- The involvement of *RFWD*^{3} protein is of the few deficient proteins in patients with FA in which enzymatic activity (ubiquitin ligase) is noticed that is helpful in massive drug screenings.

Linkage of Genetic Breakdown in Fanconi Anemia to HPV-associated Cancer[23]

A genetic malfunction which causes DNA instability in a patient with FAmay put them at high risk for squamous cell carcinomas linked to human papillomavirus (HPV). The following points are an important part of the study done by the researcher:

- A cascade of cellular abnormalities is triggered by the breakdown of a cell signaling pathway for the FA gene complex. This is then worsened by HPV cancer genes present in the skin cells and finally results in developing squamous cell carcinoma solid tumors.
- Restoration of the FA pathway leads to reversal of the irregular cell growth in HPV positive cells which may lead to a reduction of the risk of cancer.
- The blood disorder can be cured by a bone marrow transplant, but those patients who are with FA persist at the risk of solid tumors. The squamous cell carcinoma is most common in areas of head, neck, and skin or anogenital region. Surgery or radiation is mainly the current treatment for these cancers.
- The HPV cancer genes E6/E7 were artificially introduced to *FANCA*-patient skin cells and grown in the three-dimensional organotypic raft culture that stimulates the skin.
- Restoration of *FANCA* gene function in the skin helps in the reduction of skin non-uniform cell growth and thickness of the skin. However, in a reverse experiment, FA gene pathway is not broken.

DIAGNOSTIC STRATEGIES FOR FANCONI ANEMIA

Laboratory Testing

Chromosomal Fragility Testing in T-lymphocytes

This testing was illustrated by Cervenka et al. in 1981 and Auerbach in 1993 that encompassed usage of clastogenic agents, such as mitomycin C (MMC) and diepoxybutane (DEB), respectively.[24,25] Main purpose of this method is to challenge the hyposensitive FA cells in the cell culture (most frequently T-lymphocytes from peripheral blood) that are exposed to DEB and MMC and then to analyze the chromosomal aberration, breaks, rearrangements (radials exchanges). This procedure helps in distinguishing between FA cells and non-FA cells. This is labeled as the "gold standard" method in comparison to the other testing methods in the diagnosis of FA, due to it's easy, reliable, manageable, reproducible, and sensitive nature.

Skin Fibroblast Testing

It is done for the suspected cases in somatic mosaicism. Mosaicism is general in patients with FA, i.e., one shows an elevated sensitivity to MMC/DEB and the other one shows standard levels of chromosomal breakage in response to MMC/DEB.

Fanconi Anemia Complementation Group and its Determination

On the basis of specific gene defects that lead to FA, this testing is used for classification of the patients with FA. One of the chief principles of this testing is to infect FA cells that have been tested with retrovirus to contain a cDNA from a *FA* gene. This is solely for the identification of specific *FA* complementation group for the consecutive DNA sequencing of gene mutations.[26]

Analysis of the Mutation

This is done for identification of the specific gene mutations from the proband after it is being confirmed by the primary complementation group result.

This is then followed by sequence analysis that is used for all the known genes associated with *FA*. For detection of the deletions of one or more than one exons or deletion of an entire gene of any suspected case of *FA*, duplication or deletion analysis is performed.

MANAGEMENT OF FANCONI ANEMIA

According to the International Fanconi Anemia Registry, 73% of patients with *FA* develop overt bone marrow disease by age of 10 years with a median survival of 7 years. In children, hearing and development need to be evaluated and kidneys and urinary tracts should be examined by ultrasound. Androgen therapy results in the improvement of blood count in half of FA patient treated.

Hematopoietic stem cell transplantation (HSCT) can be curative for hematological symptoms using matched related sibling bone marrow, cord blood or even unrelated donors for transplantation. According to a large International Bone Marrow Transplant Registry, 2-year survival probabilities were 66% after human leukocyte antigen (HLA)-matched sibling hematopoietic stem cell transplantation. Those patients, who show sensitivity to chemotherapy and radiation, are administered with lowered doses for conditioning.

However, the patient still remains at risk for head and neck carcinomas starting 5 years after transplantation and may still develop myelodysplasia, acute myeloid leukemia, or solid tumors. Investigation for early detection of malignancies must be started at an early stage; but there should be avoidance of repeated radiographic scans. Examinations like dental and oropharyngeal check-ups, endoscopy and gynecological examination with Pap smear and rectal examination. For patients showing up with *FANCF*-mutated and *FANCB*-mutated tumors may be given treatment with crosslinking chemotherapeutic agents, like cisplastin.[27-29]

GYNECOLOGICAL AND REPRODUCTIVE CONCERNS IN FANCONI ANEMIA

Girls and women, who are with FA, encounter some standard problems such as abnormalities in puberty development and menstrual cycle irregularities.

Delayed Puberty

- It may be associated with additional growth delay
- No breast bud development by age 13
- The following can be the reasons behind pubertal delay:
  - Hormonal imbalance (hypothalamic dysfunction)
  - Low-body weight and chronic illness.

Menstrual Abnormalities

- Heavy vaginal bleeding
- Irregular vaginal spotting (may associate with hormonal or ovulatory dysfunction).

Recommendations for Preventive Gynecological Care

- Consultation with a gynecologist who has the proper knowledge and is aware of FA-associated concerns and discusses potentially sensitive issues:
  - Are you sexually active
  - Discusses contraception

  - Discusses the risk associated with sexually transmitted diseases (STDs).
- Physical examination which includes evaluation of vulva, vagina and cervix.
- Recommendation for annually STDs testing until the age of 25, i.e., testing for Chlamydia and Gonorrhea.
- Regular use of condoms for protection against STDs.

Treatment

The following are the treatment options:

- *Hormonal management:*
  - Birth control pills
  - Leuprolide acetate
  - Progestins
- *Surgical options:*
  - Hysterectomy
  - Endometrial ablation.

ASSOCIATION OF FANCONI ANEMIA WITH AN INCREASED LEVEL OF NUCHAL TRANSLUCENCY[30]

Fanconi anemia may be indicated by a heightened level of nuchal translucency in the first trimester in combination with some other significant abnormalities in the second trimester. Although there might be chances that the karyotype is normal in such cases, but the elevated risk of rare genetic syndromes should instigate the contemplation of testing for chromosomal breakage with MMC and DEB and for detection of syndromes such as FA.

CONCLUSION

Gene therapy studies are underways,[31] as are clinical trials of improved treatment for malignancies associated with FA. FA should be considered in all young adults and children who are with aplastic or hypoplastic anemia, unexplained macrocytes, myelodysplastic syndromes, acute myelogenous leukemia and epithelial malignancies with or without characteristic physical anomalies.

REFERENCES

1. D'Andrea AD, Grompe M. The Fanconianaemia/BRCA pathway. Nat Rev Cancer 2003;3:23-34.
2. Young NS, Alter BP. Aplastic Anemia: Acquired and Inherited. Philadelphia PA: WB Saunders; 1994. pp. 100-132.
3. Alter BP. Inherited bone marrow failure syndromes. In: Nathan DG, Orkin SH, Look AT, et al. (Eds). Nathan and Oski's Hematology of Infancy and Childhood, 6th edition. Philadelphia, PA: WB Saunders; 2003:280-365.
4. Saxon B. Nathan and Oski's Hematology of infancy and children. J Paediatr Child Health 2004;40:244.
5. Butturini A, Gale RP, Verlander PC, et al. Hematologic abnormalities in Fanconi anemia: an International Fanconi Anemia Registry study. Blood 1994;84:1650-1655.
6. Wajnrajch MP, Gertner JM, Huma Z, Popovic J, Lin K, Verlander PC, et al. Evaluation of growth and hormonal status in patients referred to the International Fanconi Anemia Registry. Pediatrics-English Edition 2001 Apr 1;107(4):744-754.
7. Shimamura A, Alter BP. Pathophysiology and management of inherited bone marrow failure syndromes. Blood Rev 2010;24:101-122.
8. Taylor AM. Chromosome instability syndromes. Best Pract Res Clin Haematol 2001;14:631-644.
9. Auerbach AD. Fanconi anemia and its diagnosis. Mutat Res 2009;668:4-10.
10. Dokal I. Fanconi's anaemia and related bone marrow failure syndromes. Br Med Bull 2006;77-78:37-53.
11. de Winter JP, Joenje H. The genetic and molecular basis of Fanconi anemia. Mutat Res. 2009;668:11-19.
12. Vaz F, Hanenberg H, Schuster B, Barker K, Wiek C, Erven V, et al. Mutation of the RAD51C gene in a Fanconi anemia–like disorder. Nature Genetics 2010 May;42(5):406-409.
13. Kim Y, Lach FP, Desetty R, Hanenberg H, Auerbach AD, Smogorzewska A. Mutations of the SLX4 gene in Fanconi anemia. Nature Genetics 2011 Feb;43(2):142-146.
14. Bogliolo M, Schuster B, Stoepker C, Derkunt B, Su Y, Raams A, Trujillo JP, Minguillón J, Ramírez MJ, Pujol R, Casado JA. Mutations in ERCC4, encoding the DNA-repair endonuclease XPF, cause Fanconi anemia. The American Journal of Human Genetics 2013 May 2;92(5):800-806.
15. Ten Foe JR, Rooimans MA, Bosnoyan-Collins L, Alon N, Wijker M, Parker L, et al. Expression cloning of a cDNA for the major Fanconi anaemia gene, FAA. Nature Genetics 1996 Nov;14(3):320-323.
16. The Rockefeller University. Fanconi Anemia Mutation Database. 2019. [online] Available from http://www2.rockefeller.edu/fanconi/ [Last accessed, March 2019].
17. Nakanishi K, Taniguchi T, Ranganathan V, New HV, Moreau LA, Stotsky M, et al. Interaction of FANCD2 and NBS1 in the DNA damage response. Nature Cell Biology 2002 Dec;4(12):913-920.
18. Quentin S, Cuccuini W, Ceccaldi R, Nibourel O, Pondarre C, Pagès MP, et al. Myelodysplasia and leukemia of Fanconi anemia are associated with a specific pattern of genomic abnormalities that includes cryptic RUNX1/AML1 lesions. Blood 2011 Apr 14;117(15):e161-170.
19. Rosenberg PS, Alter BP, Ebell W. Cancer risks in Fanconi anemia: findings from the German Fanconi Anemia Registry. Haematologica 2008;93:511-517.
20. Howlett NG, Taniguchi T, Olson S, Cox B, Waisfisz Q, de Die-Smulders C, et al. Biallelic inactivation of BRCA2 in Fanconi anemia. Science. 2002 Jul 26;297(5581):606-609.
21. Zhang F, Ma J, Wu J, Ye L, Cai H, Xia B, et al. PALB2 links BRCA1 and BRCA2 in the DNA-damage response. Current Biology 2009 Mar 24;19(6):524-529.
22. UAB. New gene causing Fanconi anemia identified. [online] Available from http://www.uab.cat/web/newsroom/news-detail/new-gene-causing-fanconi-anemia-identified--1345668003610.html?noticiaid=1345733384352/ [Last Accessed March, 2019].
23. Cincinnati Children's. Best Children's Hospitals. 2018. [online] Available from http://www.cincinnatichildrens.org/ [Last Accessed March, 2019].
24. Yamashita T, Wu N, Kupfer G, Corless C, Joenje H, Grompe M, et al. Clinical variability of Fanconi anemia (type C) results from expression of an amino terminal truncated Fanconi

anemia complementation group C polypeptide with partial activity. Blood 1996 May 15;87(10):4424-4432.
25. Cervenka J, Arthur D, Yasis C. Mitomycin C test for diagnostic differentiation of idiopathic aplastic anemia and Fanconi anemia. Pediatrics 1981 Jan 1;67(1):119-127.
26. Chandra S, Levran O, Jurickova I, Maas C, Kapur R, Schindler D, et al. A rapid method for retrovirus-mediated identification of complementation groups in Fanconi anemia patients. Molecular Therapy 2005 Nov 1;12(5):976-984.
27. Alter BP. Inherited bone marrow failure syndromes. In: Nathan DG, Oski SH, Ginsburg D, Look T. Hematology of Infancy and Childhood, 6th edition. Philadelphia: Harcourt Health Sciences; 2003. pp. 280-365.
28. Taniguchi T, D'Andrea AD. The molecular pathogenesis of Fanconi anemia: Recent progress. Blood 2006;107:4223-4233.
29. Rosenberg PS, Huang Y, Alter BP. Individualized risk of first adverse events in patients with Fanconi anemia. Blood 2004;104:350-355.
30. Tercanli S, Miny P, Siebert MS, Hösli I, Surbek DV, Holzgreve W. Fanconi anemia associated with increased nuchal translucency detected by first-trimester ultrasound. Ultrasound in Obstetrics and Gynecology 2001 Feb 1;17(2):160-162.
31. Cohen-Haguenauer O, Péault B, Bauche C, Daniel MT, Casal I, Levy V, et al. In vivo repopulation ability of genetically corrected bone marrow cells from Fanconi anemia patients. Proceedings of the National Academy of Sciences 2006 Feb 14;103(7):2340-2345.