Potential genetic biomarker of Saudi Arabian patients with colorectal cancer

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ABSTRACT. – Colorectal cancer (CRC) is one of the leading causes of cancer deaths globally. We implemented a comprehensive literature review regarding CRC genetics studies to offer a perception into the genes associated with CRC recognized in Saudi patients. Definite genetic variants in ABCB1, ADIPOQ, CTNNB1, SFRP3, LRP6, CYPI9A1, PARP-1, TDG genes exhibited significant protection against CRC development in Saudi population. Whereas, other gene mutations in ABCB1, ABCC1, CASR, IL-17F, NOTCH1, NOTCH4, PRNCR1, TDG, TLR2, TLR4, TLR-9, TSLP, TSLP and TNF-α genes showed irrelevant correlation with CRC risk in Saudi Arabia. On the other hand, specific mutations in ABCC1, ADIPOQ, CYPIA1, KIR, IL-17A, MMP2, NOTCH3, PRNCR1, RETN, TDG, TLR2, BRAF, PARP-1, TLR4, TLR-9, TNF-α, TSLP and XRCC1 genes demonstrated a substantial augmented CRC risk development in Saudi patients. Furthermore, ATR, ATM, BMI1, CCAT1, Chk1, Chk2, COX-2, FoxM1, FSCN1, Ki67, MALAT1, mir-29, mir-34a, mir-92, mir-182-5, PANDAR, PIK3CA, TIGAR over-expression revealed a robust association with CRC in Saudi Arabia (KSA). Moreover, gene alterations in APC, EGFR, FBXW7, TP53, PTEN, K-ras genes were concomitant in CRC. As well as, lower expression of MLH1, MSH2, MSH6, PMS2, EPCAM and MUTYH genes were recognized in LS patients and future CRC Saudi patients. These gene mutations may be used as diagnostic and/or prognostic genetic markers in CRC Saudi patients and could offer a potential therapeutic target for CRC management.

Key Words:

Colorectal cancer, Gene, Saudi Arabia, Risk, Over-expression, Mutation, SNP.

Introduction

Cancer is the second utmost cause of death all around the world. Colorectal cancer (CRC), which includes colon and rectal cancer, is the third most common cancer Worldwide after lung and breast cancers1. CRC arises due to a series of epigenetic and genetic variations with subsequent high mortality rate. The etiological and risk influences of CRC development are inconsistently emulating the multifactorial nature of the disease. Numerous risk factors are linked to the CRC development risk, for instance age (older than 50 years), inflammatory bowel diseases2, history of adenomatous polyps, high-red meat diet, smoking and limited physical activity lifestyle3. At the molecular level, CRC is very intricate and necessitates establishing comprehensive patient stratification models through the identification of patients who will benefit or will not benefit from specific targeted therapy. Thus, understanding the personalized genetic map of each patient will help in developing the utmost appropriate management of his/her case.

Incidence Rate of CRC in Saudi Arabia

Colorectal cancer is the second most common cancer after breast cancer in Kingdom of Saudi Arabia (KSA)4. In KSA, CRC incidence is about half of the incidence in the US, nevertheless the greatest incidence is at a lower age group (40-60 years in the KSA vs. 60-80 years in the US)5. Saudi Cancer Registry stated that CRC is the second most common malignancy among Saudis for all ages (10.3%), the first cancer in Saudi males (11.8%)6, and the third one among Saudi females4. Also, between 1994 and 2003 age-standardized rates for CRC in KSA nearly doubled6. Between 2001 and 2003, whereas the annual percent change (APC) of CRC incidence among Saudi females displayed an insignificant rise, Saudi males showed extremely rising incidence, with an APC reaching 20.5%. Furthermore, it is anticipated that
by the year 2030, the CRC incidence in KSA may upsurge four-fold. Almatroudi (2020) preformed a retrospective observational population-based epidemiological study of CRC established on the Saudi Cancer Registry information, which included all CRC patients January 2006 to December 2016. The results verified that the highest mean age-standardized incidence rates (ASIRs) of the CRC were in Riyadh, Makka, and Eastern Province regions, whereas the lowest mean ASIRs were stated Jazan and Najran regions.

Risk Factors
In KSA, there is no nationwide policy for CRC screening despite the escalation incidence. Notably, most of CRC cases were identified throughout clinical assessments rather than through screening programs. Aljumah and Aljebreen (2017) recommended that the developing and applying CRC screening policy would be cost-effective application, which would eventually decline the financial burden on government spending, as well as improve the populations’ health status. Saudi Centre for Evidence-Based Healthcare panel meeting in 2015 agreed on the existence of a lack of nationwide incidence data concerning adenomatous polyps or the age groups in which the CRC incidence surges. Furthermore, there were no national clinical trials assessing the effectiveness of the diverse modalities of screening for CRC and their influence on mortality. It is crucial to recognize the risk factors connected to CRC development. Al-Zalabani (2020) recognized some of the population attributable fractions (PAFs) of CRC Saudi cases. The study results displayed that the largest fraction of attributable CRC cases among men and women was triggered by physical inactivity (16.13% and 16.45%), followed by extra weight (obesity: 9.71% and 6.93%; overweight 6.05% and 1.9%); and smoking (present smoker: 3.04% and 0.18%; prior smoker: 3.29% and 0.12%). Moreover, harmful actions such as consanguinity among the Saudi population, which were and still are detrimental factors for CRC increasing risk among KSA population. Subsequently, the risks for transporting genetic variations to the next generations in KSA are higher than in Europe and America, where there are less consanguineous marriages, and the family size is generally smaller. Although many advances have been made to diagnose and treat CRC in KSA, the latest cancer incidence report from the National Cancer Registry (NCR) indicated a worrying escalation in CRC patients. Accordingly, additional studies are still indispensable to conclude the diverse genotype biomarkers that can be used to predict the Saudi patients who are at risk of developing CRC.

The aim of the existing review is to offer an understanding into the genes recognized in CRC Saudi patients. These gene mutations may be used as diagnostic and/or prognostic genetic markers in CRC Saudi patients and could offer a potential therapeutic target for CRC management.

To achieve the current review goal, we accomplished a comprehensive state-of-art literature review regarding the genetics of CRC in KSA. A literature search was executed using online available databases like PubMed, ClinVar, Online Mendelian Inheritance in Man (OMIM), Phenotype-Genotype Integrator (PheGenI), DisGeNET and GWAS Catalog to retrieve all genetic studies executed on Saudi CRC patients or samples until December 2021. The search was done using the following strings: (colorectal), (Saudi), (gene), (colon), (rectal) and (cancer). All published articles including Saudi CRC patients were included in this review.

Genes Associated with Colorectal Cancer
Several genes’ mutations were included in this review, some of which were associated with increased risk of colorectal cancer, whereas others genetic mutations, were not associated with the development of CRC. Additionally, some genetic mutations showed a protective role against CRC development in Saudi patients. More than 40 genes mutations were included and for each of these we tried to explain a little about the gene associated proteins and their roles in CRC development as shown in the Supplementary Table I.

ABC
Adenosine Triphosphate (ATP) binding cassette (ABC) transporters play an indispensable part in the development of numerous disorders, including cancers, through drug resistance mechanism.

ABCB1
ABCB1 gene is positioned on chromosome 7 and encodes a P-glycoprotein (Pgp) which is responsible for the active efflux of drugs from cells. Al Qahtani et al (2019) studied the genotype distributions and the allele frequencies of two major variants in ABCB1 gene, C3435T and T129C, and linked them with CRC in Saudi Arabia. They recognized no significant association between ABCB1, 3435C>T and 129T>C polymorphisms
with CRC risk. Furthermore, patients with 3435 homozygous (TT) genotypes had lower risk of developing CRC risks. The same group of research implemented another study on 62 CRC patients to determine the genotypic distribution and allele frequency of another two ABCB1 Single nucleotide polymorphisms (SNPs), T1236C and G2677T, in Saudi CRC patients. The outcomes displayed no noteworthy variations in T1236C in CRC patients than controls. However, G2677T showed a protecting action against CRC progression.

**ABCC1**

Abdulkhaleq et al (2019) conducted a case-control study on 51 colon cancer patients to recognize the effect of two SNPs; G128C and C218T in the **ABCC1** gene on CRC development. The results showed an association between heterozygous (CT) genotype for variant C218T and an increased risk of colon cancer (3 times over) and high-grade stages (III and IV). They concluded that the CT genotype of variant C218T in **ABCC1** gene might intensify the risk of developing colon cancer among Saudi population, suggesting that this variant can be used as a prognostic marker for colon cancer. In contrast, the variant G128C exhibited no association with colon cancer.

**ADIPOQ**

Adiponectin is an adipose-specific protein, which has anti-atherogenic, anti-inflammatory and anti-diabetic actions. In order to appraise the influence of the adiponectin gene **ADIPOQ** polymorphisms to the risk of colon cancer, Al-Harithy and Al-Zahrani (2012) conducted a case-control study on 60 colon cancer patients. They examined the link between two SNPs, rs1501299 (G276T) and rs2241766 (T45G), in the **ADIPOQ** gene and CRC risk. The results showed that carriers of the heterozygous (GT) genotype of G276T displayed a higher risk of colon cancer than carriers of (GG) genotype. By contrast, the G allele in position 45 of the **ADIPOQ** gene had a lower risk of colon cancer than carriers of the normal (TT) genotype. The results suggested that the G276T SNP contributes to the genetic risk of colon cancer, while the presence of the G allele in position 45 of the **ADIPOQ** gene acted as a defensive factor against colon cancer.

**ALK**

Anaplastic lymphoma kinase (ALK) is a tyrosine kinase that was acknowledged as part of chromosomal rearrangement as a fusion partner of nucleophosmin. Bavi et al (2013) identified the frequency and nature of **ALK** alterations via recruiting 770 Saudi CRC patients. CRC prognosis was poor in patients with **ALK** gene amplification and gain in copy number as compared to CRC patients with standard **ALK** gene copy number. **ALK** gene amplification and gain in copy number were considered as an independent prognostic marker for poor survival in CRC across all stages.

**APC**

The adenomatous polyposis coli (APC) gene mutation is one of the primary events in CRC. Ninety-five tumor samples were retrospectively recruited in a study accomplished by Almuzzaini et al (2021), and 96% of the samples exhibited at least one confirmed **APC** gene pathogenic variant. Five novel variants, at the time, out of total 38 variants were detected in the **APC** gene. These variants included c.1696G>A (p.V566I) missense mutation at exon 14, c.1697delT (p.V566X) frame shift mutation at exon 14, c.2680_2681del-GTinsTA (p.Val894Ter) stop gain mutation at exon 16, c.3917delA (p.E1306X), frame shift mutation at exon 16, and c.4320-4341del ACCCTCCTCA (p. PPPPQ-TAQ1440-1447X). Additional study investigated 99 CRC cases via targeted sequencing, which led to the identification of frequent mutations in **APC**. **APC** gene was the second most commonly mutated gene in that cohort, with 36.4% of the cases examined demonstrating missense, nonsense or frameshift mutations in the hotspot regions of this gene. The most common mutation identified was the p. Arg1450Ter change, resulting in the expression of truncated **APC** and thus loss of control on nuclear β-catenin mediated gene expression and poorly regulated WNT pathway.

**ATR and ATM**

The expression of four telomere-associated proteins, hTERT, TRF1, TRF2, POT1 were studied by Aljarbou et al (2018). There are six individual proteins associated with telomeric DNA, collectively called shelterin complex. They are essential in preventing the recognition of the telomere as single or double strand breaks via the inhibition of Ataxia telangiectasia mutated (ATM) and Ataxia telangiectasia and Rad3 related (ATR) dependent by DNA damage response (DDR) pathway. The expression of ATR, ATM and Chk1, Chk2 were significantly escalated in cancer tissues. Thus, the expression of ATR/Chk1 and ATM/Chk2, may serve as a therapeutic re-
Table I. Summary of the genes mentioned in this review and mutations occurred in them, as well as their association with CRC development risk.

<table>
<thead>
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<th>Gene of interest</th>
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<tbody>
<tr>
<td>ABCB1</td>
<td>No significant association between ABCB1 3435C &gt; T and 129T &gt; C polymorphisms with CRC risk. Patients with 3435 homozygous (TT) genotype had lower risk of developing CRC risk.</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td>No noteworthy variations in T1236C in CRC patients. G2677T showed a protecting against CRC progression.</td>
<td>16</td>
</tr>
<tr>
<td>ABCC1</td>
<td>An association between heterozygous (CT) genotype for variant C218T and increased risk of colon cancer and high-grade stages (III and IV).</td>
<td>17</td>
</tr>
<tr>
<td>ADIPQ</td>
<td>Carriers of the heterozygous (GT) genotype of G276T displayed a higher risk of colon cancer than (GG) genotype. By cont rast, the G allele of T45G had a lower risk of colon cancer than the normal (TT) genotype.</td>
<td>18</td>
</tr>
<tr>
<td>ALK</td>
<td>CRC prognosis was poor in patients with ALK gene amplification and gain in copy number as compared with CRC patients with standard ALK gene copy number.</td>
<td>20</td>
</tr>
<tr>
<td>APC</td>
<td>96% of the samples exhibited at least one confirmed APC gene pathogenic variant.</td>
<td>21</td>
</tr>
<tr>
<td></td>
<td>APC gene was the second most commonly mutated gene. Most common mutation identified was the p. Arg1450Ter change, resulting in the expression of truncated APC.</td>
<td>22</td>
</tr>
<tr>
<td>ATR &amp; ATM</td>
<td>The expressions of ATR, ATM and Chk1, Chk2 were escalated in cancer tissues.</td>
<td>23</td>
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<tr>
<td>BCAR4</td>
<td>V600E mutation was detected in female patients, deletion of A at c.1758 bp, causing a frameshift and truncated protein, insertion of A/C at c.1860 bp and an insertion of A, leading to frameshift and stop codon. Mutation at c.1780G &gt; A (D594N), and c.1826insT, resulted in frame shift and truncated protein. BRAF frameshift mutations E586E, Q609L, and M620I lead to truncated defective BRAF proteins.</td>
<td>24</td>
</tr>
<tr>
<td>CASR</td>
<td>Intron 4 variant (rs3804594) in CASR gene was not correlated to CRC risk.</td>
<td>25</td>
</tr>
<tr>
<td>CCAT1&amp;C-CAT2</td>
<td>The expression level of CCAT1 was augmented in CRC patients. No noteworthy CCAT2 expression in CRC samples.</td>
<td>26</td>
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<tr>
<td>COX-2</td>
<td>COX-2 over expression in CRC cases and associated with shorter survival.</td>
<td>27</td>
</tr>
<tr>
<td>CTNNB1</td>
<td>Genetic variants in CTNNB1 (β-catenin) (rs4135385), SFRP3 (rs7775), and LRP6 (rs2284396) genes were associated with a defense against CRC development. GG genotype was associated with lower risk CRC developing relative to AA genotype at rs4135385 of CTNNB1 signifying an association of the rs4135385 in CTNNB1 gene with a reduced CRC risk.</td>
<td>28</td>
</tr>
<tr>
<td>CYP1A1</td>
<td>High distribution of CYP1A1wt/*24 genotype in CRC patients, reflecting a significant rise of cancer risk associated with CYP1A1wt/*24 genotype.</td>
<td>29</td>
</tr>
<tr>
<td>CYP2E1</td>
<td>CYP2E1 wt/*6 was not related to CRC risk in Saudi populations.</td>
<td>30</td>
</tr>
<tr>
<td>CYP19A1</td>
<td>Three SNPs rs4774585, an A &gt; G transition, rs936308, C &gt; G transversion, and rs4775936 C &gt; T transition. AA genotype of rs4774585, GG state of rs936308 and TT state of rs4775936 exhibited a negative association with CRC development in Saudi patients.</td>
<td>31</td>
</tr>
<tr>
<td>EGFR</td>
<td>Frequent mutations in EGFR were associated with young age of onset and poor disease-specific survival in CRC.</td>
<td>32</td>
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<tr>
<td>FBXW7</td>
<td>Frequent mutations in FBXW7 were identified</td>
<td>33</td>
</tr>
<tr>
<td>FOXM1</td>
<td>FoxM1 protein overexpression was elevated in CRC tissues and was related with poorly differentia-ted and highly proliferative tumors.</td>
<td>34</td>
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<tr>
<td>FSCN1</td>
<td>FSCN1 intensified in CRC patients and was accompanied by reduced OS and DFS. Furthermore, high BMI/FSCN1 patients experienced the worst OS and DFS.</td>
<td>35</td>
</tr>
<tr>
<td>GSTM1</td>
<td>Majority of CRC cases harbored the null genotype (GSTM1*0/*0).</td>
<td>36</td>
</tr>
<tr>
<td>GSTP1</td>
<td>None of the genotypes of GSTP1 studied were associated with an increased risk of CRC.</td>
<td>37</td>
</tr>
<tr>
<td>hBD</td>
<td>hBD-1 mutations and mutation 1 of hBD-3 lead to truncated pre-proteins. hBD-3 mutation 2 protein was greatly destabilized. These outcomes established a significant reduction of hBDs in cancer tissues.</td>
<td>38</td>
</tr>
<tr>
<td>hTERT</td>
<td>All CRC cases expressed hTERT; however, there was no difference between tumor and adjacent mucosa.</td>
<td>39</td>
</tr>
</tbody>
</table>

Table continued
Gene’s polymorphisms in colorectal cancer Saudi patients

Table I. (Continued). Summary of the genes mentioned in this review and mutations occurred in them, as well as their association with CRC development risk.

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<tbody>
<tr>
<td>IL-17</td>
<td>Males harboring the A allele of IL-17A G197A SNP exhibited higher risk of developing CRC. No connection between IL-17F (rs763780) polymorphism and CRC susceptibility.</td>
<td>46</td>
</tr>
<tr>
<td>KIR</td>
<td>Five activating KIR genes (2DS1, 2DS2, 2DS3, 2DS5, and 3DS1) were significantly more prevalent in the CRC patients. The highest risk was associated with the 3DS1 gene, followed by the 2DS1 gene. Additionally, no associations were found between 3DL1 and 2DS4 and CRC, whereas 2DS2 was inversely associated with CRC risk.</td>
<td>50</td>
</tr>
<tr>
<td>K-ras</td>
<td>No mutations were identified in codon 61 of exon 3. Exon 4 aberrations harbored p.Ala146Thr (a missense mutation), p.Ala134Val, p.Arg135Lys, p.Gln150Stop, p.Lys147Lys, p.Gln150Stop, p.Gly138Gly, p.Arg149Gly, p.Gly138Gly, and p.Gly138Gly mutations. Missense K-ras mutations altering the amino acid sequence of the protein (A134V, R135K, E143K and R149G) were distinguished. Moreover, two synonymous K-ras mutations (G138G and K147K) and a nonsense truncating mutation (Q150X) were detected. E143K mutation is predicted to have a damaging effect on the protein. R149G mutation is predicted to be neutral, but molecular modeling showed that this mutation caused changes in the helix and loop chains near the GTP binding pocket. Q150X mutation introduced a premature stop codon.</td>
<td>5</td>
</tr>
<tr>
<td>LRP6</td>
<td>Decreased risk of developing CRC in CC genotype compared to TT genotype at SNP rs2284396 in LRP6 gene.</td>
<td>34</td>
</tr>
<tr>
<td>MALAT1</td>
<td>The expression levels of MALAT1 were significantly high in CRC patients.</td>
<td>24</td>
</tr>
<tr>
<td>MED12</td>
<td>Three MED12 somatic mutations in CRC patients were found.</td>
<td>57</td>
</tr>
<tr>
<td>MEG3</td>
<td>No significant expression of MEG3 in CRC was detected.</td>
<td>24</td>
</tr>
<tr>
<td>miRNAs</td>
<td>Significant rises in miR-29 and miR-92 and their expression levels in CRC, while miR-145 and miR-195 decreased in CRC tissues.</td>
<td>95</td>
</tr>
<tr>
<td>MLH1</td>
<td>Structural loss in the genomic regions of MLH1 (3p23-p14.2), MSH2, MSH6, EPCAM (2p21-p16.3), PMS2 (7p22.1) and MUTYH (1P34.1-p33) in LS patients. This structural loss resulted in lower expression of MLH1, MSH2, MSH6, PMS2, EPCAM and MUTYH genes. Positive MLH1 correlation was found in around 27% and that MSI was present in more than 90% of CRC patients with Lynch syndrome.</td>
<td>12</td>
</tr>
<tr>
<td>MMP2</td>
<td>C1306 T mutation was significantly more common in colon Saudi patients.</td>
<td>65,66</td>
</tr>
<tr>
<td>NOTCH1</td>
<td>SNPs rs3124591 in NOTCH1 and rs3820041 in NOTCH4 did not exhibit any association with CRC. rs1043994 in NOTCH3 displayed a significant association with CRC in males.</td>
<td>68</td>
</tr>
<tr>
<td>PANDAR</td>
<td>The expression levels of PANDAR were significantly enlarged in CRC patients.</td>
<td>24</td>
</tr>
<tr>
<td>PARP-1</td>
<td>SNPs in PARP-1 gene, including Met129Thr, Val762Ala, and Lys940Arg, did not show any association with CRC risk in Saudi population. Lys933Asn and Lys945Asn showed significant association with CRC among Saudis. SNP rs8679 diminished susceptibility to colorectal cancer at heterozygous TC allele and at minor allele C.</td>
<td>69</td>
</tr>
<tr>
<td>PCAT6</td>
<td>No significant expression of PCAT6 was detected in CRC patients.</td>
<td>24</td>
</tr>
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Table continued
Table I. (Continued). Summary of the genes mentioned in this review and mutations occurred in them, as well as their association with CRC development risk.

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<tr>
<td><strong>POLD1 and POLE</strong></td>
<td>Numerous variants in POLE gene were associated with an increased risk for CRC. Low POLE protein expression exhibited a substantial link with lymph node involvement and grade III tumors. Whereas for POLD1, low expression was related with adenocarcinoma histology, larger tumor size and stage III tumors.</td>
<td>75</td>
</tr>
<tr>
<td><strong>PRNCR1</strong></td>
<td>One SNPs, rs1456315, in PRNCR1 gene showed an association with CRC with the homozygous CC variant allele, among younger age patients (≤ 57) and in female patients. Three other SNPs, rs1016343 (C &gt; T), rs13252298 (A &gt; G), and rs16901946 (A &gt; G), in PRNCR1 gene did not display any association with CRC.</td>
<td>74</td>
</tr>
<tr>
<td><strong>P35 and PTEN</strong></td>
<td>P35 positive expression was established in 25.4% CRC, whereas loss of PTEN expression was recognized in 32.3% CRC patients.</td>
<td>64</td>
</tr>
<tr>
<td><strong>RETN</strong></td>
<td>SNPs in RETN gene rs1862513 (C &gt; 202G) and rs3745367 (G &gt; 299A) had increased the risk of colon cancer. Carriers of the heterozygous (GA) genotype of SNP 299 had a significantly higher colon cancer risk than carriers of the wild (GG) genotype.</td>
<td>77</td>
</tr>
<tr>
<td><strong>SFRP3</strong></td>
<td>Genetic variant (rs7775) was correlated to considerable protection against CRC progression. Women having Gly at codon 324 (rs7775) of SFRP3 have 2.5-fold lower risk of developing CRC compared to those having Arg at this locus.</td>
<td>74</td>
</tr>
<tr>
<td><strong>SMAD4</strong></td>
<td>SMAD4 gene was identified and correlated to CRC development.</td>
<td>25</td>
</tr>
<tr>
<td><strong>TDG</strong></td>
<td>SNP rs1435113 showed a significant risk association of its genotype AA and of the minor allele A in CRC Saudi patients. SNP rs1866074 presented a protective association with the GG allele and the additive (AG+GG) allele in CRC patients. Other four SNPs (rs4135050, rs4135066, rs3751209, and rs1882018) showed no association with CRC patients in the Saudi population.</td>
<td>79</td>
</tr>
<tr>
<td><strong>TIGAR</strong></td>
<td>TIGAR expression was found in 68% of the tumor samples with nuclear localization and was significantly amplified in early stage (stage I and II) and late stage (stage III and IV) of CRC.</td>
<td>81</td>
</tr>
<tr>
<td><strong>TLR2</strong></td>
<td>TLR2 rs3804099 and TLR2 rs4696480 SNP were associated with CRC susceptibility, while TLR2 (rs3804100 C &gt; T) disclosed no association with CRC susceptibility in Saudi patients.</td>
<td>83</td>
</tr>
<tr>
<td><strong>TLR4</strong></td>
<td>A clear association between TLR4 rs10759931 polymorphism, the G allele, and susceptibility to CRC development risk was detected. TLR4 rs2770150 is associated with CRC in women aged over 50 years. Whereas SNP rs10759932 and rs4986790 appeared not to have any association with colon cancer.</td>
<td>84</td>
</tr>
<tr>
<td><strong>TLR6</strong></td>
<td>rs3796508 is a protective factor against CRC in the older male Saudi population. Two other non-synonymous SNP S249P and V327M were common in a few patients and were predicted as damaging.</td>
<td>85</td>
</tr>
<tr>
<td><strong>TLR9</strong></td>
<td>An association between the rs187084 SNP and CRC risk was found in female patients. T allele exhibited lower frequency in female cancer patients. Additionally, rs352139 and rs352144 SNPs were found to be correlated with colon cancer development. SNPs rs352144, rs187084 and rs5743839 were not associated with colorectal cancer in males.</td>
<td>87</td>
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<tr>
<td><strong>TNF-α</strong></td>
<td>SNPs -308 and -857 were not associated with CRC, while -238 (G/A) genotype was significantly concomitant with high risk of CRC. AA genotype of -238 G/A SNP was higher proportion in CRCs.</td>
<td>88</td>
</tr>
<tr>
<td><strong>PTEN</strong></td>
<td>Loss of PTEN has been reported in CRC.</td>
<td>89</td>
</tr>
<tr>
<td><strong>TP53</strong></td>
<td>KRAS or PIK3CA mutations were significantly associated with poor disease-specific survival in cases with wild-type TP53.</td>
<td>22</td>
</tr>
<tr>
<td><strong>TSLP</strong></td>
<td>SNP rs10043985 presented a strong correlation with CRC Saudi patients, whereas rs2289276 SNP did not show any relation with CRC.</td>
<td>93</td>
</tr>
<tr>
<td><strong>UCA1</strong></td>
<td>No significant expression of UCA1 detected in 63 CRC.</td>
<td>24</td>
</tr>
<tr>
<td><strong>VDR</strong></td>
<td>No association between the four VDR polymorphisms with CRC risk was found in the overall analysis. Apal and BsmI loci were associated with CRC in elderly and female patients.</td>
<td>95</td>
</tr>
<tr>
<td><strong>XPD and XRCC1</strong></td>
<td>No significant difference in XPD Lys751Gln polymorphism in CRC. An association between the GG genotype of XRCC1 polymorphism and the increased risk of CRC was detected. Also, XRCC1 (AG + GG) polymorphism may be associated with increased clinic pathological parameters of CRC.</td>
<td>99</td>
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</table>
response biomarker to certain therapies that induce these DNA damages\(^2\).

**BCAR4**

Siddique et al\(^2\) (2019) designed a study to measure the expression of several oncogenic lncRNAs, including BCAR4. The study revealed that there was no significant expression of BCAR4 in CRC blood samples. However, a small number of subjects limited the statistical power in some comparisons.

**BMI1**

BMI1 polycomb gene promotes cancer cell survival via p53-dependant cell death suppression\(^2\). BMI1 expression was related to cancer progression and poor clinical outcome. Alajez (2016)\(^2\) demonstrated a significant escalation of BMI1 in CRC Saudi patients. Furthermore, high BMI1 expression was concomitant with reduced overall survival (OS) and reduced disease-free survival (DFS).

**BRAF**

BRAF (B-Raf proto-oncogene, serine/threonine kinase) is a member of the rapidly accelerated fibrosarcoma (RAF) family. BRAF regulates the mitogen-activated protein kinase (MAPK)/extracellular signal-regulated kinase (ERK) signaling pathway, thus affecting cell division, differentiation, and secretion\(^2\). Rasool et al\(^2\) (2021) recognized numerous BRAF mutations in 14% of CRC Saudi patients studied. Of these, 5% of these mutations were in the V600E position, especially in female patients. The second most common mutation was the deletion of A at c.1758 bp, causing a frameshift and truncated protein of 589 amino acids only. Thirdly, heterozygous insertion of A/C at c.1860 bp and an insertion of A, leading to frameshift and stop codon after 629aa. Additionally, a mutation in at c.1780G > A (D594N), and c.1826insT, that resulted in frame shift and truncated protein of 610 amino acids. BRAF frame-shift mutations E586E, Q609L, and M620I were detrimental, leading to truncated defective BRAF proteins. Accordingly, these frameshift mutations predict the worst clinical prognosis, as well as impaired response to therapy.

**CASR**

CASR (calcium sensing receptor) expression exhibits a protective role in CRC patients via several signaling transductions, such as stimulating cell differentiation, prompting apoptosis and constraining proliferation\(^2\). However, Al-Ghafari (2019)\(^3\) verified that intron 4 variant (rs3804594) in CASR gene did not correlate with CRC risk.

**CCAT1 and CCAT2**

Siddique et al\(^2\) (2019) showed that the expression level of CCAT1 was significantly augmented in CRC patients, concluding that CCAT1 expression could be used as possible biomarkers for CRC prognosis. Alternatively, no noteworthy CCAT2 expression has been found in CRC samples when related to control\(^2\).

**COX-2**

Numerous inflammatory mediators and cytokines are elaborated in the cancer pathogenesis, one of them belonging to the family of cyclooxygenases\(^3\). Albashri et al\(^3\) (2018) conducted a retrospective study including 324 CRC diagnosed cases. They observed COX-2 over expression in 40% of normal colonic mucosa, 65% of colorectal adenoma and 84.6% of CRC cases. Furthermore, elevated patients showing an elevated COX-2 expression were found to suffer from shorter survival.

**CTNNB1 (β-catenin)**

The N-terminus of β-catenin harbors highly conserved residues, S33, S37, S45, and T41 encoded by exon 3 of the human CTNNB1 gene. Alteration in any of these amino acid residues in CTNNB1 gene exon 3 produces a stabilized β-catenin. Stabilized β-catenin could not be phosphorylated, causing constitutively active transactivation complexes, with subsequent loss of cell growth control\(^3\). Parine et al\(^3\) (2019) studied thirteen SNPs in 8 genes including CTNNB1 (β-catenin) (rs4135385, rs13072632), SFRP3 (rs7775), APC (rs454886, rs459552), LR6P (rs2075241, rs2284396), DKK4 (rs3763511), DKK3 (rs6485350), TCF4 (rs12255372) and AXIN2 (rs3923086, rs3923087, rs4791171) in CRC patients. From the 13 SNPs, only genetic variants in CTNNB1 (β-catenin) (rs4135385), SFRP3 (rs7775), and LR6P (rs2284396) genes were associated with considerable defense against CRC development. GG genotype was associated with a lower risk of CRC developing relative to AA genotype at rs4135385 of CTNNB1, translating into an association of the rs4135385 in CTNNB1 gene with a reduced CRC risk\(^3\).

**CYP**

**CYP1A1**

CYP1A1 is important for the conversion of carcinogenic polycyclic aromatic hydrocarbons\(^3\).
Saeed et al. (2013) demonstrated that a high distribution of CYP1A1 wt/*2A genotype in CRC patients, reflecting a significant rise of cancer risk associated with CYP1A1 wt/*2A genotype.

CYP2E1
CYP2E1 enzyme is responsible for the metabolic oxidation of low molecular weight carcinogens. A nucleotide replacement (7632T>A) in intron 6 resulted in the absence of a Dral restriction enzyme site (CYP2E1*6 allele, rs. 6413432). However, CYP2E1 wt/*6 was not related to CRC risk in Saudi population.

CYP19A1
Al-Mukaynizi et al. (2017) designed a case-control study to explore the effect of three SNPs in the CYP19A1, on CRC risk. Two of the SNPs, rs4774585, an A > G transition and rs936308, C > G transition, are positioned in the promoter region. The third SNP rs4775936 is situated in the intronic region and is a C > T transition. AA genotype of rs4774585, GG state of rs936308 and TT state of rs4775936 exhibited a negative association with CRC development in Saudi patients.

EGFR
A study has investigated 99 CRC cases, leading to the recognition of frequent mutations in EGFR (11%). EGFR mutations were relatively frequent and significantly associated with young age of onset and poor disease-specific survival in CRC.

FBXW7
Dallol et al. (2016) identified frequent mutations in FBXW7 in 99 CRC cases via targeted sequencing.

FOX
Forkhead box protein M1 (FoxM1) is a member of the FoxM family, which initiates cell cycle progression and evasion of growth arrest. Consequently, FoxM1 deregulation was found to be involved in cancer pathogenesis. FoxM1 protein overexpression was revealed in 66% of 448 CRC tissues and was related with poorly differentiated and highly proliferative tumors. Thus, FOXM1 gene may serve as a valuable molecular biomarker, as well as a possible therapeutic target.

FSCN1
FSCN1, an actin-binding protein, stimulates cancer cell relocation, invasion and metastasis.

GST
Glutathione S-transferases (GSTs) are family of phase II enzymes vital in carcinogen detoxification. One of these GSTs is the GSTM, a protective enzyme that detoxifies several carcinogens. GSTM*0/*0 genotype exhibits deficiency in enzyme activity with subsequent reduced carcinogen-detoxification ability. A study implemented using 83 CRC patients, showed that the majority (83%) of CRC cases harbored the null genotype (GSTM1*0/*0). The remaining (17%) cases had either the GSTM1 wt/wt or the GSTM1 wt/*0 genotype. Therefore, among the control cases, 65% had the null genotype (GSTM1*0/*0) and 35% had either the GSTM1 wt/wt or the GSTM1 wt/*0 genotype. Another study found GSTM1*0/*0 genotype in only 2% of CRC patients, suggesting that GSTM1*0/*0 is not a risk factor in CRC patients.

GSTP1
In the fifth exon at the codon 105 of Glutathione S-transferase pi (GSTP1), A changes to G which triggers substitution of isoleucine (Ile) with valine (Val) (Ile105Val), with a subsequent low enzyme activity. A study tried to examine the potential influence of GSTP1 (Ile105Val) polymorphism in CRC risk in Saudi patients. None of the genotypes of GSTP1 was associated with an increased risk of CRC development.

hBD
Human β-defensins (hBDs) belong to a family of antimicrobial peptides that constitute an important part of the innate immune defense. To date, four hBDs (1-4) have been identified in human tissues. Semlali et al. (2015) designed a study to analyze the expression and genetic variations in hBDs (hBD-1, hBD-2, hBD-3 and hBD-4) and their putative association with CRC in Saudi population. Numerous mutations, generally insertions, were recognized in different exons (1/2).
These mutations contributed to significant changes in the protein structure of hBDs (i.e., hBD-1, hBD-3 and hBD-4). hBD-1 mutations and mutation 1 of hBD-3 lead to truncated pre-proteins with no predicted mature hBD-1 protein synthesis. hBD-3 mutation 2 protein is greatly destabilized because of the absence of a disulfide bridge caused by the substitution of cysteine to leucine (Cys63Leu). In addition, in the same mutant, lysine to glutamic acid (Lys67→Glu) substitution introduced a negatively charged, acidic residue in a positively charged, hydrophobic C-terminal section. These outcomes established a significant reduction of hBDs in cancer tissues related to normal tissues.

hTERT
The expression of four telomere-associated proteins, hTERT, TRF1, TRF2, POT1 were studied by Aljarbou et al23 (2018). All CRC samples expressed hTERT; however, there was no difference between tumor and adjacent mucosa. Tissues adjacent to tumors showed detectable hTERT mRNA levels, while normal tissues do not express hTERT. Thus, Aljarbou et al23 (2018) findings can be attributed to the presence of cancer-associated genetic modifications. Additionally, a positive correlation between the age of the patients and hTERT expression was identified23.

IL-17
IL-17 is a major cytokine created by Th17 cells to prompt inflammatory cytokines and chemokines production by neutrophils and macrophages, thus playing a crucial role in human malignancies27. Al Obeed et al48 (2018) detected that males harboring the A allele of IL-17A G197A SNP exhibited higher risk of developing CRC. No connection between IL-17F (rs763780) polymorphism and CRC susceptibility48 has been found.

KIR
Natural killer (NK) cells play a fundamental role in the immunity regulation against infected and malignantly transformed cells through their killer cell immunoglobulin-like receptors (KIRs). KIRs interacts with human leukocyte antigen (HLA) molecules49. Al Omar et al50 (2015) verified five activating KIR genes (2DS1, 2DS2, 2DS3, 2DS5, and 3DS1), which were significantly more prevalent in CRC patients. The highest risk was associated with the 3DS1 gene, followed by the 2DS1 gene. Additionally, no association was found between 3DL1 and 2DS4 and CRC, whereas 2DS2 was inversely associated with CRC risk.

K-ras
In K-ras gene, the majority of somatic mutations occur at codons 12 and 13 (situated in exon 2). Other less frequent mutations occur in exon 3 (codons 59/61) and exon 4 (codons 117/146)51. Wild type K-ras protein resides in the GDP-bound state on the plasma membrane in inactive cells, whereas mutated K-ras is in a continuous stimulation. Subsequently, mutant K-ras proteins are driving tumor formation and progression52. Additionally, activated K-ras is associated with increased aggressiveness of CRC and reduced responsiveness to targeted therapies27. Numerous studies5,22,53,54 explored K-ras mutations in Saudi Arabia. These studies5,22,53,54 showed different prevalence of K-ras gene mutations including 35%, 42.85%, 42% and 56%. It is not clear why there is a wide range difference in the percentage of K-ras mutations in Saudi Arabia. A study2 was performed on K-ras mutations in cancerous tissue obtained from 56 Saudi sporadic CRC patients from the Eastern Province. K-ras gene mutations were detected in the cancer tissue of 24 out of the cases studied. Of these, 11 had exon 4 mutations localized between codons 134 and 150, while 13 had mutations in exon 2, affecting codons 12 and 13. No mutations were identified in codon 61 of exon 3. The 11 cases with exon 4 aberrations harbored p. Ala146Thr (a missense mutation), p. Ala134Val, p. Arg135Lys, p. Gln150Stop, p. Lys147Lys, p. Gln150Stop, p. Gln150Stop, p. Gly138Gly, p. Arg149Gly, p. Gly138Gly, and p. Gly138Gly mutations. Missense K-ras mutations which altered the amino acid sequence of the protein (A134V, R135K, E143K and R149G) were distinguished. Moreover, two synonymous K-ras mutations (G138G and K147K) and, a nonsense truncating mutation (Q150X) were detected. E143K mutation is predicted to have a damaging effect on the protein. R149G mutation is predicted to be neutral, but molecular modeling showed that this mutation caused changes in the helix and loop chains near the GTP binding pocket. Q150X mutation introduced a premature stop codon.

Another study53, one of the largest studies investigated K-ras mutations, was accomplished via examining 300 CRC patients in KSA. Most mutations detected were at codon 12 (89%) and were associated with metastasis. The prevalence of mutated K-ras was 42% in patients and mostly in stages II-IV, suggesting that K-ras mutations were concomitant with advanced stage of CRC, shorter RFS and OS. Additionally, mutations in
K-ras
most commonly p. G12D, codon 13 (20%).
tients.
and clinicopathological features in CRC pa-
and its correlation with patients' characteristics
rospectively the frequency of
mutions were in codon 12 (75%),
right-sided tumors (57.1%). A
mutions were higher in young patients (≤ 50,
alyzed ret-
missions in four
cods (12, 13, 17, and 31) were recog-
in 26 patients. Several mutations in K-ras
were acknowledged in codon 12 (61.5% of all
 mutation, glycine was substituted by aspartate
Glycine substituted by arginine (G12R), glycine
substituted by alanine (G12A), and glycine sub-
stituted by cysteine (G12C). The presence of gly-
cine at position 12 seemed to be imperative for
appropiate K-ras gene functioning and disrup-
tion, or the replacement of this amino acid led to
failure in function efficiency. Other mutations in
K-ras recognized in codon 13 included glycine
was replaced by aspartate (G13D) and glycine sub-
stituted by cysteine (G13C). As for codon 17,
mutions in which serine was substituted with
arginine (S17R) were detected. Whereas in co-
don 31, a very rare mutation was identified, c.91
G > A, in which glutamic acid was replaced by
lysine.

Another study22 has investigated 99 CRC cas-
esia, identifying frequent mutations in K-ras (35%)
which were associated with poor disease-specif-
ic survival in cases with wild-type TP53.

LRP6
Parine et al34 (2019) verified a fourfold de-
creased risk of developing CRC in CC genotype,
compared to TT genotype at SNP rs2284396 in
LRP6 gene34.

MALAT1
Siddique et al24 Alsufiani (2019) displayed that
the expression levels of metastasis-associated
lung adenocarcinoma transcript 1 (MALAT1)
were significantly escalated in CRC patients.

MED12
MED12 encodes a member of mediator, a mu-
tiprotein complex involved in the transcrip-
tional regulation of many genes by mediating the in-
teraction of RNA polymerase with various tran-
scriptional factors. Siraj et al35 (2018) identified
three MED12 somatic mutations in 27 CRC pa-
tients, which expand the role of MED12 as a tu-
mor suppressor in CRC.

MEG3
Siddique et al24 (2019) revealed no significant
expression of MEG3 in CRC when compared with
control.

miRNAs
Several classes of non-coding RNAs (ncRNAs),
including microRNAs (miRNAs), exhibit diffe-
rential expression in many types of cancer, in-
cluding CRC, and their dysregulation promote
carcinogenesis. Al-Sheikh et al36 (2016) showed
significant rises in miR-29 and miR-92 and their
expression levels. Whereas miR-145 and miR-195
decreased in CRC tissues compared with adjacent
neoplasm-free mucosal tissues. Another study
performed by Fawzy et al37 (2020) identified note-
worthy escalation in miR-34a in CRC colon spec-
imens, specifying that miR-34a rs2666433 AA
and AG genotype carriers were more likely to
develop cancer than GG carriers. Additionally,
Al-Sheikh et al38 (2019) demonstrated that miR-
182-5p gene was amplified in CRC patients.

MLH1
Lynch syndrome (LS) is associated with ear-
ly onset of CRC and enhanced risk of many ex-
tra colonic malignancies. LS is mainly caused
by germline pathogenic mutations in DNA mis-
match repair (MMR) genes, mostly in four of the
genesis, MutL Homolog 1 (MLH1), MutS Homolog
2 (MSH2), MutS Homolog 6 (MSH6) and PMS1
Homolog 2 (PMS2). Rasool et al32 (2020) detect-
ed structural loss in the genomic regions of
MLH1 (3p23-p14.2), MSH2, MSH6, EPCAM (2p21-p16.3),
PMS2 (7p22.1) and MUTYH (1p34.1-p33) in LS
patients. This structural loss resulted in lower
expression of MLH1, MSH2, MSH6, PMS2, EPCAM
and MUTYH genes. Moreover, Ahmed
(2017)44 detected a positive MLH1 correlation in
around 27% and that MSI was present in more
than 90% of CRC patients with Lynch syndrome.

MMP-2
MMPs overexpression is associated with tumor
invasion, metastasis, and a worse prognosis. Two
studies confirmed that MMP2 C1306 T mu-
tation was significantly more common in colon
Saudi patients.
**NOTCH1**

The **NOTCH** gene family consists of four receptors (**NOTCH1**, **NOTCH2**, **NOTCH3**, and **NOTCH4**). Notch signaling plays an important role in several cellular processes, including proliferation, epithelial cell polarity/adhesion and apoptosis. Alanazi et al. (2021) demonstrated that SNPs rs3124591 in **NOTCH1** and rs3820041 in **NOTCH4** did not exhibit any association with CRC. Although rs1043994 in **NOTCH3** was not associated with CRC in the overall analysis, it displayed a significant association with CRC in males. The GA heterozygote males of this SNP were two-fold higher risk of CRC development compared to GG homozygotes.

**PANDAR**

Siddique et al. (2019) showed that the expression levels of the promoter of CDKN1A antisense DNA damage-activated RNA (**PANDAR**) were significantly enlarged in CRC patients.

**PARP-1**

Poly (ADP-ribose) polymerase-1 (**PARP-1**) has a crucial role in DNA damage repair and is involved in many cellular processes. Thus, **PARP-1** gene polymorphisms are associated with the risk of various carcinomas, including colon cancer. Alshammari et al. (2014) demonstrated that SNPs in **PARP-1** gene, including Met129Thr, Val762Ala, and Lys940Arg, did not show any association with CRC risk, while Lys933Asn and Lys945Asn showed significant association with CRC among Saudis. Another study formulated by Alhadheq et al. (2016) demonstrated that SNP rs8679 diminished the susceptibility to colorectal cancer at heterozygous TC allele and at minor allele C.

**PCAT6**

Siddique et al. (2019) revealed no significant expression of **PCAT6** in CRC patients.

**PIK3CA**

**PIK3CA** mutations were significantly associated with poor disease-specific survival in cases with wild-type **TP53** in CRC Saudi patients.

**POLD1 and POLE**

**POLD1** and **POLE** encode the catalytic subunit of the polymerase enzyme complexes Epsilon (ε) and Delta (δ), which play an imperative part in DNA replication and repair. Siraj et al. (2020) demonstrated that four variants in **POLE** gene were associated to an increased risk of CRC, besides the three **POLE** variants p. His342Tyr, p. Gly395Glu, and p. Thr457Met, which were found in early onset CRC patients. Furthermore, **POLD1** variant of c.932G > A: p. Arg311His was established in a late onset patient, causing loss of function of **POLD1**. Generally, low **POLE** protein expression exhibited a substantial link with lymph node involvement and grade III tumors. Whereas for **POLD1**, low expression was related to adenocarcinoma histology, larger tumor size and stage III tumors.

**PRNCR1**

Prostate cancer non-coding RNA (**PRNCR1**) propagates colon cancer from epithelial cells, causing an increase the tumor size in CRC patients. One SNPs, rs1456315, in **PRNCR1** gene revealed an association with CRC with the homozygous CC variant allele. This risk association was observed among younger age patients (≤ 57) and in female patients. Three other SNPs rs1016343 (C > T), rs13252298 (A > G), and rs16901946 (A > G) in **PRNCR1** gene did not display any association with CRC.

**P35 and PTEN**

Mutation of **p53** frequently happens in almost half of all human malignancies and contributes to tumor progression. A retrospective cohort study was performed out over a five-year period in which 130 samples were recruited. The study revealed that a **P53** positive expression was found in 25.4%, whereas loss of **PTEN** expression was recognized in 32.3% CRC patients.

**RETN**

Resistin gene (**RETN**) codes a peptide hormone called resistin, which is secreted predominantly by adipose tissue, in particular from adipocytes and macrophages. A study accomplished by Alharithy (2014) indicated that SNPs in **RETN** gene rs1862513 (C-420G) and rs3745367 (G+299A) had increased the risk of colon cancer. Additionally, carriers of the heterozygous (GA) genotype of SNP 299 had a significantly higher colon cancer risk than carriers of the wild (GG) genotype.

**SFRP3**

As already mentioned, Parine et al. (2019) verified that genetic variants in **SFRP3** (rs7775) gene were correlated with considerable protection against CRC progression. Women having Gly at codon 324 (rs7775) of **SFRP3** have 2.5-fold lower risk of developing CRC compared to those having...
Arg at this locus. Thus, SFRP3 gene may serve as a protective marker in female patients harboring the minor allele G14.

**SMAD4**

Frequent mutations in SMAD4 were identified and correlated with CRC development, as showed by a study done by Dallol et al22 (2016).

**TDG**

In addition to its DNA repair function, thymine DNA glycosylase (TDG) is also involved in other critical cellular processes78. SNP rs4135113 in TDG gene showed a significant risk association between its genotype AA and the minor allele A in CRC Saudi patients in general, and in patients aged more than 57 years. On the other hand, SNP rs1866074 in TDG gene presented a protective association between the GG allele and the additive (AG+GG) allele in CRC patients. Other four SNPs (rs4135050, rs4135066, rs3751209, and rs1882018) in TDG gene showed no association with CRC patients in the Saudi population79.

**TIGAR**

The TP53-induced glycolysis and apoptosis regulator (TIGAR) regulates glycolysis by acting as fructose bis-phosphatase (FBPase) and modulate reactive oxygen species80. Al-Khayal et al81 (2016) revealed that TIGAR expression was found in 68% of the tumor samples with nuclear localization and was significantly amplified in early stages (stage I and II) and late stages (stage III and IV) of CRC. Thus, TIGAR expression may be used as a biomarker for CRC recognition and even as a target for developing therapeutics for CRC treatment81.

**TLRs**

Toll-like receptors (TLRs) represent the first line of defense against invading pathogens, initiating inflammatory responses; thus, they play a key role in immune cell regulation, survival, and proliferation82.

**TLR2**

A study83 goal was to determine the association of TLR2 SNPs (rs3804099, rs3804100, and rs4696480) and the risk of colon cancer development in a Saudi Arabia population83. TLR2 rs3804099 and TLR2 rs4696480 SNP were closely associated with CRC susceptibility. However, TLR2 (rs3804100 C > T) disclosed no association with CRC susceptibility in Saudi patients.

**TLR4**

Semlali et al84 (2016) revealed a clear association between TLR4 rs10759931 polymorphism, the G allele, and susceptibility to CRC development risk in the Saudi Arabian population. Also, the TLR4 rs2770150 is associated with CRC in women aged over 50 years and is linked to the decreased levels of female sex hormones during the post-menopausal period. Whereas TLR4 SNPs rs10759932 and rs4986790 appeared not to have any association with colon cancer84.

**TLR6**

Semlali et al85 (2019) illustrated that Val/Met genotype of rs3796508 of TLR6 gene had a significantly higher frequency in the control group than in the CRC male cases, suggesting that TLR6 rs3796508 is a protective factor against CRC in the older male Saudi population. Two other non-synonymous SNPs S249P and V327M were common in a few patients and were predicted as being damaging85.

**TLR 9**

TLR 9 is the only TLR which is administered systemically and has shown substantial evidence of anticancer activity in human clinical trials86. Semlali et al87 (2016) clarified a significant association between the TLR-9 rs187084 SNP and CRC risk in female patients. T allele exhibited lower frequency (2.8 times) in female cancer patients. Additionally, TLR-9 rs352139 and rs352144 SNPs were found to be suggestively correlated with colon cancer development when the tumor was located in the rectal area, but not in the colon area localization. On the other hand, all three TLR-9 SNPs rs352144, rs187084 and rs5743839 were not associated with colorectal cancer in males.

**TNF-α**

Hamadien et al88 (2016) verified that TNF-α SNPs, -308 and -857, were not associated with CRC. TNF-α-238 (G/A) genotype was significantly concomitant with high risk of CRC. This is because AA genotype of -238 G/A SNP was observed at considerably higher proportion in CRCs.

**PTEN**

Loss of PTEN has been reported89 in many types of cancers, including CRC. The overall loss of PTEN expression (negative) was identified in 32.3% of the CRC patients84. In another study in KSA, PTEN was inactivated in 66.1% of the 51 CRC cases, and PTEN loss was more frequent in
Gene’s polymorphisms in colorectal cancer Saudi patients

**CRC**

A third study identified **PTEN** mutations in 13% of CRC cases.

**TP53**

Interestingly, **KRAS** or **PIK3CA** mutations were significantly associated with poor disease-specific survival in cases with wild-type **TP53**. Mutant **TP53** may serve as an emerging target for cancer treatment using small molecule therapeutics that restores wild-type **TP53** function, inducing cell cycle arrest and apoptosis.

**TSLP**

**TSLP**, **IL-7** like cytokine, triggers **STAT1**, **STAT3**, **STAT4**, and **STAT5**, stimulating the proliferation, development, differentiation, migration, and death of apoptotic cells, depending on the type of stimuli and cells. **Semlali et al.** (2021) established that **TSLP** rs10043985 presented a strong correlation with CRC Saudi patients, indicating that this mutation in the promoter region of **TSLP** gene might play a detrimental role in CRC. However, rs2289276 SNP of **TSLP** gene did not show any relation with CRC. On the other hand, **IL-7R** rs1053496 SNP showed no association with CRC in female subjects or in CRC patients who are more than 57 years of age.

**UCA1**

**Siddique et al.** (2019) revealed no significant expression of IncRNA uterine carcinoma-associated 1 (**UCA1**) in 63 CRC cases when compared with control.

**VDR**

More than sixty SNPs of the **VDR** gene, located in the promoter region, have been related to cancer occurrence and prognosis. **Apol** (rs7975232), **TaqI** (rs731236), **BsmI** (rs1544410) and **FokI** (rs10735810) are **VDR** SNPs that affect **VDR** gene expression and mRNA stability. **Alkhayal et al.** (2016) did not observe any association of the four **VDR** polymorphisms with CRC risk in the overall analysis. However, **Apol** and **BsmI** loci were associated with CRC in elderly and female patients, respectively. In contrast, **Apol** and **BsmI** loci displayed an increased risk for the disease. In contrast, heterozygous (Bb) and homozygous (bb) carriers of the **BsmI** SNP (rs1544410) had significantly lower risk for CRC. Finally, for the **FokI** SNP (rs2288570), there was no association with CRC risk. Another study performed by **Al-Ghafari et al.** (2020) found that the **VDR** SNPs **Apol** and **TaqI** upsurge the risk of CRC, whereas **BsmI** lessens the risk of CRC in the selected Saudi population.

**XPD and XRCC1**

The xeroderma pigmentosum group D (**XPD**) protein participates in nucleotide excision repair (**NER**), one of DNA repair pathways. X-ray repair cross-complementing group 1 (**XRCC1**) is acknowledged to participate in base excision repair (**BER**). **Karam et al.** (2016) demonstrated no significant difference in **XPD** Lys751Gln polymorphism in CRC. Regarding **XRCC1** polymorphism, they demonstrated there was an association between the GG genotype of **XRCC1** polymorphism and the increased risk of CRC. Moreover, **XRCC1** (AG + GG) polymorphism may be associated with increased clinic pathological parameters of CRC.

**Genetic Variation Irrelevant to CRC**

**ABCB1** (C3435T, T129C and T1236C), **ABCC1** (G128C), **CASS** (rs3804594), **IL-17F** (rs763780), **NOTCH1** (rs3124591), **NOTCH4** (rs3820041), **PRN** (rs1016343), **SNP** (rs13252298), and **TDS** (rs16901946), **TDG** SNPs (rs4135050, rs4135066, rs3751209, rs1882018), **TLR2** (rs3804100), **TLR4** (rs10759932, rs4986790), **TLSP** (rs2289276), **TSLPR** (rs36139698, rs36177645, rs36133495), and **TNF-α** (−308 and −857) polymorphisms were unrelated to CRC risk in KSA. Additionally, Met129Thr, Val762Ala, and Lys940Arg and **XP** Lys751Gln polymorphisms in **PARP-1** were unconnected to CRC risk in Saudi population. Moreover, genotypes **CYP2E1**T6, **GSTM1**T0 were not associated with the CRC development. Besides, no significant expressions of **BCAR4**, **CCAT2**, **MEG3**, **PCAT6**, **UCA1** were found in CRC samples, nor did any genotypes of **GSTP1** showed association with CRC development. Finally, three **TLR-9** SNPs (rs352144, rs187084 and rs5743839) were unrelated to colorectal cancer in males, whereas **IL-7R** rs1053496 SNP showed insignificant association with CRC in female subjects or in CRC patients who were more than 57 years of age.

**Genetic Variation Protected Against CRC**

Genetic variants in **ABCB** (rs3435, TT genotype, G2677T, female), **ADIPQ** (T45G, G allele), **CTNNBI** (rs4135385, GG genotype), **SFRP3**
Genetic Variation Strongly Associated with CRC

ABCC1 (C218T, CT genotype), ADIPQ (G276T, T allele), CYP1A1 (wt*2A genotype), KIR (3DS1, 2DS1), IL-17A (G197A, A allele in males), MMP2 (C1306 T), NOTCH3 (rs1043994, GA genotype, males), PRNCR1 (rs1456315, CC, young age, female), RETN (rs1862513 and rs3745367), TDG (rs41351130, AA genotype), TLR2 (rs3804099, rs4696480) polymorphisms exhibited a substantial augmented CRC risk of development in CRC Saudi patients. Additionally, TLR4 (rs1075993, G allele; rs2770150 women aged over 50 years), TLR-9 (rs187084, female), TNF-α (-238 (G/A) genotype), XRCC1 (GG genotype) mutations displayed a significant amplified CRC development. Furthermore ATR, ATM, BM1, CCAT1, Chkl, Chk2, COX-2, FoxMi, FSCN1, Ki67, MALAT1, miR-29, miR-34a, miR-92, miR-182-5, PANDAR, PIK3CA, TIGAR over-expression showed a correlation with CRC Saudi inhabitants. Besides, significant associations between BRAF (E586E, Q609L, and M620I) and PARP-1 (Lys933Asn and Lys945Asn) and CRC risk have been detected. Moreover, ALK gene amplification and gain in copy number and gene mutations in APC, EGFR, FBXW7, TP53, PTEN, K-ras were concomitant in CRC Saudi population. Structural loss of MLH1 (3p23-p14.2), MSH2, MSH6, EPCAM (2p21-p16.3), PMS2 (7p22.1) and MUTYH (1p34.1-p33), with subsequent lower expression of MLH1, MSH2, MSH6, PMS2, EPCAM and MUTYH genes, were recognized in LS patients and future CRC patients.

Conclusions

In this review, we performed a comprehensive literature review concerning CRC genetics to offer an insight into the CRC genes in Saudi Arabia patients. All these gene mutations may be used as diagnostic and/or prognostic genetic marker in CRC Saudi patients and could offer a potential therapeutic target for CRC management. For each of these genes, we tried to explain a little about the gene and its role in cancer development and clinical phenotype on the Saudi patients and the mutation occurring in these genes. Several genes’ mutations were included in this review, from which some genetic variations were either associated or strongly related to, nor even protected against the CRC development.

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Conflicts of Interest

The authors declare no conflicts of interest.

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Authors’ Contributions


References


Gene's polymorphisms in colorectal cancer Saudi patients


