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UNDERSTANDING DIVERSE SIGNALING PATHWAYS INVOLVED IN COLORECTAL CARCINOMA AND POTENTIALS OF NANOTECHNOLOGY IN THEIR DIAGNOSIS AND THERAPEUTICS

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ABSTRACT

Colorectal carcinogenesis is among the 3rd most significant primary reasons for fatality. Colorectal cancer (CRC) growth and development were linked to a variety of underlying pathways. It progresses from invasive cancer to adenomatous polyps via genetic and epigenetic mutations. Various risk factors, including genetic and ecological, play a vital part in progression. Colorectal carcinoma develops in stages, starting from the adenomatous polyps (benign type) in the large intestine and rectum, particularly on the inner surface. It later progressed to complex adenomatous cancer in situ along with invasive cancer. Even though recent studies have focus attention on the pathogenesis of colorectal carcinogenesis and provided improved diagnostic strategies, the fraction of CRC is still on the surge. Numerous signal transduction pathways were found to be uncontrolled in CRC, resulting in the initiation of pernicious phenotypes. Elaborating the pathogenesis of CRC progression and pharmacotherapy, it is imperative to analyze signaling pathways associated with CRC metastasis. The biomarker is a compound found in samples including blood, stool, and tissues used to diagnose pathological medical conditions. The latest evidence has focused on advancing these biomarkers as a tool for the prior detection of biomarkers to aid in the early detection and predictive stratification of CRC. As a result, extensive attempts have been made to target signaling pathways to develop effective treatment plans. The signaling pathway implicated in the pathogenesis and advancement of colorectal cancer is discussed in this paper. The various invasive and noninvasive colorectal cancer screening tests and molecular biomarkers involved in colorectal cancer are explored. In addition, the management of the CRC through novel drug delivery systems are also addressed.

Keywords: *Colorectal cancer, screening test, biomarkers, signaling pathway*

INTRODUCTION

Carcinoma is an uncontrollable cell growth that can affect any organ in the body. Human tumors are defined in layman's terms as abnormal growth that impacts the surrounding tissues.[1] According to World Health Organization data from 2020, colorectal carcinoma contribution is around 10% of the total cancer cases as per the report.[2] Among them, the top-ranked cancers in males include liver, prostate, colorectal, stomach etc, while, in the case of females, thyroid, cervices, mammary, lungs, and colorectal are predominant. A healthier lifestyle and a change in habits could prevent nearly 30 percent of cancer deaths. A series of genetic and epigenetic changes are responsible for the development of colorectal cancer.[3] The vast majority of CRCs arise from adenomas (adenoma-carcinoma sequence), and the neoplastic conversion time is estimated to be around 10-15 years, indicating the appropriate time to assess and eradicate these adenomas before they evolve into invasive carcinoma. The progression of tiny polyps results in CRC formation, which may appear harmless at first but grows uncontrollably and solidly over time (Figure 1).

Cancer mortality is exceptionally high in Asian and African populations. It is well understood that dietary habits influence the occurrence of CRC. Fiber-rich, low-fat diets have been shown to reduce the risk of CRC.[4] The food we eat has an impact on our health. Fatty foods, processed meats, and processed foods are all linked to an increased risk of CRC. The cornerstones of CRC therapies are surgery, targeted therapy, neoadjuvant radiotherapy, and adjuvant chemotherapy. Novel biomarkers are required for CRC diagnosis and treatment.[5] As CRC progresses, numerous signaling pathways, including TGF-β, EGFR, EMT, Wnt, MAPK, and others, are downregulated. Epidermal growth factor receptor (EGFR) a type of tyrosine kinase receptor linked to cell membrane receptors of the ErbB family.[6] These EGFRs were important in antitumor targeted therapies. HER2 (Erb-2), HER3 (Erb-3), HER4 (Erb-4) are among the other receptors of the Epidermal growth factor receptor family. Such receptors have a ligand-restricting area on the cell surface, EGFR has an atomic load of 170kDa, and its activity is prolonged due to receptor over-articulation and co-articulation with its ligands.[7] The variety of cellular processes such as cell apoptosis, differentiation, invasion, and cell growth arrest have the indolent of TGF-β. TGF-β promotes invasion and migration of carcinoma by epithelial-mesenchymal transition induction. As a result, TGF-β inhibition is considered a possible restorative alternative for numerous cancer types, including CRC.[8] The Wnt pathway, on the other hand, has been linked to the progression of CRC through both canonical and non-canonical pathways. The canonical pathway is essential in the β-catenin dependent pathway, but it does not affect the non-canonical pathway.[9] The synthetic/herbal medicines such as quercetin, isoquercetin, anthocyanin, genistein, epigallocatechin-3-gallate (EGCG), gallic acid, gllagic acid, resveratrol, curcumin, gefitinib, erlotinib, cetuximab, panitumumab, ursolic acid, sulforaphane are used to treat CRC. This review deals with multiple signaling pathways and their effect on colorectal carcinoma. It also highlights various delivery systems for the management of CRC.

EPIDEMIOLOGY

The incidence and mortality rates of CRC vary considerably across countries. According to the World Health Organization's GLOBOCAN database, CRC is the third most widely diagnosed carcinoma in men and the third most common carcinoma in worldwide.[10] Males have significantly greater rates of both incidence and mortality than females. In 2020, there are expected to be 147,950 new cases of CRC in the United States, including 104,610 cases of colon cancer and 43,340 cases of rectal cancer. Whereas the significant proportion of these eventuate in people over the age of 50 years and older, 17,930 new cases of CRC (12%) will be diagnosed in people under the age of 50 years (Figure 2). Furthermore, there will be a projected 53,200 CRC deaths in 2020, with 3640 decedents (7 percent) being under the age of 50.[11]

The prevailing incidence rates are in Australia and New Zealand, Europe, and North America, and the lowermost rates are originated in Africa and South-Central Asia. These regional variations appear to be due to differences in food and environmental exposures imposed on a backdrop of genetically determined vulnerability.[12] Since the mid-1980s, mortality rates from CRC have steadily fallen in the United States and most other Western countries. This improved prognosis can be ascribed, at least in part, to the diagnosis and eradication of colonic adenoma, the timely diagnosis of CRCs, and more appropriate treatment and adjuvant therapies. Conversely, at least in the United States, the reduction in CRC mortality began long before comprehensive CRC screening and effective adjuvant therapy became generally available.[13]

ESSENTIAL CONSEQUENCES OF MOLECULAR PATHWAYS

Multiple and sequential genetic alterations should arise for melanoma to establish, regardless of the specific pathway.[14] Also, every genetic alteration creates a new clone, with a 'successful cancer' necessitating up to ten clonal events, each characterized by a relative density advantage.[15] These pre-cancerous cells should create a cellular interface that helps for future genetic and probably epigenetic events, resulting in genomic and epigenomic instability. Genomic instability is crucial in oncogenesis because it ensures that consequent events occur with increasing probability.[16] It hastens metastatic progression by raising the mutation rate triggered by the background mutagenic obstacle. Without genomic instability, the accumulation of new mutations would be far too inefficient for melanoma to establish during a human lifetime.[17] Diverse range of molecular pathways has been identified for colorectal cancer (Figure 3) and their therapeutic agents and targets (Table 1) these include Wnt/β-catenin, EGFR, TGF-β, P53, EMT pathways etc.

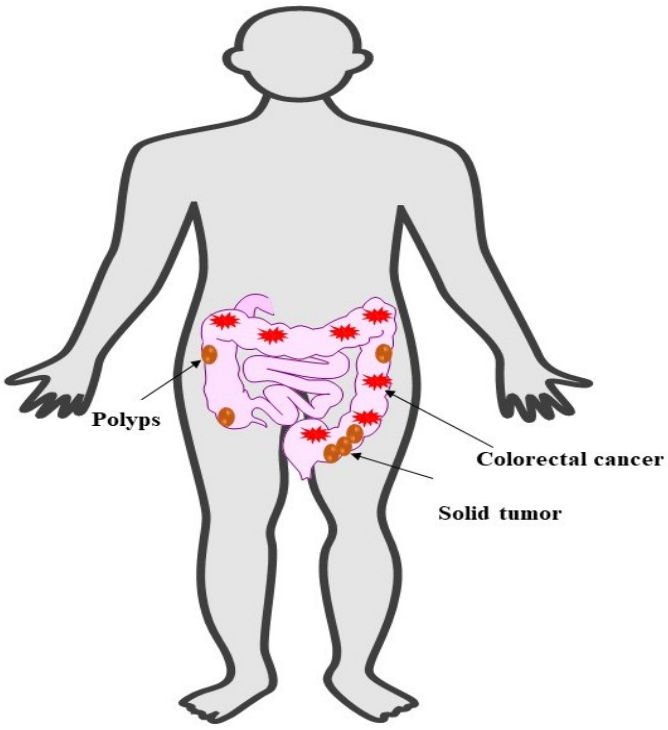

Fig 1: Progression of CRC in the human body

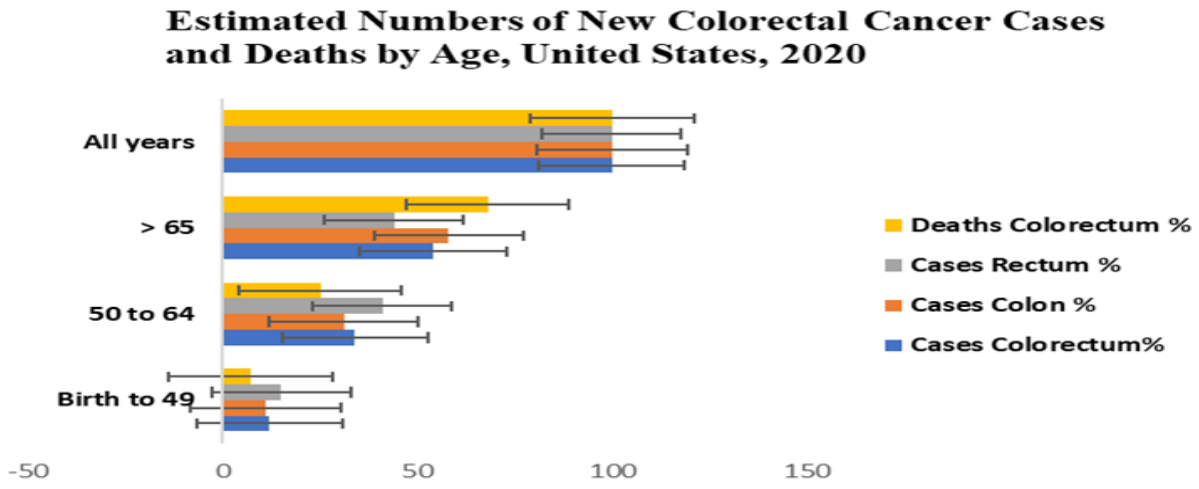

Figure 2: Estimated number of new colorectal cancer cases and deaths by age in the United States, 2020

Wnt/β-catenin signaling pathway

The Wnt/β-catenin signaling pathway is categorized into two stages: on-stage and off-stage. According to the Wnt off state region, cytoplasmic catenin is phosphorylated by a destructive complex made up of Axin, APC, GSKβ3, and CK1, after which it is ubiquitinated and focused for proteasome degradation. Whereas Wnt on state consists of Wnt ligands binding to its receptor, Dvl establishes the devastation of the catenin destruction complex, which provokes β-catenin stability. Initiating the Wnt target genes, β-catenin is relocated to the nucleus as a cofactor of TCF/LEF.[18] The eGFR signaling mechanism starts with ligand interaction, which induces the eGFR to dimerize and activate. Following autophosphorylation of tyrosine residues, downstream signaling is activated. The activation of Ras (also known as KRas) leads to the stimulation of Raf protein (i.e; B-Raf), hence phosphorylation of MEK and MAPK takes place in the cell nucleus. Cell proliferation, differentiation, and sustainability are thought to be controlled by the Ras-Raf-MAPK signaling pathway.[19]

Table 1: Therapeutic agent for multiple signaling pathways

Pathways	Therapeutic agents	Targets
Wnt/β-catenin signaling pathway	BC2059	β-catenin
	PNU-74654	Wnt/β-catenin
	Sulindac	Dishevelled
EGFR signaling pathway	Cetuximab	EGFR
	Sorafenib	RAF
	Erlotinib	EGFR
	Pertuzumab	HER2
TGF-β signaling pathway	LY2109761	TGFBRI/II
	AP11014	TGF-β
	AP15012	TGF-β1
P53 signaling pathway	CTM	R175H
	ZMC1	R175H
	PEITC	R175H
EMT signaling pathway	Emodin	CK2alpha
	Sorafenib	SHP1
	Gefitinib	EGFR
	Cisplatin	EGFR/HDAC
	Galunisertib	TGFβRI

EGFR signaling pathway

The PI3K-AKT pathway is another axis of the EGFR signaling cascade that is crucial in colorectal carcinogenesis. PI3K is transferred to the cellular membranes and adheres to tyrosine phosphate after the EFGR tyrosine residues are phosphorylated.[20] PI3K then promotes the activation of AKT. The presence of activated AKT (p-AKT) in the cytoplasm then stimulates multiple targets, leading to cell development, multiplication, and preservation.[21]

TGF-β signaling pathway

TGF pathway activation binds with Type I and Type II TGF receptors (known as TβRI and TβRII, respectively). Another step in the phosphorylation of the canonical TGF pathway leads to activating Smad proteins (i.e, R-Smads: Smad2 and Smad3) to the active TβRI. The Smad proteins (i.e, Smad2 and Smad3) binds with the TβRI receptor and then phosphorylates the receptor complexes before interrelating with co-Smad (i.e, Smad4). Thus, the clusters of Smad protein (i.e; Smad2/3/4) enters to the nucleus, where it unites DNA and regulates the appearance of many genes in collaboration with cofactors.[22]

p53 pathway

The p53 pathway is a crucial transcription feature that plays a vital role in tumor development and growth suppression via signaling factors. Because of its importance in maintaining DNA integrity, it can be attributed to as the "Guardian of the Genome," but p53 signaling is frequently liberalized in colorectal cancer.[23] Under standard homeostatic conditions, p53 interaction in cells is suppressed by its negative regulators (MDM2 and MDM4), which together elicit ubiquitination and proteosomal degradation of p53 and keep cellular intensities of the tumor suppressor at ineffectively reduced numbers. Subsequent DNA smash-up, p53 binds unswervingly to the CDKN1A locus and stimulates CDKN1A, PANDA, and LincRNA-p21 transcription. PANDA suppresses NF-YA to prevent cell death, and LincRNA-p21 promotes apoptosis by mediating gene silencing via hnRPK recruitment. p53 induced eRNAs are responsible for activating p53-target genes and regulating cell cycle arrest caused by p53. LncRNA-LOC285194 is a 2 kb long RNA found at chr3q13.31 that has prevalent focal copy number alterations (CNAs) and loss of heterozygosity (LOH) in primary osteosarcoma specimens and cell lines from other cancers.[24]

EMT pathway

The initiation of the signaling pathway controls the responses of EMT program. The EMT program can also provoke through various signaling pathways such as TGF-β, Wnt, and receptor tyrosine kinases (RTKs), and relying on the cellular environment, the importance of each of these may differ. The phosphorylation of Smad proteins takes place via interaction with TGF-β, and therefore stimulates Smad2 and Smad3 to the nucleus with Smad4 to transactivate SNAI1 expression.[25] The major role of Smad4 act as a negative regulator in EMT pathway is that activation of STAT3 is inhibited by Smad4, which directly relates to EMT and ZEB1 expression. Whereas RAF, Raf-1 kinase inhibitor protein (RKIP) acts as a negative regulator. Apart from this, another negative regulator (i.e; PTEN) inhibits the PI3K signaling pathway. Cells of the immune system, carcinoma-associated fibrocytes (CAF), stem cells, endothelial cells, hypoxia, soluble factors, signaling molecules, and extracellular matrix components are all involved in forming the tumor. IL-6 is a key cytokine in the microenvironment. The IL-6/IL-6R/STAT3/miR-34a feedback loop leads to CRC proliferation in the context of EMT.[26]

SCREENING TEST AND MOLECULAR MARKERS OF COLORECTAL CANCER

Screening is a method for the identification of carcinoma, which is based on symptomatic criteria. This can aid in the detection of cancer at an initial point. It could be possible to diagnose cancerous tissues if it is discovered early.[27] Tumor may have propagated by the time symptoms appear. Colorectal cancer is frequently preventable via routine screening, which detects polyps until they become cancerous. Numerous screening tests (table 2), both noninvasive and invasive, are generally used to screen for CRC.[28] Among them, multi-targeted fecal DNA (FIT-DNA), Guaiac fecal occult blood test, (gFOBT), Fecal immunochemistry test (FIT), CT colonoscopy, sigmoidoscopy, and SEPT9 test are being commercially employed.[29] In practical use gFORT is not being utilized to detect advanced colorectal cancer because of sensitivity issues. Quantitative

matching is the most widely used method for the screening of the CRC. gFOBT helps to detect the blood in the fecal sample using guaiac as a chemical.[30] In the case of the FIT test the antibodies are used to detect blood in a fecal sample.

As a combination test FIT-DNA is used to detect the DNA changes in a stool sample.[31] The rectal and sigmoid colon are being investigated and screened for abdominal-related problems including polyps, through a sigmoidoscope. An instrument known as a sigmoidoscope is introduced intra-rectally for this purpose. This instrument has a small light attached to it to assist the screening with the attachment for removing tissues section and polyps, which can further be examined for malignancy.[32] Colonoscopy is done by inserting it in the rectum to carry out the screening and examination of rectum and colon for malignancy and other ailments.[33] Despite this there is various nanotechnology-based detection method for the treatment of colorectal cancer, the brief outlook of these methods explained in table 3.

In the present scenario, the CRC disease pathogenesis can well be understood with progressive molecular and biological features. With this advancement, it has now become possible to create biomarkers for the identifications of tumor management, diagnosis and surveillance. This biomarkers rationalization can be further utilized to detect and select the medicines to prevent and reduce the death rate.[34] A better CRC biomarker must be quantitatively necessary to assess, intensely accurate, selective, and provable. The biomarkers must be equipped with the quality to differentiate between risk-based patients and patients with the requirement of 2nd line therapy (endoscopy and radiology). These objectives should preferably be met using a noninvasive and limited cost technique that uses widely accessible urine and fecal samples.[35]

A molecular marker, also recognized as a biomarker, is a substance identified in serum or tissue and used to diagnose peculiar ailments. Biomarkers have an elevated predictive and prognostic significance and are a valuable method of primary CRC diagnosis, therapeutic interventions, and patient outcome (Table 4). Biomarkers can be categories as predictive, diagnostic, and prognostic with the ability of early diagnosis and disease detection. Predictive biomarkers analyze patients' compliance and response to therapy, which helps decide the future treatment schedule for positive results.[36] They can even be used to calculate the appropriate drug dosage and avoid cytotoxic activity. Prognostic biomarkers enable predicting the disease's natural course and categorize tumors into 2 categories: positive or negative. These are the moieties responsible for various processes, including tumor formation, demarcation, angiogenic, invasion, and metastasis. KRAS, BRAF, and MSI mutations are prominent during the diagnosis and treatment of CRC to help determine the best handling.[37] The vast majority of CRC cancers are sporadic (70–80%), with an estimated 20% genetic background. Colorectal cancer is a diverse ailment usually caused by a mixture of genetic and epigenetic changes. The most widely mutant allele in CRC patients is APC (about 80–82 percent of cases), TP53 (48–59 percent), KRAS (40–50 percent), and PIK3CA (14–18 percent). Assisting in cancer progression, surveillance, and treatment, new regulations for testing molecular biomarkers in CRC tumour cells have recently been developed.[38] Only a few biomarkers are involved with DNA mismatch repair system impairments or are dependent on the mutational status of genes associated with CRC carcinogenesis (NRAS, KRAS, BRAF).

NANOMEDICINE APPROACHES FOR THE MANAGEMENT OF CRC

Various therapies are currently employed to handle colorectal cancer, including chemotherapy, surgery, radiotherapy, and targeted drug therapy. A substantial preponderance involving preclinical studies and formulation research has been carried out to design the targeted approach for drug

delivery to the colon via alternate means/routes.[39] The four fundamental strategies to colon appropriate cancer therapy via oral route are drug delivery with temporal control, drug delivery with pH control, enzyme-based drug delivery systems, and pressure-based systems.[40] Alternative methods being suggested and employed for the colon-specific drug delivery includes microbial triggered system, osmotically controlled drug delivery, prodrug approach.[41] Each formulation method has advantages and disadvantages. The limitation can be overcome by different nanocarriers system and maximize colon specialized therapeutic agents, incarnation involving two or more formulations was engaged. As a result, for the nano medication of colorectal cancer, synthetic/herbal agents have been reported to be beneficial (Figure 4).

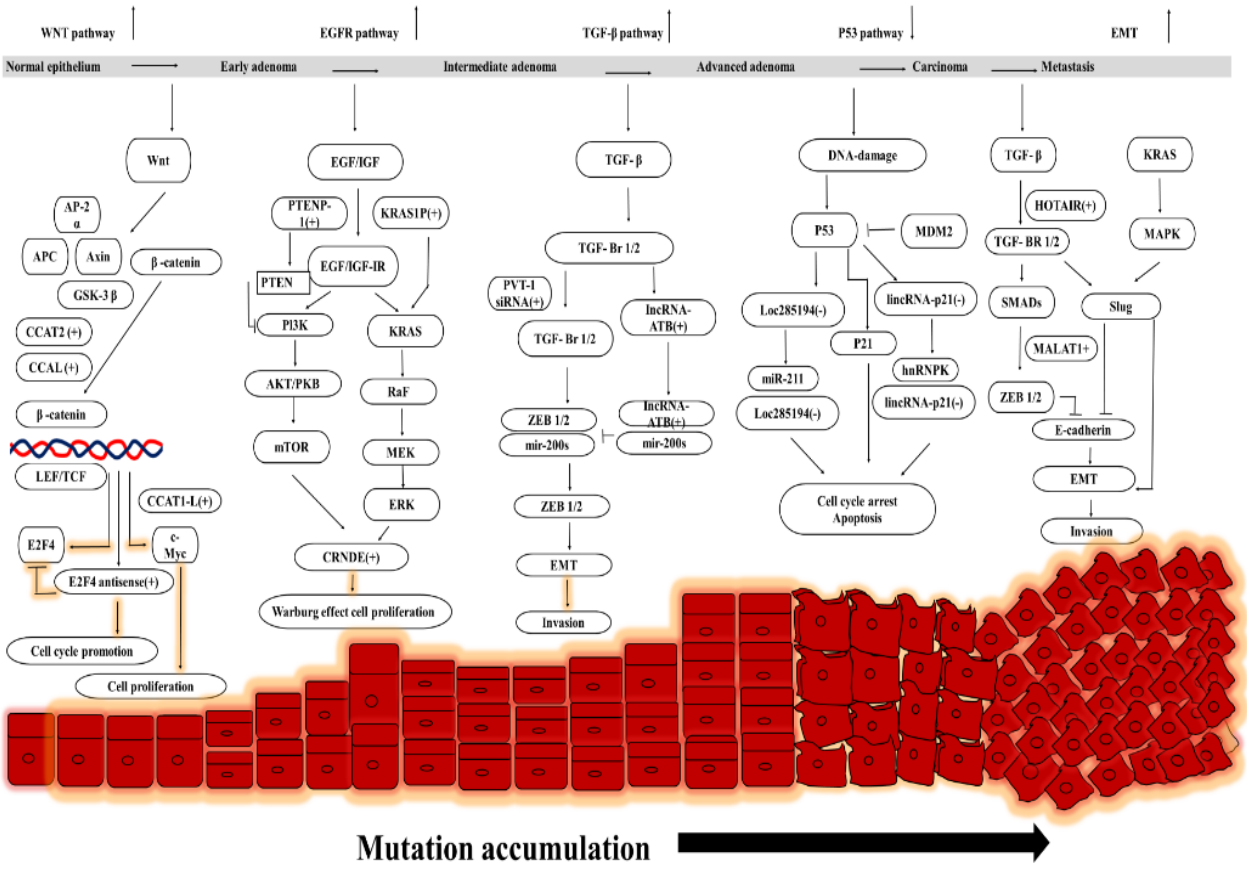

Fig 3: Molecular pathways of CRC

Table 2: Different screening techniques for CRC and their merits and demerits

Method	CT colonography	Colonoscopy	Stool tests (FIT or FOBT)	Sigmoidoscopy
Description	The entire colon images were captured by CT scanner. The obtained images are in 2D and 3D format, and helps radiologist to identified the tumor	The polyps can be detected in the colon/rectum by this method	These are blood or DNA testing kits that can detect abnormalities. The FOBT detects blood using guaiac. It utilises antibodies to detect blood in the faeces instead of using enzymes	Using this device, a physician may examine the lining of the colon and rectum
Merits	No sedation is required in this test and it is noninvasive	It may able to detect vary minute polyps, also it has the capability to detect large polyps and reduces the risk of death from CRC	The colon does not need to be cleansed. In some cases, it is possible to collect samples from home. When compared to other bowel cancer screening procedures, the cost of this test is very inexpensive	If polyps or malignancies are found, they can be detected with great accuracy
Demerits	Usually bowel preparation is required to clear up the colon. Besides polyps or cancer, it may also identify various abnormalities in the colon or rectal area	Required sedation, and it can cause severe bleeding/tear in the intestinal wall	Non-bleeding tumours cannot be detected by the device	A polyp or malignancy placed on the right side (cecum, hepatic flexure or part of the transverse colon) cannot be detected

Table 3: Detection and treatment of CRC using nanotechnological techniques

Nano-system	Structure	Characteristics	Application in CRC
Iron oxide nanocrystals	It consists of size ranges in between 1-100nm	Due to their superparamagnetic characteristics and their potential uses in various sectors, these materials have received considerable study	Detection
Liposomes	Structures comprised of lipid biolayers that self-assemble are called colloidal biolayers	It's an artificially produced vesicle composed of two bilayers of phospholipid, and it's been around for a while. These small, water-soluble particles can be transported into or out of a cell by liposomes	Detection and treatment
Dendrimers	As the name suggests, they are radially developing hyperbranched synthetic polymers containing repeating units	Branched, repeating molecules with a flawless structure	Detection and treatment
Quantum Dots	It's a semiconducting substance having a diameter of 2–10 nanometers (nm)	In addition, quantum dots have outstanding optical characteristics, including great brightness and adjustable wavelenghts	Detection and treatment
PLGA nanoparticles	Multiple drug delivery methods have been developed using different PLGA copolymers with varying characteristics	Hydrolysis of PLGA contributes to metabolite monomers such as lactic acid and glycolic acid	Detection and treatment

Table 4: Outline of Diagnostic, Prognostic and Predictive biomarkers

Biomarker	Types Tissue biomarker	Explanation
Diagnostic biomarker	Cytokeratin's	Approximately 65-95% of CK7–/CK20+ sequence is identified in CRC
	Telomerase	Telomerase itself is a ribonucleoprotein which mostly adds TTAGGG repeats to telomeres in order to maintain their integrity. Cancerous cells avoid DNA damage-induced inhibiting transcription factors by modulating telomerase. Telomerase shows 95% sensitivity and specificity in CRC when coupled with TBT.
	CDH17 (Cadherin 17)	CDH17 are often cell–cell interaction molecules that, under optimal conditions, cause changes in tissue structure. As per the reports, CDH17 is detected in 96-100% of primary CRC and documented about 100% of malignant CRC.
	CDX2 (Caudal type homeobox 2)	CDX2 expression has a specificity and sensitivity of over 90% in CRC diagnosis. Because CDX2 expression cannot distinguish between GI adenocarcinomas on its own,13,14 it can be used in conjunction with CKs.
	Biomarker in the blood	**Explanation**
	CfDNA (Circulating cell free DNA)	Even in cancer cells, cfDNA is dispersed with many more fragments than in healthy cells. During CRC diagnosis, quantitative analysis of circulating cfDNAs divulged sensitivity of 73 percent –90 percent and specificity of 97 percent –85 percent by comparing the longer to shorter DNA fragments or assessing cfDNA integrity number.
	Long Non coding RNAs (IncRNA)	Considering an AUC of 0.960, HIF1A-AS1 demonstrated exceptional diagnostic capabilities for CRC. Patients with CRC who had high HIF1A-AS1 expression had a lower 5-year mortality incidence than those who had low HIF1A-AS1 expression. CRNDE-h exceeded CEA in terms of diagnostic value. The diagnostic value increased when merged with CEA. Further, GAS5, ZFAS1, and NEAT1 are examples of markers that have showed better diagnostic or prognostic markers.
	Biomarker in the faeces	**Explanation**
	sDNA (Stool DNA)	The Cologuard test was much more responsive than the FIT and gFOBT, but it does have a greater proportion of false positives.
	gFOBT (Guaiac Fecal Occult Blood Test)	Using the gFOBT as a CRC clinical diagnosis lowered death rates by 11%–33% over 20 years. Nevertheless, it cannot distinguish between upper and lower GI bleeding between human and non-human heme.

Prognostic biomarker	Tissue biomarker	Explanation
	BRAF	Cell proliferation, migration, angiogenesis is all correlated with the mitogen-activated protein kinase pathway. A BRAF mutation is associated with a shorter PFS and OS. It proposed that it be studied for its prognostic role.
	CIMP	CIMP's prognostic significance is unknown. Despite this, a number of studies have found that CIMP+/CIMP-high CRC patients have an even worse prognosis than CIMP–/CIMP–high CRC patients.
	MSI	MSI-high tumors have a better prognosis than MSI-low tumors. Patients with MSI CRC had a longer OS and DFS than patients with MSS CRC.
	APC	The APC has been linked to FAP and a large proportion of sporadic CRC cases. Many transgenes are uncontrolled transcribed as a consequence of APC mutations. Patients with APC mutations and elevated amounts of miR-21 shown to have a shorter lifespan.
	Blood biomarker	**Explanation**
	CEA	CEA concentrations, according to reports, are significantly and positively related to patient outcomes. In patients with CRC metastasis to the liver, preoperative CEA levels were significantly related to prognosis.
	cfDNA	Among CRC patients, increased cfDNA levels were linked with an increased risk of recurrence and a very shorter overall survival. Serum KRAS, APC, and p53 mutations also were linked to a greater rate of surgical metastasis and relapse.
	NLR	Even after treatment, patients with elevated NLR had substantially lower cumulative mortality and progression-free survival (PFS). CRC patients who had an NLR of 5 before therapy had a better 5-year OS and DFS. Furthermore, in individuals with hepatic metastases, a high pretreatment NLR was linked to a shorter OS and RFS.
Predictive biomarker	**Tissue biomarker**	**Explanation**
	NRAS and KRAS	Numerous molecular pathways, including the EGFR pathway, are regulated by the GTPase protein (KRAS) encoded by the KRAS proto-oncogene. Even in tumour cells with wild-type RAS, anti-EGFR antibody treatment was clinically efficacious. KRAS mutations can predict an inadequate response to EGFR inhibitors.
	BRAF	According to reports, there is a lack of evidence that BRAF mutations could be used to anticipate the benefit of anti-EGFR antibody therapy.
	Biomarker in the blood	**Explanation**
	cfDNA	Upon primary resection, the amount of cfDNA reduced, but when the CRC reappeared, the level of cfDNA surged. The cfDNA concentration in participants with rectal cancer who received chemoradiotherapy results in greater non-compliance than compliance.

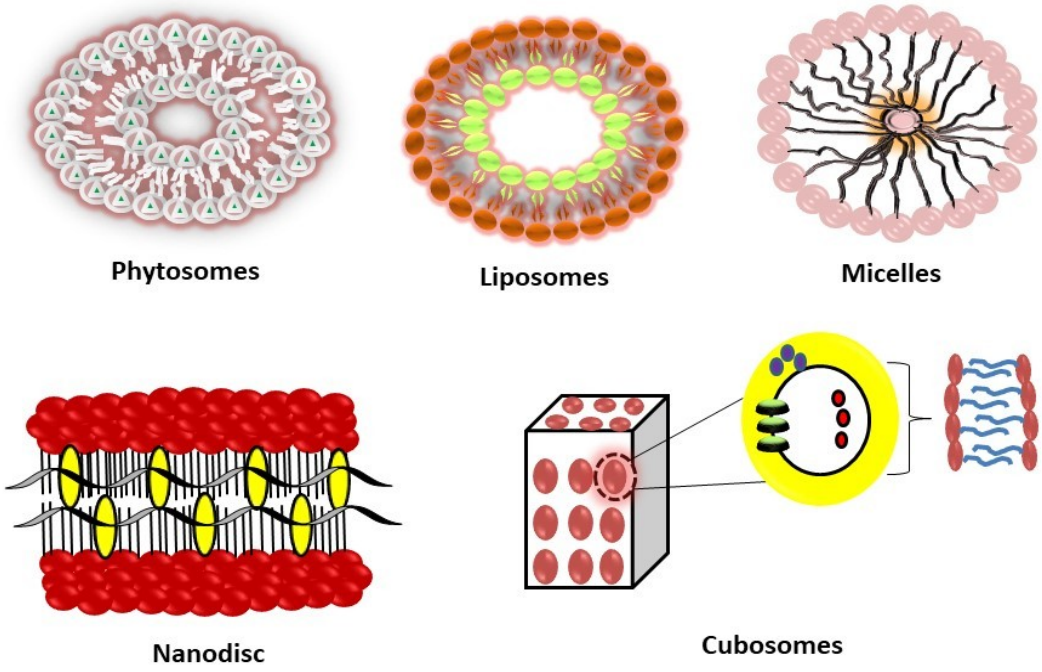

Figure 4: Nanocarrier based structure for therapeutic delivery

Phytosomes

Polyphenol molecules are linked to phosphatidylcholine molecules via intermolecular bonding in phytosomes complexes (PC). When a polyphenol is consumed orally, the amphipathic PC molecules likely "usher" the polyphenol via the intestinal epithelial cell outer membrane, allowing it to reach the bloodstream.[42] They are phospholipids with polar and non-polar parts in their structures and are utilised to form phytosomes. As a result of the phospholipids alcohols, glycerophospholipids and sphingomyelins can be separated. Vegetable oils (such as soybean, cotton seed, corn, sunflower and rapeseed) and animal tissues are the major sources of these compounds (such as egg yolk and bovine brain).[43] Marjaneh *et al.*, 2018 used a phytosomal preparation to investigate the impact of curcumin and 5–fluorouracil in colitis allied colon carcinoma using mouse model. Curcumin phytosomal preparation showed the enhanced anti-proliferative effect of 5-Flurouracil in vitro and in vivo compared to previous studies. The two- and three-dimensional cell culture model and colitis allied colon carcinoma mouse model was being used to investigate the cytoprotective effect of phytosomal curcumin formulation. It was discovered that the combined effect of curcumin and 5-FU retards the progression and invasiveness of colorectal cancer.[44]

Liposomes

Liposomes sphere-shaped vesicles made up of one or more phospholipid layers, and they've

been around for a while. They can be made from phospholipids, cholesterols, non-toxic surfactants, sphingolipids, glycolipids, long-chain fatty acids or membrane proteins. Liposomal drug delivery is distinguished by slow and delayed-release passive targeting via the EPR effect and high drug loading, which leads to dose reduction.[45] As a medication carrier for cancer treatment, liposomes have attracted a considerable lot of attention in biomedical research in recent decades. The efficacy of anticancer medicines can be improved by using liposomal carriers. This can result in better therapeutic benefits, reduced toxicity, greater drug stability, and improved drug dispersion.[46] Messerer *et al.*, 2004 developed an irinotecan liposomal formulation to treat generalized hepatic CRC in mice. Patients with LS180 tumors or colorectal metastasis (orthotopc LS174T) were given the prepared formulation intravenously. The proposed study discovered that the LS180 solid tumor model. It took 34 days to progress to a 400-mg tumor after injecting the stat liposomal irinotecan injection at 50mg/kg compared to the same dose inculcated for 22 days in the sample model with free drug. The liposomal irinotecan in the dose of 50mg/kg in total of 3 doses given for 4 days showed the survival (median) time of 79 days. Mice treated with different dose of NS (normal saline) lived for 34 to 53 days.[47]

Micelles

These micelles are composed of amphiphilic copolymers and are nanoscale in size (a hydrophilic shell and a hydrophobic core). This makes them a good choice for medication delivery. Since they lengthen the circulation duration of the therapeutic drug in blood, their use as a carrier system enhances therapeutic action.[48] As a result of many research studies, PM is a viable delivery method for CRC treatment. Prednisolone, retinoic acid, or DOX encapsulated in polymeric micelles (PM) can overcome the multidrug resistance of colon cancer cells more effectively than a medication when delivered alone.[49]

Lu *et al.*, 2019 designed the RNA based gene therapy to provide the optional method for cancer treatment. The micelles formulation was developed using the PEG-PCL copolymer. The formulation was introduced in the mouse having colon cancer as a sample model. DMP/siMcl1 and DMP/siBcl-xl complexes were used as the sample formulation showing the good potential as antitumor agent in vivo.[50] Yang et al., 2015 have developed curcumin-loaded micelles using MPEG-PCL copolymer incorporating trimethylene carbonate (TMC) to form MPEG-PCL-TMC micelles. TMC was incorporated in the formulation process to maintain polymer stability. In vitro study was performed using CT-26 cell lines and the outcomes of the proposed study found that curcumin micelles shows higher cellular uptake in CT-26 cell lines. Treatment with curcumin micelles shows lower tumor weight than free curcumin, resulting in vivo antitumor effects. Raveendran et al., 2013 developed curcumin micelles using copolymer Pluronic/Polycaprolactone. The outcome of the study revealed that compared to free curcumin, micelles improved absorption and bioavailability in Caco-2 cells.[51]

Nanodisc

An amphipathic protein-coated phospholipid bilayer, also known as a membrane scaffolding protein (MSP), forms the hydrophobic edge of nanodiscs, a synthetic membrane system. Apolipoprotein A1 (apoA1), the major component of high-density lipoproteins, is the MSP in some nanodiscs (HDL). Disk-like HDL replicates a more natural habitat than liposomes and micelles.[52]

Park *et al.*, 2017 developed a high-density mimicking nanodisc encapsulating doxorubicin. The nanodisc improved immune blockade in the murine colorectal cancer model. Immunogenic cell death in colorectal

cancer cells is induced by doxorubicin HDL nanodics, ensuing in exceptional anti-cancerous effect with no obvious off-target effects. CD8+ T cells anti carcinogenic responses are elicited with the administration of nanodics. This resulted in the reorganization of antigens, neo-antigens and intact cancer cells with augmentation of T cells.

Moreover, the combined effect of anti-programmed death-I and nanodisc established the relation between CT26 and MC38 CRC tumor in greater than 80% of the sample models. The re-occurrence of the cancerous effects in surviving animal models was not observed. This study found that nanodisc-based chemotherapy can stimulate antitumor immunity and make tumors more susceptible to immune checkpoint blockade.[53]

Cubosomes

Cubosomes are crystalline, isotropic nanoparticles stabilized by Poloxamers such as F127 and F108 (F127 and F108, respectively). An indeterminate periodic minimum surface of cubic symmetry is used to create a network of two distinct water channels produced by a three-dimensional, non-intersecting lipid bilayer. Cubosomes are characterized by many unusual properties, including their cubic shape, which allows for incorporating extremely lipophilic, hydrophilic, and amphiphilic medications.[54] Phytantriol and monoolein are biodegradable and biocompatible lipid excipients utilized in the production of cubosomes. These cubic nanoparticles are therefore considered safe carriers for drug delivery. As a result, the dissociation rate of cube-shaped lipid nanoparticles is slowed and drugs are retained better.[55]

Saber *et al.*, 2018 formulated the nano-cubosomes containing Cisplatin alone and Cisplatin metformin combination utilizing emulsification method. An enhanced cytotoxic effect was observed with Cisplatin encapsulated nano-cubosomes compared to untreated Cisplatin (HCT-116).

Moreover, the depletion of glucose level, decreasing the energy balance and disruptions of multiple metabolic pathways like mTOR inhibition and AMPK activation are responsible for inducing colorectal cancer cell apoptosis with Cisplatin metformin combination. The combined nano cubosomal formulation also concealed p-Akt (Ser473) and increased reactive oxygen species concentrations, resulting in an increase in nicotinamide-adenine-dinucleotide-phosphate (NADPH) oxidase, a decrease in LDH, and a significant increase in caspase-3 activity. Utilization of the combined formulation alters the molecular mechanism and thereby indicating the future implementation of nano-cubosomes for the treatment of CRC.[56]

FUTURE PROSPECTS AND RECOMMENDATIONS

CRC has become a major public health concern because of its excessive incidence and death rate. Medical professionals will get a new perspective on CRC by reviewing the latest discoveries in the study of CRC research and the newest results in diagnostic and therapy techniques. Researchers are studying a mix of genetic and environmental variables that contribute to the development of CRC. Their study is vital for the development of novel preventative measures. These genes have been discovered over the previous decade, and their pathogenic mutations have been characterized by high susceptibility to colon cancer. In the development of CRC, there are several molecular mechanisms involved. Nanotechnology plays a crucial importance in the therapy of CRC. Nanoscience is proven to be a key and hopeful strategy for the eventual eradication or at least chronic management of cancer. Nanotechnology had a revolutionary influence on cancer detection and treatment. Research

in biomedicine has prioritized developing drug delivery methods that may alter biodistribution, tissue absorption, and pharmacokinetics. Nanomedicine has the potential to play a major role in the diagnosis and treatment of human CRC and the enhancement of normal human physiology, despite the obstacles that limit its implementation.

CONCLUSION

An acute set of cellular mechanisms characterizes colorectal tumor, yet if sporadic or hereditary. The genes and pathways formed in the initial stages of oncogenesis appear to play essential roles in regulating normal crypt homeostasis. The APC gene, in specific, is significant in the onset of colorectal cancer, but it also plays a role in the normal gut's tissue homeostasis. This brief description includes details regarding the specific biomarker which can help in improving the prior detection and diagnosis of colorectal cases, thereby improving CRC patients' prognoses. This section highlights the most recent CRC system of biomarkers, prognostic agents, and diagnostic techniques prevailing in the present scenario. Research also focused on the detailed descriptions of various carrier systems that have been used in the management of CRCs. This review may help the researchers to develop effective medication/diagnostic tests for the management of CRC.

CONFLICT OF INTEREST

None

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