

BIOINFORMATICS DISCOVERY OF ANTI CANCEROUS COMPOUNDS FOR THE TREATMENT OF OVARIAN CANCER

(Submitted in partial fulfillment of the requirements Under Bachelor of technology
degree in Biotechnology)

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SATHYABAMA

**INSTITUTE OF SCIENCE AND TECHNOLOGY
(DEEMED TO BE UNIVERSITY)**

Accredited with Grade "A" by NAAC

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BONAFIDE CERTIFICATE

This is to certify that this Project Report is the bonafide work of GURAJA SREEJA (37230021) and V.E.MOHANALAKSHMI (37230047) who carried out the project entitled **BIOINFORMATICS DISCOVERY OF ANTI CANCEROUS COMPOUNDS FOR THE TREATMENT OF OVARIAN CANCER** under the supervision of DR.BHAVANILATHA, Ph.D. Sathyabama Institute of Science and Technology, Chennai from JANUARY 8TH TO MARCH 20th 2021

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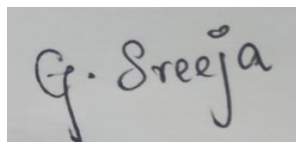
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DECLARATION

I GURAJA SREEJA (37230021) hereby declare that the Project Report entitled **BIOINFORMATICS DISCOVERY OF ANTI CANCEROUS COMPOUNDS FOR THE TREATMENT OF OVARIAN CANCER** done by me under the guidance of Dr.BHAVANILATHA, Ph.D., at Sathyabama Institute of science and technology is submitted in partial fulfillment of the requirements for the award of Bachelor of Engineering in Biotechnology

DATE: 15.01.2021
PLACE: Chennai

A rectangular box containing a handwritten signature in black ink. The signature appears to be 'G. Sreeja'.

SIGNATURE OF THE CANDIDATE

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ABSTRACT

Cancer is one of the main reason for death which has been constantly increasing day by day. However, Ovarian cancer has been reported in large number every year constantly as per the statistics also treatments like chemotherapy have side effects which are evident in most of the research .Therefore the majority of research work is carried out on cancer-targeted drugs which are derived from plant. Nigella sativa also called as Black cumin is a natural compound with anti-cancer properties. A bioinformatics approach was carried out to understand the activity of phytochemicals of black cumin against ovarian cancer. The active molecules were also subjected to pharmacokinetic and pharmacodynamics properties. However, the efficiency of the novel drug compounds could be validated only through In-Vivo and In- Vitro results

Keywords: Cancer, anti-cancerous compounds, phytochemicals of nigella sativa, docking, PubChem, PDB.

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LIST OF ABBREVIATIONS

FTC - Fallopian Tube Cancer

PPC - Primary Peritoneal Carcinoma

CCC - Clear Cell Carcinoma

ADME - Absorption, Distribution, Metabolism, Excretion

NCBI - National Center for Biotechnology Information

GCT- Granulosa cell tumor

SLCT- Sertoli-Leydig cell tumor

SDF - The Spatial Data File

3D – 3 Dimensional

PDB- Protein Data Bank

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1. INTRODUCTION

Cancer is a group of diseases which involves rapid cell growth and spread to other parts of the body. Our body is mainly composed of many millions of tiny cells and each a self-contained living unit. Normally, each cell combines with the others that compose tissues and organs of our body. Normal cells in the body grow and divide for a period of time and then it stops growing and dividing. Unlike normal cells, cancer cells continue to grow fast and divide for their whole lives, replicating into more and more harmful cells. The abnormal growth and division occurred in cancer cells can cause damage in these cells. As cancer cells divide and replicate themselves, they often form into a lump of cancer cells known as a tumor. Tumors spread and invade into nearby tissues. As tumors grow some cancer cells and spread to other parts of the body through blood or the lymph system and form new tumors. Tumors are two forms benign and malignant. Benign tumors are not cancerous, thus they do not grow and spread to the extent and form cancerous tumors. Benign tumors are usually not life threatening. On the other hand, malignant tumors grow and spread to other areas of the body.

THE MOST COMMON CAUSES OF CANCER ARE

- Smoking and tobacco
- Diet and physical activity
- Sun and other types of radiation

THE SIGNS AND SYMPTOMS OF CANCER ARE

- A lump
- Abnormal bleeding
- Prolonged cough
- Unexplained weight loss and
- Changes in the bowel moments.

STAGES OF CANCER:

1. Stage 0: This stage of cancer is considered to be precancer or cancer that hasn't spread from where it started and it's called in situ.
2. Stage 1: In this stage the cancer is small and hasn't spread. It's also known as localized.
3. Stage 2: In this stage the cancer is locally spread in surrounding tissue.
4. Stage 3: In this stage the cancer has spread regionally or into the lymph node.
5. Stage 4: This stage is said to be the advanced stage of cancer where the cancer cell spread to the distant tissues or organs.

1.1 OVARIAN CANCER:

A woman's reproductive system has two ovaries, connected with fallopian tubes which connects the ovary to uterus followed by cervix, vagina and vulva. These are the other organs which help to understand how ovarian cancer has spread and the stages of ovarian cancer.

- **VULVA:** This is the external female genitalia. These are the parts outside the body
- **VAGINA:** This is the passage that connects the uterus to the outside of the body. It's also called as the birth canal.
- **CERVIX:** This is the lower part of the uterus that connects to the vagina.
- **UTERUS:** This is a hollow, pear-shaped organ that holds a growing baby. It's also called the womb.
- **TWO FALLOPIAN TUBES:** The Fallopian tubes, also known as uterine tubes, salpinges or oviducts, are tubes that stretch from the uterus to the ovaries, and are part of the female reproductive system. The fertilized egg from the ovaries of female mammals passes through the fallopian tube to the uterus. The Fallopian tubes are simple hair-like extensions which are called as the cilia that carries the fertilized egg.

Ovarian cancer is cancer that mainly starts in the ovaries or at the end of the fallopian tubes next to the ovary. Fallopian tubes are set of tubes that connect the ovaries to the uterus. Fallopian tube is the path where the egg travels from the ovaries to the uterus. Many types of tumors can start growing in the ovaries, some are benign. This means that they are not cancer. Benign tumors don't spread. They can usually be treated by removing one ovary or part of ovary, ovarian cancer, however, is a malignant tumor. If a cancerous tumor isn't treated, it can grow and spread to other parts of the body. The ovary is made up of many layers of cells Early-stage ovarian cancer, in which the disease is confined to the ovary and easy to be treated successfully. At this final stage, ovarian cancer is more difficult to treat.

1.1.1 STAGES OF OVARIAN CANCER:

1. Stage 1: In this stage the cancer has not spread beyond the ovaries

Stage 1A: In this stage the cancer is only in one ovaries.

Stage 1B: In this stage the cancer is in both ovaries.

Stage 1C: In this stage cancer is found inside the ovaries or on the surface of the ovaries

2. Stage 2: In this stage of ovarian cancer, the cancer is in both the ovaries and has spread within the pelvis.

Stage 2A: In this stage it is found that the cancer has gone from the ovaries to the fallopian tubes, the uterus or both.

Stage 2B: This stage indicates that the cancer has been migrated to the nearby organ like the bladder, rectum.

3. Stage 3: This stage of ovarian cancer indicates that the cancer is found in one or both ovaries, it is also found that the cancer is spread to the lining of the abdomen and to the lymph nodes in the abdomen.

Stage 3A: In this stage the cancer is found in the other pelvic organs and in lymph nodes within the abdominal cavity or the abdominal lining.

Stage 3B: This stage of cancer indicates that the cancer has spread to nearby organs within the pelvis. The cancer cells can be found in the outside of the spleen or liver or in the lymph nodes.

Stage 3C: In this stage the cancer cells has spread in a large amount and the cancer cells are found outside the spleen or liver or it has spread to lymph nodes.

4. Stage 4: This stage of ovarian cancer is the most advanced stage of ovarian cancer. In this stage the cancer has spread to the distant area or the organ in the body.

Stage 4A: The cancer cells are found in fluid around the lungs.

Stage 4B: This stage of cancer indicates that the cancer is reached to the inside of the spleen or liver, distant lymph node or the distant organs such as skin, lungs or brain.

1.1.2 TYPES OF OVARIAN CANCER:

Ovarian cancers are classified into 3 major types which are named after the type of cell they come from, they are,

- Epithelial ovarian tumor
- Ovarian germ cell tumor
- Ovarian sex cord-stromal tumor

These three types can develop into a different types of tumors, each type differs in how it spreads and how it's treated.

1.1.3 EPITHELIAL OVARIAN TUMOR:

Epithelial ovarian cancer is said to be the most common type of ovarian cancer. It is one of the four types of tissue in the body which covers the outer surfaces of the organs and blood vessels and the inner surfaces of cavities in many internal organs. Most ovarian cancers approximately 85-90% fall into this category. Epithelial ovarian cancer is one of the most common gynecologic malignancies, in which 50% of all cases occurring in women who are older than 65 years. These cancers are diseases in which malignant cancer cells form in the tissue covering the ovary, or lining the fallopian tube or peritoneum and are called carcinomas. Fallopian Tube Cancer (FTC) and Primary Peritoneal Carcinoma (PPC) and epithelial ovarian cancer these three

are often grouped together because they found to be most similar and they follow similar treatments.

Epithelial ovarian cancer types:

- Serous carcinoma (high and low grade),including primary peritoneal and fallopian tube
- Clear Cell Carcinoma(CCC)
- Endometrioid
- Mucinous

1.1.4 OVARIAN GERM CELL TUMOR:

Ovarian germ cell tumors begin in the egg or sperm of the body. In women, this type begins in the cells that forms eggs in the ovaries, known as germ cells.

Benign Germ Cell Tumors

Most germ cell tumors are benign (non-cancerous). Mature teratomas are considered to be the most common type of benign germ cell tumor. These tend to occur in women of reproductive age mostly teens and it is often called a dermoid cyst. Women with benign germ cell tumors such as mature teratomas are treated by removing the part of the ovary that has the tumor (ovarian cystectomy) or by removing the entire ovary. There may be few symptoms like abdominal pain or constipation may also occur, and when the tumor gets bigger, an abdominal mass may be seen.

Malignant Ovarian Germ Cell Tumors

Ovarian germ cell malignant tumors are uncommon, and approximately causes 5 percent of ovarian cancer cases. This cancer can occur in women of any age, but are more often found in young girls. These tumors often affect only one ovary and are generally curable and this can be found and treated early.

Immature Teratomas

Immature teratomas are one of the type of malignant ovarian germ cell tumors. In which they typically occur in young girls under the age of 18. In cases where tumors have not spread beyond the ovary, this cancer can be treated by surgical removal of the ovary and if they spread beyond the ovary, chemotherapy is typically recommended in addition to surgery.

Dysgerminoma

Dysgerminoma, is considered to be the most common malignant form this form mainly affects women in their teens or twenties. These types of tumors don't spread quickly, and are usually treatable. More than 75% of women are cured by the removal of the ovary, and if the cancer has spread, surgery along with radiation or chemotherapy can control or cure the disease in approximately 90% of patients. New types of treatment are being tested through clinical trials, such as high-dose chemotherapy with bone marrow transplant. Malignant germ cell tumors follow the same staging system as epithelial cancers and primary peritoneal cancers.

1.1.5 OVARIAN SEX-CORD STROMAL TUMOR:

Malignant ovarian stromal tumors are considered to be rare and represent approximately 1.2% of all primary malignant ovarian tumors. Sex cord-stromal tumors are basically present with signs of hormonal production, such as hirsutism and virilization, menstrual changes or early puberty and are often found in young adult. Ovarian stromal tumors can be detected early and have a 75% survival rate.

Benign Ovarian Sex Cord-Stromal Tumors

- Fibromas
- Thecomas

Malignant Ovarian Sex Cord-Stromal Tumors

- Adult granulosa cell tumor (GCT)
- Juvenile granulosa cell tumor
- Sertoli-Leydig cell tumor (SLCT)

Mixed Forms can include

- Granulosa and Sertoli-Leydig elements
- Granulosa and Sertoli elements
- Gynandroblastoma

Recently individuals who has sex cord-stromal tumors receive treatment that is similar to that of the individuals with germ cell or epithelial tumors. Recurrent disease may be treated with surgery. Radiation therapy is considered to be useful in settings of recurrent disease.

1.2 LIST OF OVARIAN CANCER CAUSING GENES:

- BRCA1
- BRCA2
- TP53
- NBN
- RAD50
- RAD51
- CHEK2
- PIK3CA
- MMR

1.2.1 BRCA1:

The risk of ovarian cancer for the average woman is about 2% in her lifetime. The risk of ovarian cancer in women with a BRCA1 mutation is estimated to be 39–46% by age 70 years. Mutations in BRCA1 gene located area are associated with increased risks for breast and ovarian cancer. In cancer risk evaluation programs the increasing numbers of BRCA1 gene mutation carriers are being identified and said that the highest cancer risk are derived from families. Risks for these families are estimated at 85% for breast cancer and 60% for ovarian cancer.

1.2.2 BRCA2:

The secondary hereditary breast cancer gene BRCA2 is a tumor-suppressor gene that participates in DNA damage repair. There is an increased risk of ovarian cancer development in patients with germline BRCA2 mutations. BRCA2 is a gene on chromosome 13 that normally helps to suppress cell growth. A person who has increased mutations in a BRCA2 gene has a higher risk of getting breast, ovarian and other types of cancer. In women, the risks associated with a BRCA2 mutation appear to be about the same as those for BRCA1 mutation around 50 to 85 percent risk of developing the disease by the age of 70. The risk of ovarian cancer is also increased between 16 and 27 percent by late age.

1.2.3 TP53

TP53 is a nuclear transcriptional regulator which takes part in multiple cellular process. In order to maintain homeostatic and genomic integrity the TP53 gene binds to DNA and controls the expression of hundreds of target genes. TP53 is found to be the most frequently mutated gene in cancer as more than 50% of the types of human cancer have TP53 mutation. Over 36,000 TP53 mutation were found and approximate of 80% of them were missense mutation with amino acid and substitutions. Through the analysis of IARC Database it is identified that 2,329 kinds of TP53 mutations causes human ovarian cancer.

1.2.4 NBN:

NBN gene is responsible for making a protein called Nibrin. This gene is located on chromosome 8. NBN gene encodes protein and it is a member of MRE/RAD50 double stranded break repair complete. The NBN gene has been altered 1.63% in the ovarian carcinoma patients.

1.2.5 RAD50:

RAD50 gene encodes a protein that functions in double- strand break repair.

Mutations, and frame shift deletions and insertions are observed in cancers such as ovarian cancer, intestinal cancer, and stomach cancer. RAD50 is modified in 1.05% of ovarian carcinoma patients. RAD50 is an inclusion criterion in 14 clinical trials for ovarian carcinoma, of which 10 are open and 4 are closed. The trials that contain RAD50 status and ovarian carcinoma as inclusion criteria 5 are phase 1 and 4 is open, 2 are phase 1/phase 2 and 1 is open, 6 are phase 2 and 5 is open, and 1 is phase 3 and 0 open.

1.2.6 RAD51:

The RAD51 gene is potentially associated with the risk of ovarian cancer. RAD51 mutation carriers has a family history of ovarian cancer. It is analyzed that there are three RAD51 gene in germ line DNA. Mutations in the RAD51B, RAD51C and RAD51D genes has risk for invasive epithelial ovarian cancer. In an overall amount of epithelial ovarian cancer cases 0.81% of the cases had a mutation in one of these three genes.

1.2.7 CHEK2:

CHEK2 gene is also called as checkpoint kinase 2 .People who inherit mutations in the gene are at an increased risk for certain types of cancer. The gene is responsible for making a protein known as CHEK2, which becomes active when DNA within the cell is damaged or DNA strand breaks. CHEK2 mutations are found in a variety of cancer types, including ovarian, breast, colon, prostate, kidney, thyroid, and other cancers. The mutations is also found in some brain tumors and in osteosarcoma, a form of bone cancer.

1.2.8PIK3CA:

The PIK3CA gene is responsible for making the P110 alpha protein and it belongs to the phosphatidylinositol 3- kinase (PI3Ks) family, which plays an important role in proliferation and cell survival through the PI3CK/AKT signaling pathway. The somatic activating mutation of this gene causes several types of cancer. The PIK3CA mutation were analyzed with 109 advanced ovarian carcinoma.

1.2.9 MMR:

The MMR genes are otherwise called as DNA miss match repair genes. Deficiency of the MMR gene is considered to be the most common cause of ovarian cancer. In this gene the deficient cells usually have many DNA mutations and that leads to cancer.

1.3 TREATMENT OF OVARIAN CANCER:

The treatment for ovarian cancer mainly involves surgery, chemotherapy, targeted therapy and immune therapy or it can combine both chemotherapy and surgery.

1.3.1 SURGERY

Operations to remove ovarian cancer include:

Surgery to remove one ovary: In early stages when the cancer hasn't spread beyond one ovary, surgery may involve removing the affected ovary and its fallopian tube.

Surgery to remove both ovaries: In this case the surgeon may remove both the ovaries and the fallopian tube.

Surgery for advanced cancer: In the advanced stage of cancer the doctor may recommend chemotherapy followed by surgery to remove the cancer as much as possible.

1.3.2 CHEMOTHERAPY

Chemotherapy is drug treatment that has a combination of two drug or a single drug and it uses chemicals to kill fast-growing cells in the body, including cancer cells. Chemotherapy drugs can be used in many different types it can be injected into a vein or can be taken by mouth. Sometimes the drugs can be directly injected into the abdomen. In advanced stages chemotherapy is often used after surgery to kill any cancer cells that might remain. It can also be used before surgery to control or stop the growth of cancer cells.

1.3.3 TARGETED THERAPY

Targeted therapy are drug or medicines that are used to target treat specific genes and proteins that are involved in the growth of the cancer cells. Targeted therapy are used to treat advanced epithelial ovarian cancer. This therapy attack only the cancer cells and leaves the normal cells unaffected.

1.3.4 IMMUNOTHERAPY

Immunotherapy is also called as Immune-Oncology in which the immune system fights cancer cells. These treatment strengthens immune system so that the immune system can find and attack cancer or stops them from growing.

1.3.5 SIDE EFFECTS:

TABLE 1.3.5.1 Side effects of surgery, chemotherapy, targeted therapy, immunotherapy.

S.NO	THERAPY	SIDE EFFECTS
1.	SURGERY	Bleeding and need for blood transfusion, bladder problem, Lymphedema, fertility problem, blood clots, wounds or blood infection and bowel obstruction.
2.	CHEMOTHERAPY	Nausea,vomiting, loss of appetite, sore mouth and throat, low blood cell count,infection, hairloss, fatigue and fertility problem.
3.	TARGETED THERAPY	Fatigue, headaches,muscle and joint pain,sore mouth, loss of apetite, nausea , vomiting, diarrhea and low blood cell count
4.	IMMUNOTHERAPY	Skin rash, fatigue, nausea, vomiting, joint pain, itching,skin rash and cough

Most of the therapy result in side effects but natural plant are not harmful and there are very less chances of getting side effects so we choose plant spices for the treatment of ovarian cancer. Many siddha and ayurveda treatments have used *Nigella sativa* for ovarian cancer treatments and resulted with less and no side effects.

Ovarian cancer is well known for its highest mortality rate in gynecological malignancy. *Nigella sativa* is known for its potential as a traditional herbal healing medicine for many ailments. *Nigella sativa* is a potent anti-ovaraian cancer agent, which inhibits tumor growth by modulating apoptotic activity.

1.4 INTRODUCTION TO NIGELLA SATIVA:

Nigella sativa commonly known as black cumin is a small black seed that comes from a flowering plant in the Ranunculaceae family. *Nigella sativa* originates from southwest Asia and the Mediterranean, now it grows throughout India, the Middle of East, and Europe. *Nigella sativa* has been used as herbal medicine for centuries to treat certain health conditions including asthma, bronchitis, and inflammation. It is also used as a food preservative and it is used as a spice. The seeds of *Nigella sativa*, commonly known as black seed or black cumin, *Nigella sativa* seeds have wide therapeutic effects and have been reported to have significant effects against many ailments such as diseases in skin, gastrointestinal problems, anorexia, jaundice, dyspepsia, conjunctivitis, diabetes, hypertension, cough, headache, bronchitis, influenza, fever, eczema, rheumatism. Around 2000 years ago *N. sativa* seeds have been used as folk medicine traditionally in middle eastern for the treatment of many disease . The seeds of *nigella sativa* has been used were used as pungent appetizer ,aromatic, thermogenic, diuretic, expectorant, purgative, stimulant, sudoriferous, sedative, and carminative. Black cumin seeds have a history of use in Arabic herbal medicine to treat many diseases such as skin disease. Therefore *nigella sativa* is said to be the best medicine in curing disease.

From literature survey the list of phytochemicals of *nigella sativa* has been taken.

1.4.1 LIST OF PHYTOCHEMICALS OF NIGELLA SATIVA:

- Alpha phellandrene (0.45%)
- Alpha pinene (1.11%)
- Alpha thujene (6%)
- Beta myrcene (0.21%)
- Beta pinene (7%)
- Borneol (0.43%)
- Camphene (11%)
- Camphor (1.0%)
- Carvacrol (1%)
- Carvone (0.32%)
- GAMMA-terpinene (5.12%)
- Limonene (0.13%)

- P-cymene (32.05%)
- Sabinene (1%)
- Terpinolene (0.23%)
- Thymol (10.12%)
- Thymoquinone (20.32%)

1.5 BIOINFORMATICS DRUG DISCOVERY:

Bioinformatics Drug discovery is a step by step process by which new drug are discovered. The drug discovery has its beginning in the 19th century by John Langley in the year 1905. Bioinformatics deals with the growth and development in the databases like protein sequences, nucleic acid sequences and structures. The major thing to identify a drug is to find the drug target and then study the target. This process helps us to check the potential for failure in clinical testing and approval. The aim of the study is to identify lead compounds and the further study can be done in two stages preclinical and clinical pharmacology.

2.LITERATURE SURVEY

Cancer, the uncontrolled growth of cells, is a major cause of death throughout the world. Donna S. Shewach, *et al.*, (July 25 2014) describes that In 2007, it killed ~7,900,000 people worldwide, a value that represents ~13% of total deaths. In the U.S., the number of deaths caused by cancer is second only to that from cardiovascular disease. While great strides have been made in the treatment of cancer over the past 50 years, it continues to be a major health concern and therefore, Donna S. Shewach's efforts have been devoted to searching for new therapeutic approaches.

Pei-Wei Zhang, *et al.* (march 30 2015) describes that, Identifying cancer-specific genes involved in tumor genesis and cancer progression is one of the major ways to understand the pathophysiologic mechanisms of cancers and to find therapeutic drug targets. Many efforts have been made to identify cancer biomarkers by using gene expression profiles. However, the robustness of microarray-derived biomarkers is very poor; this is in part because the robustness can be easily influenced in gene expression levels by small environmental changes. Without the evaluation of protein expression levels, there would be no way to illustrate causes of tumor proliferation and differentiation. Therefore, better understanding of the translational states of these genomes will bring us a step closer to finding potential drug targets and to illustrating off-target effects in cancer medicine.

Giuseppina D Andrioli, *et al.* (february 6 2008) Cancer is frequently considered to be a disease of the cell cycle; alterations in different families of cell cycle regulators cooperate in tumor development. Molecular analysis of human tumors has shown that cell cycle regulators are frequently mutated in human neoplasms, which underscores how important the maintenance of cell cycle commitment is in the prevention of human cancer. The regulatory pathways controlling cell cycle phases include several oncogenes and tumor suppressor genes which display a range of abnormalities with potential usefulness as markers of evolution or treatment response in epithelial ovarian cancer and Summarizing the current knowledge about these aberrations in malignant tumors of the ovary and to focus our attention on the genes involved in the

development of tumors arising from the ovarian epithelium, which are the most common types of ovarian malignancies.

Zohre Momenimovahed, et al..(April 30 2019) describes that Ovarian cancer is one of the most common gynecologic cancers that has the highest mortality rate. Considering the fact that knowledge on the incidence, mortality of ovarian cancer, as well as its risk factors is necessary for planning and preventing complications and examining the epidemiology and risk factors of ovarian cancer in the world, further he concluded with the significant evidence about ovarian cancer, Considering the heavy burden of ovarian cancer on women's health, preventive measures as well as health education and early detection in high risk groups of women are highly recommended. Although some risk factors cannot be changed, a focus on preventable risk factors may reduce the risk of ovarian cancer.

Agella toss, et al..(2015 May 17) describes that more than one-fifth of ovarian tumors have hereditary susceptibility and, in about 65–85% of these cases, the genetic abnormality is a germ line mutation in BRCA genes. Nevertheless, several other suppressor genes and oncogenes have been associated with hereditary ovarian cancers, including the mismatch repair (MMR) genes in Lynch syndrome, the tumor suppressor gene, TP53, in the Li-Fraumeni syndrome, and several other genes involved in the double-strand breaks repair system, such as CHEK2, RAD51, BRIP1, and PALB2.

Helen M. Berman et al..(2000) describes that Protein Data Bank (PDB) was established at Brookhaven National Laboratories (BNL) in 1971 as an archive for crystal structure of biological macromolecules

Sunghwan Kim et al..(2016) confirms that PubChem is committed to continue serving as a key chemical information resource not only to the biomedical research community but also to the scientific community as a whole. Report of September 2015 suggests that pubchem has more than 157 million depositor-provided data's from biologist and scientist on chemical substance descriptions. In addition 60 million unique chemical structures and its biological assay descriptions majorly covered about 10 thousand

unique protein target sequences are been deposited. These data's are updated regularly.

N. Siddharthan, et al.,(Sep 03 ,2017) describes Drug discovery is the step-by-step process by which new candidate drugs are discovered. Bioinformatics deals with the exponential growth and the development in primary and secondary databases like nucleic acid sequences, protein sequences and structures. Drugs are usually only developed when the particular drug target for those drugs' actions have been identified and studied. Drug target validation helps us to check the potential for failure in the clinical testing and approval phases. Drug discovery also involves several phases from target identification to preclinical development .

3. AIM AND SCOPE

3.1 AIM:

- To study the efficacy of phytochemicals of *Nigella sativa* against ovarian cancer causing genes through computational approach.

3.2 SCOPE:

- Retrieving the targets from PDB and retrieving the ligands from PubChem and doing the further pharmacodynamics and pharmacokinetic analysis reduces the cost and time of finding the compounds on any disease.
- In silico approach has wide application and makes work easier and even more effective in invitro and in vivo analysis.

4. MATERIALS AND METHODS

4.1 SELECTION OF THE TARGET:

The targets are selected based on documentary evidence and the target structure are retrieved from PDB (Protein Data Bank) which is a database for the three-dimensional structural data of large biological molecules. Total of 9 targets have been analyzed and studied for causing ovarian cancer.

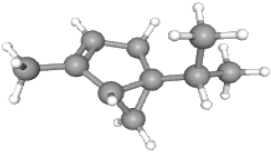
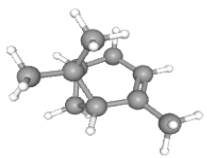
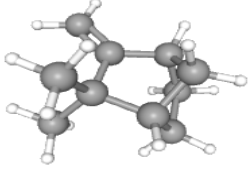
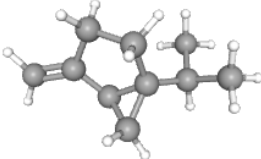
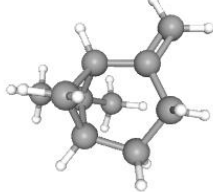
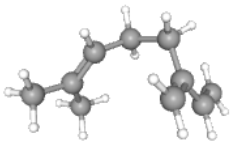
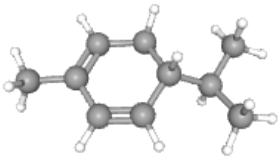
Table 4.1.1 Target genes are selected for causing ovarian cancer

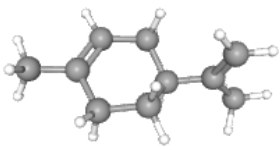
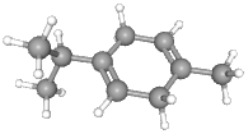
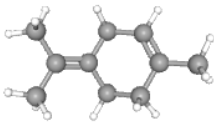
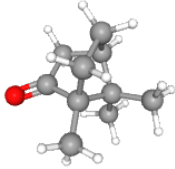
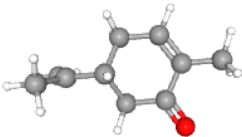
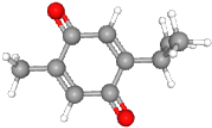
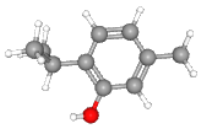
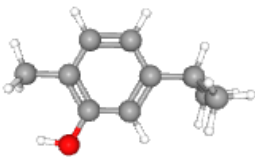
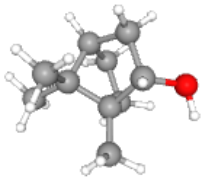
Target genes	PDB ID
BRCA1	3FA2
BRCA2	3EU7
TP53	ITUP
NBN	5WQD
RAD51	IB22
RAD50	5G0X
CHEK2	2WTI
PIK3CA	7K71
MMR	6MFQ

4.2 SELECTION OF LIGANDS:

Ligands are selected based on documentary evidence on the phytochemical compounds of nigella sativa (black cumin). The compounds are retrieved from PubChem, 3D conformer structure is obtained for interaction studies. PubChem provides information on chemical properties, synonyms, chemical structures, SMILES, InChI strings, bioactivity and links to structurally related compounds as well as other related compounds. It also includes other NCBI databases like PubMed.

TABLE №4.2.1 LIST OF LIGANDS

LIGANDS	PUBCHEM ID	STRUCTURE
Alpha thujene	6451618	
Alpha pinene	6654	
Camphene	6616	
Sabinene	18818	
Beta-pinene	14896	
Beta-mycrene	31253	
Alpha-phellandrene	7460	

Limonene	22311	
Gamma-Terpinene	7461	
Terpinolene	11463	
Camphor	2537	
Carvone	7439	
Thymoquinone	10281	
Thymol	6989	
Carvacrol	10364	
Borneol	64685	

4.3 ONLINE SMILES TRANSLATOR:

Online smiles translator is used to further carry out the pharmacodynamics studies of the ligands. This online smile translator is used for the conversion of the ligands since pharmacodynamics studies using pre-ADME tools need SDF Format. All ligands 3D structure was not available in PubChem so it can be obtained using online smiles translator.

4.4 PHARMACOKINETIC STUDY

Pharmacokinetic study was carried out to find the approximate prediction of the ligands using online tool called pre-ADMET which includes drug-likeness, ADME studies and Toxicity.

4.6. PHARMACODYNAMICS ANALYSIS

Pass online server was used to predict the lead compounds and their biological activities. The adverse effects are analysed using Pass online server .This study mainly focused on the activity related to ovarian cancer.

4.7. MOLECULAR DOCKING

The targets and ligands are docked to predict the binding affinity and their interaction by using the discovery studio 2020 (a latest and updated version has been used) from Madras Veterinary College, BTIS centre. This updated version was useful in exact interaction study on the selected targets and ligands.

Discovery studio visualizer is the latest version was used to obtain the interacting residue, number of hydrogen bonds interacted. Software was also used to measure the bond length on accordance with the interacted target and the ligand. These detailed study may be useful to find drug even more effectively with no or low failure chances in in vivo and invitro studies.

5. RESULTS AND DISSCUSION

5.1. DRUGLIKENESS

5.1.1 Drug likeness study of the ligands

LIGANDS	RULE OF FIVE
P-cymene	Suitable
Alpha thujene	Suitable
Alpha pinene	Suitable
Camphene	Suitable
Sabinene	Suitable
Beta-pinene	Suitable
Beta-mycrene	Suitable
Alpha-phellandrene	Suitable
Limonene	Suitable
Gamma-Terpinene	Suitable
Terpinolene	Suitable
Camphor	Suitable
Cavone	Suitable
Thymoquinone	Suitable
Thymol	Suitable
Carvacrol	Suitable
Borneol	Suitable

5.2. ADME STUDY:

TABLE 5.2.1. ADME STUDIES OF P-cymene, Alpha-thujene, Alpha –pinene, Camphene

ID	P-cymene	Alpha-thujene	Alpha -pinene	Camphene
BBB	4.96983	5.5333	5.5333	5.75623
Buffer_solubility_mg_L	1047.8	1368.72	1368.72	164.943
Caco2	23.4337	23.6324	23.6324	23.4938
CYP_2C19_inhibition	Inhibitor	Non	Non	Non
CYP_2C9_Inhibition	Inhibitor	Inhibitor	Inhibitor	Inhibitor
CYP_2D6_Inhibition	Non	Non	Non	Non
CYP_2D6_substrate	Non	Non	Non	Non
CYP_34A_inhibitor	Inhibitor	Inhibitor	Non	Inhibitor
CYP_34A_substrate	Weakly	Substrate	Substrate	Substrate
HIA	100.00000 0	100.00000 0	100.00000 0	100.00000 0
MDCK	237.507	304.815	304.815	300.517
Pgp_inhibition	Non	Inhibitor	Inhibitor	Inhibitor
Plasms_Protein_Binding	100.00000 0	100.00000 0	100.00000 0	100.00000 0
Pure_water_solubility_mg_L	80.701	133.362	133.362	113.777
Skin_Permeability	-0.805344	-1.44458	-1.44636	-1.33638
SKlogD_value	3.559720	2.918210	2.918210	2.952770
SKlogP_value	3.559720	2.918210	2.918210	2.952770
SKlogS_buffer	-2.107540	-1.997980	-1.997980	-2.916960
SKlogS_pure	-3.220940	-3.009260	-3.009260	-3.078240

5.2.2 Sabinene, Beta- pinene, Beta- myrcene and Alpha phellandrene

TABLE 5.2.2.1 ADME STUDIES on Sabinene, Beta-pinene, Beta- myrcene, Alpha – phellandrene

ID	Sabinene	Beta-pinene	Beta-Myrcene	Alpha-Phellandrene
BBB	5.75623	5.75623	9.1018	7.17054
Buffer_solubility_mg_L	164.943	164.943	3.13858	1227.08
Caco2	23.4924	23.4924	23.6306	23.4164
CYP_2C19_inhibition	Non	Non	Inhibitor	Inhibitor
CYP_2C9_Inhibition	Inhibitor	Inhibitor	Inhibitor	Inhibitor
CYP_2D6_Inhibition	Non	Non	Non	Non
CYP_2D6_substrate	Non	Non	Non	Non
CYP_34A_inhibitor	Inhibitor	Inhibitor	Non	Non
CYP_34A_substrate	Substrate	Substrate	Substrate	Weakly
HIA	100.000000	100.000000	100.000000	100.000000
MDCK	300.517	300.517	216.252	267.707
Pgp_inhibition	Inhibitor	Inhibitor	Inhibitor	Non
Plasms_Protein_Binding	60.971697	100.000000	100.000000	100.000000
Pure_water_solubility_mg_L	113.777	113.777	129.406	141.466
Skin_Permeability	-1.36792	-1.35832	-0.632987	-0.882293
SKlogD_value	2.952770	2.952770	4.084600	3.387220
SKlogP_value	2.952770	2.952770	4.084600	3.387220
SKlogS_buffer	-2.916960	-2.916960	-4.637560	-2.045420
SKlogS_pure	-3.078240	-3.078240	-3.022340	-2.983640

5.2.3. Limonene, Gamma-terpinene, Terpinolene, Camphor

TABLE 5.2.3.1 ADME STUDIES On Limonene, GammaTerpinene, Terpinolene, Camphor

ID	Limonene	Gamma-terpinene	Terpinolene	Camphor
BBB	8.27823	8.03745	8.90455	8.67174
Buffer_solubility_mg_L	14.068	116.738	11.1059	20539.8
Caco2	23.6317	23.6401	23.6372	26.8158
CYP_2C19_inhibition	Inhibitor	Inhibitor	Inhibitor	Non
CYP_2C9_Inhibition	Inhibitor	Inhibitor	Inhibitor	Inhibitor
CYP_2D6_Inhibition	Non	Non	Non	Non
CYP_2D6_substrate	Non	Non	Non	Non
CYP_3A4_inhibitor	Non	Non	Non	Inhibitor
CYP_3A4_substrate	Substrate	Substrate	Substrate	Substrate
HIA	100.000000	100.000000	100.000000	100.000000
MDCK	238.434	244.909	221.545	90.0816
Pgp_inhibition	Inhibitor	Non	Inhibitor	Inhibitor
Plasms_Protein_Binding	100.000000	100.000000	93.161811	100.000000
Pure_water_solubility_mg_L	100.696	118.03	98.4766	483.111
Skin_Permeability	-0.834346	-0.885719	-0.883947	-1.84121
SKlogD_value	3.669300	3.634740	3.882260	2.085710
SKlogP_value	3.669300	3.634740	3.882260	2.085710
SKlogS_buffer	-3.986060	-3.067080	-4.0888740	-0.869920
SKlogS_pure	-3.131280	-3.062300	-3.140960	-2.498470

5.2.4. Carvone, Thymoquinone, Thymol

TABLE 5.2.4.1ADME STUDIES on Carvone, Thymoquinone,Thymol.

ID	Carvone	Thymoquinone	Thymol
BBB	1.06069	1.78938	6.38802
Buffer_solubility_mg_L	75.1636	357.209	903.915
Caco2	47.7427	23.037	38.0122
CYP_2C19_inhibition	Inhibitor	Inhibitor	Inhibitor
CYP_2C9_Inhibition	Inhibitor	Inhibitor	Inhibitor
CYP_2D6_Inhibition	Non	Non	Non
CYP_2D6_substrate	Non	Non	Non
CYP_34A_inhibitor	Non	Non	Inhibitor
CYP_34A_substrate	Substrate	Substrate	Weakly
HIA	100.000000	99.286337	100.000000
MDCK	97.5458	61.0892	87.3307
Pgp_inhibition	Non	Non	Inhibitor
Plasms_Protein_Binding	58.045472	100.000000	100.000000
Pure_water_solubility_mg_L	636.47	61.7082	180.95
Skin_Permeability	-1.40341	-1.78115	-1.06531
SKlogD_value	2.599010	2.514900	3.405370
SKlogP_value	2.599010	2.514900	3.405370
SKlogS_buffer	-3.300720	-2.662460	-2.220600
SKlogS_pure	-2.372950	-3.425040	-2.919170

5.2.5. Carvacrol, Borneol

TABLE 5.2.5.1ADME Studies on carvacrol and Borneol

ID	Carvacrol	Borneol
BBB	6.38799	3.80575
Buffer_solubility_mg_L	903.915	9139.02
Caco2	38.0121	24.2343
CYP_2C19_inhibition	Inhibition	Non
CYP_2C9_Inhibition	Inhibition	Inhibitor
CYP_2D6_Inhibition	Non	Non
CYP_2D6_substrate	Non	Non
CYP_34A_inhibitor	Inhibitor	Inhibitor
CYP_34A_substrate	Weakly	Substrate
HIA	100.000000	100.000000
MDCK	87.3307	91.738
Pgp_inhibition	Inhibitor	Non
Plasms_Protein_Binding	100.000000	100.000000
Pure_water_solubility_mg_L	180.95	574.037
Skin_Permeability	-1.04552	-1.88173
SKlogD_value	3.405370	2.076990
SKlogP_value	3.405370	2.076990
SKlogS_buffer	-2.220600	-1.227330
SKlogS_pure	-2.919170	-2.429290

5.3 TOXICITY STUDY:

5.3.1 P-cymene, Alpha-thujene, Alpha- pinene, Camphene

TABLE 5.3.1.1 P-Cymene, Alpha- thujene, Alpha- pinene, Camphene

ID	P-Cymene	Alpha-thujene	Alpha-pinene	Camphene
Carcino_Mouse	Positive	Negative	Negative	Negative
Carcino_Rat	Negative	Positive	Positive	Positive

5.3.2 Sabinene, Beta-pinene, Beta- Mycrene and Alpha phellandrene

TABLE 5.3.2.1Sabinene, Beta-pinene, Beta- mycrene, Alpha- phellandrene

ID	Sabinene	Beta-pinene	Beta-mycrene	Alpha-phellandrene
Carcino_Mouse	Negative	Negative	Negative	Positive
Carcino_Rat	Positive	Positive	Positive	Positive

5.3.3. Limonene, Gamma-Terpinene, Terpinolene, Camphor

TABLE 5.3.3.1 Limonene, Gamma-Terpinene, Terpinolene, Camphor

ID	Limonene	Gamma-Terpinene	Terpinolene	Camphor
Carcino_Mouse	Negative	Positive	Positive	Negative
Carcino_Rat	Positive	Positive	Positive	Positive

5.3.4 Carvone, Thymoquinone, Thymol,Carvacrol, Borneol

TABLE 5.3.4.1Carvone, Thymoquinone, Thymol, Carvacrol, Borneol

ID	Carvone	Thymoquinone	Thymol	Carvacrol	Borneol
Carcino_Mouse	Negative	Positive	Negative	Negative	Negative
Carcino_Rat	Positive	Positive	Negative	Negative	Positive

5.4 PHARMACODYNAMIC RESULTS

The pharmacodynamics studies were done and the smiles of the ligands were retrieved using PubChem and using pass online server the possible activity were retrieved.

5.4.1 p-cymene

The smiles notation for p- cymene (CC1=CC=C(C=C1)C(C)C)

pa	pi	Activity
0,919	0,004	Mucomembranous protector
0,888	0,008	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,884	0,006	Antieczematic
0,644	0,024	Anti-inflammatory
0,623	0,011	Anti-infective
0,607	0,005	Muscular dystrophy treatment
0,606	0,018	Cholesterol antagonist

5.4.2 Alpha-thujene

The smiles notation for alpha thujene (CC1=CCC2(C1C2)C(C)C)

Pa	pi	Activity
0,866	0,008	Antieczematic
0,850	0,014	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,807	0,006	Anti-inflammatory
0,732	0,005	Adenomatous polyposis treatment
0,643	0,036	Antineoplastic
0,634	0,087	Mucomembranous protector

5.4.3 Alpha pinene

The smiles notation for Alpha pinene CC1=CCC2CC1C2(C)C

Pa	pi	Activity
0,863	0,012	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,760	0,049	Phobic disorders treatment
0,654	0,004	Transplant rejection treatment
0,680	0,066	Mucomembranous protector
0,565	0,103	Antieczematic

5.4.4 Camphene

The smiles notation for Camphene CC1(C2CCC(C2)C1=C)C

Pa	pi	Activity
0,882	0,006	Antieczematic
0,873	0,010	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,782	0,040	Phobic disorders treatment
0,511	0,039	Apoptosis agonist

5.4.5 Sabinene

The smiles notation for Sabinene CC(C)C12CCC(=C)C1C2

Pa	pi	Activity
0,947	0,003	Antieczematic
0,891	0,005	Antineoplastic
0,853	0,005	Anti-inflammatory
0,840	0,016	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,800	0,004	Ant psoriatic
0,641	0,084	Mucomembranous protector

5.4.6 beta-pinene

The smiles notation for beta-pinene (CC1(C2CCC(=C)C1C2)C)

Pa	pi	Activity
0,902	0,005	Antieczematic
0,857	0,013	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,752	0,053	Phobic disorders treatment
0,657	0,034	Antineoplastic
0,611	0,029	Anti-inflammatory
0,596	0,022	Adenomatous polyposis treatment
0,539	0,012	Ant metastatic

5.4.7 beta mycrene

The smiles notation for beta mycrene (CC(=CCCC(=C)C=C)C)

Pa	pi	Activity
0,941	0,004	Mucomembranous protector
0,896	0,005	Antineoplastic
0,892	0,004	Antineoplastic (breast cancer)
0,836	0,012	Antieczematic
0,744	0,011	Apoptosis agonist
0,763	0,035	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,584	0,020	Antifungal
0,590	0,030	Immunosuppressant
0,555	0,004	Antineoplastic (carcinoma)

5.4.8 Alpha- phellandrene

The smiles notation for Alpha- phellandrene (CC1=CCC(C=C1)C(C)C)

Pa	pi	Activity
0,828	0,019	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,772	0,025	Antieczematic
0,727	0,005	Adenomatous polyposis treatment
0,750	0,054	Phobic disorders treatment
0,751	0,034	Mucomembranous protector

5.4.9 Limonene

The smiles notation for Limonene (CC1=CCC(CC1)C(=C)C)

Pa	pi	Activity
0,896	0,005	Antieczematic
0,816	0,007	Apoptosis agonist
0,812	0,010	Antineoplastic
0,753	0,038	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,714	0,015	Immunosuppressant

5.4.10 Gamma-terpinene

The smiles notation for Gamma-terpinene (CC1=CCC(=CC1)C(C)C)

Pa	pi	Activity
0,860	0,013	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,854	0,009	Antieczematic
0,803	0,033	Phobic disorders treatment
0,789	0,021	Mucomembranous protector
0,714	0,006	Adenomatous polyposis treatment

5.4.11 Terpinolene

The smiles notation for Terpinolene (CC1=CCC(=C(C)C)CC1)

Pa	pi	Activity
0,890	0,005	Antieczematic
0,850	0,014	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,790	0,037	Phobic disorders treatment
0,670	0,010	Adenomatous polyposis treatment
0,667	0,018	Apoptosis agonist
0,579	0,007	Anti metastatic
0,562	0,114	Mucomembranous protector

5.4.12 Camphor

The smiles notation for Camphor (CC1(C2CCC1(C(=O)C2)C)C)

Pa	pi	Activity
0,936	0,004	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,746	0,031	Antieczematic
0,751	0,053	Phobic disorders treatment
0,729	0,044	Mucomembranous protector
0,671	0,010	Adenomatous polyposis treatment

5.4.13 Carvone

The smiles notation for Carvone (CC1=CCC(CC1=O)C(=C)C)

Pa	Pi	Activity
0,772	0,015	Antineoplastic
0,782	0,030	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,737	0,034	Antieczematic
0,694	0,015	Apoptosis agonist
0,657	0,022	Immunosuppressant
0,567	0,005	Antineoplastic (ovarian cancer)
0,485	0,021	Ant carcinogenic

5.4.14 Thymoquinone

The smiles notation for Thymoquinone (CC1=CC(=O)C(=CC1=O)C(C)C)

Pa	Pi	Activity
0,881	0,005	Mucomembranous protector
0,873	0,010	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,802	0,004	Antiseptic
0,762	0,027	Antieczematic
0,740	0,058	Phobic disorders treatment
0,650	0,013	Adenomatous polyposis treatment
0,609	0,025	Apoptosis agonist
0,601	0,031	Antiinflammatory

5.4.15 Thymol

The smiles notation for Thymol (CC1=CC(=C(C=C1)C(C)C)O)

Pa	Pi	Activity
0,922	0,004	Mucomembranous protector
0,913	0,003	Antiseptic
0,866	0,011	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,788	0,021	Antieczematic
0,698	0,008	Adenomatous polyposis treatment

5.4.16 Carvacrol

The smiles notation for Carvacrol (CC1=C(C=C(C=C1)C(C)C)O)

Pa	Pi	Activity
0,880	0,005	Mucomembranous protector
0,866	0,011	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,824	0,004	Antiseptic
0,811	0,016	Antieczematic
0,784	0,005	Anti-infective
0,674	0,019	Anti-inflammatory

5.4.17 Borneol

The smiles notation for Borneol (CC1(C2CCC1(C(C2)O)C)C)

Pa	Pi	Activity
0,962	0,002	Testosterone 17beta-dehydrogenase (NADP+) inhibitor
0,756	0,033	Mucomembranous protector
0,566	0,008	Antimetastatic
0,538	0,046	Anti-inflammatory

5.5 DOCKING RESULTS

5.5.1 Interaction of BRCA1-3FA2 with carvacol

The 3 dimensional structure of BRCA1 binding with carvacol

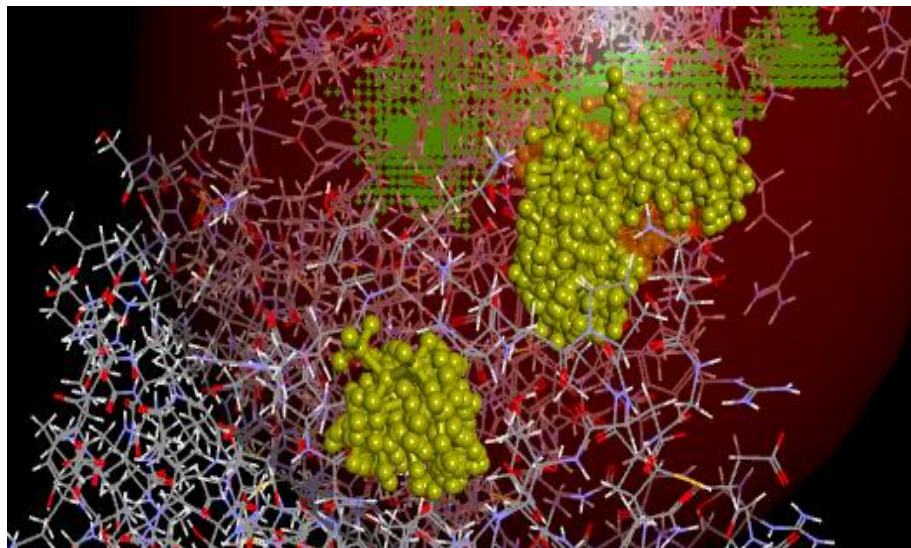


Fig 5.5.1.1 Interaction of BRCA1-3FA2 with carvacol

The 2 dimensional structure of BRCA1 binding with carvacol

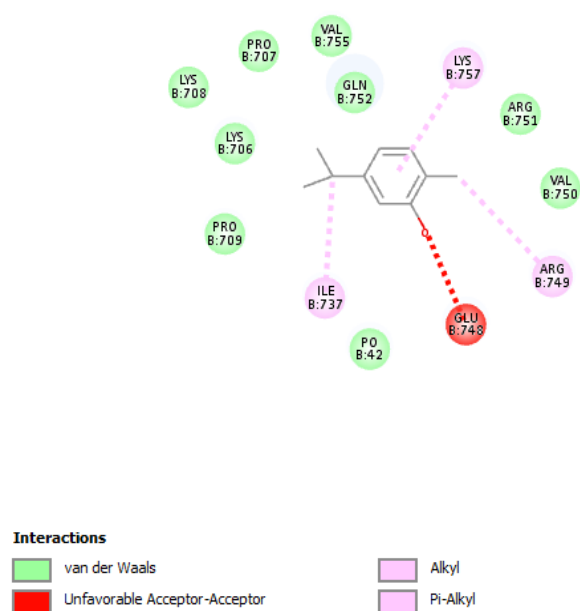


Fig 5.5.1.2 2D structure of BRCA1 binding with carvacol

5.5.2 Interaction of TP53 -1TUP with carvacrol

The 3 dimensional structure of TP53 binding with carvacrol

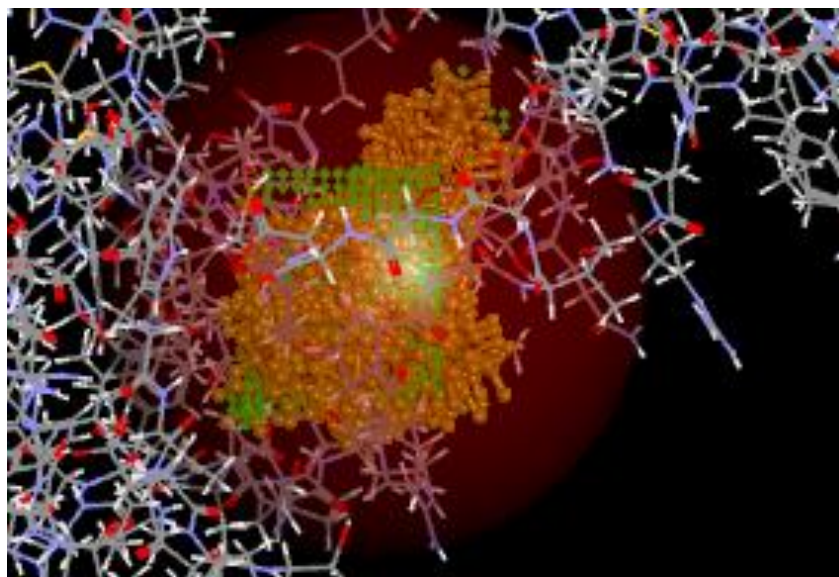


Fig 5.5.2.1 Interaction of TP53 -1TUP with carvacrol

The 2 dimensional structure of TP53 binding with carvacrol

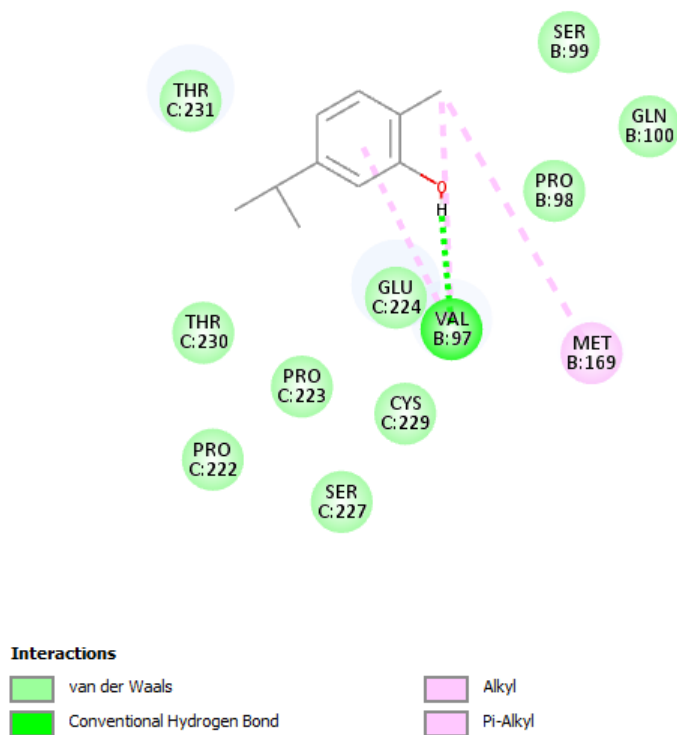


Fig 5.5.2.2 2D structure of TP53 binding with carvacrol

5.5.3 Interaction of MMR- 6MFQ with thymoquinone

The 3 dimensional structure of MMR binding with thymoquinone

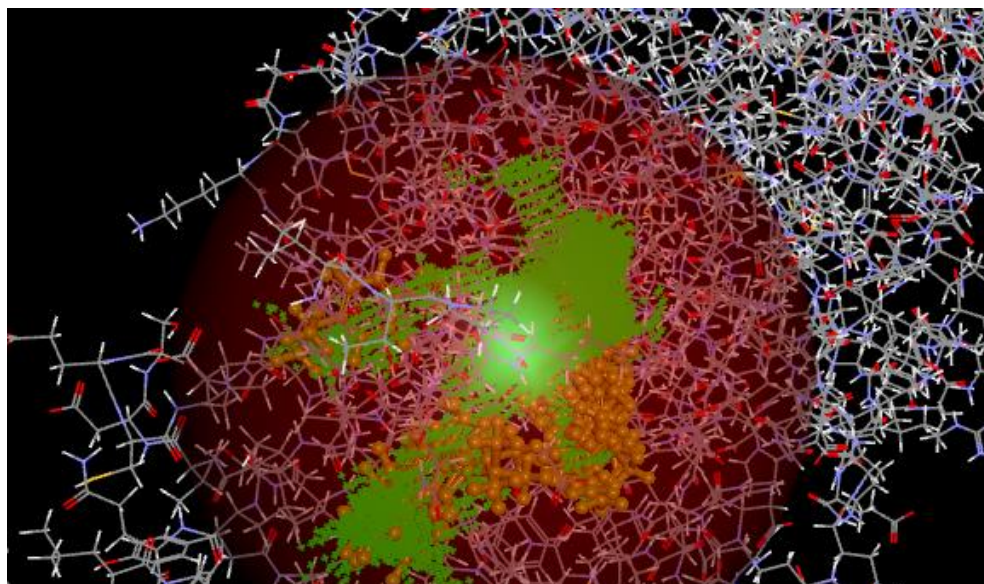


Fig 5.5.3.1 Interaction of MMR- 6MFQ with thymoquinone

The 2 dimensional structure of MMR binding with thymoquinone

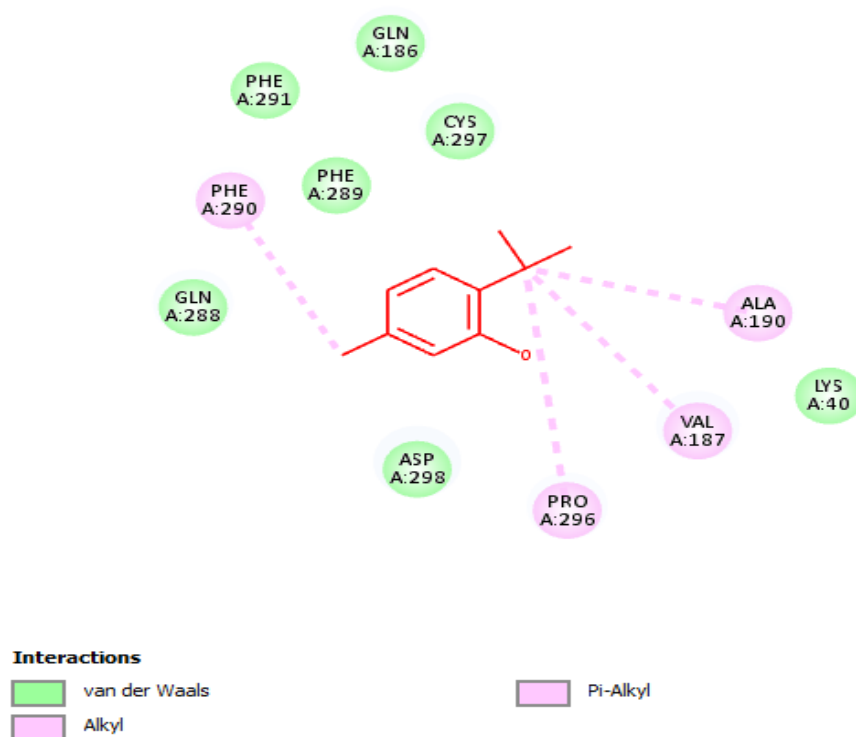


Fig 5.5.3.2 2D structure of MMR binding with thymoquinone

5.5.4 Interaction of CHEK2 -2WTI with thymoquinone

The 3 dimensional structure of CHEK2 binding with thymoquinone

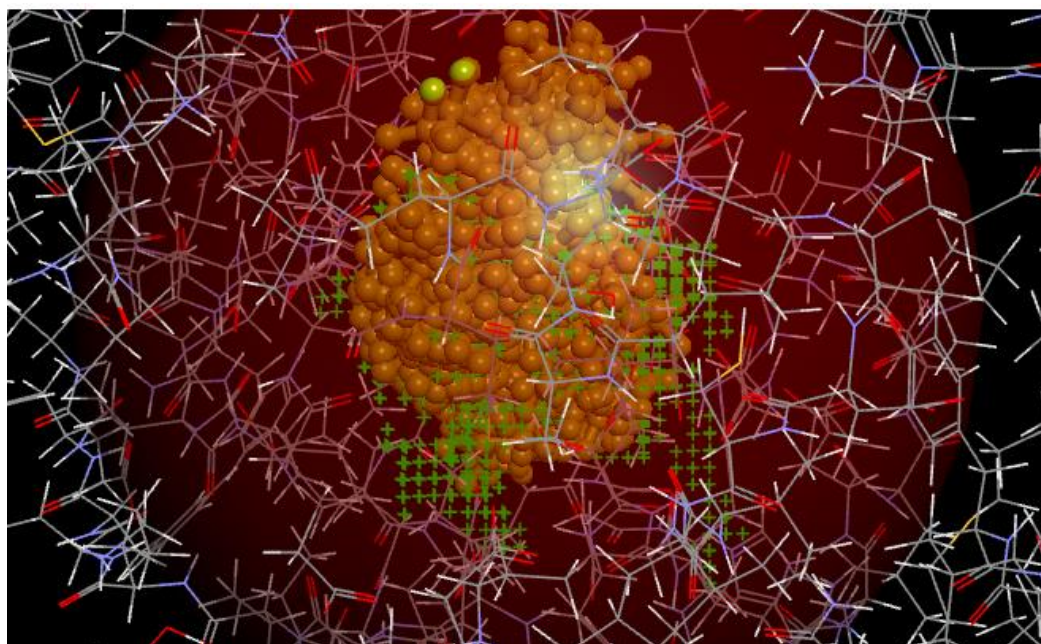


Fig 5.5.4.1 Interaction of CHEK2-2WTI with thymoquinone

The 2 dimensional structure of CHEK 2 binding with thymoquinone

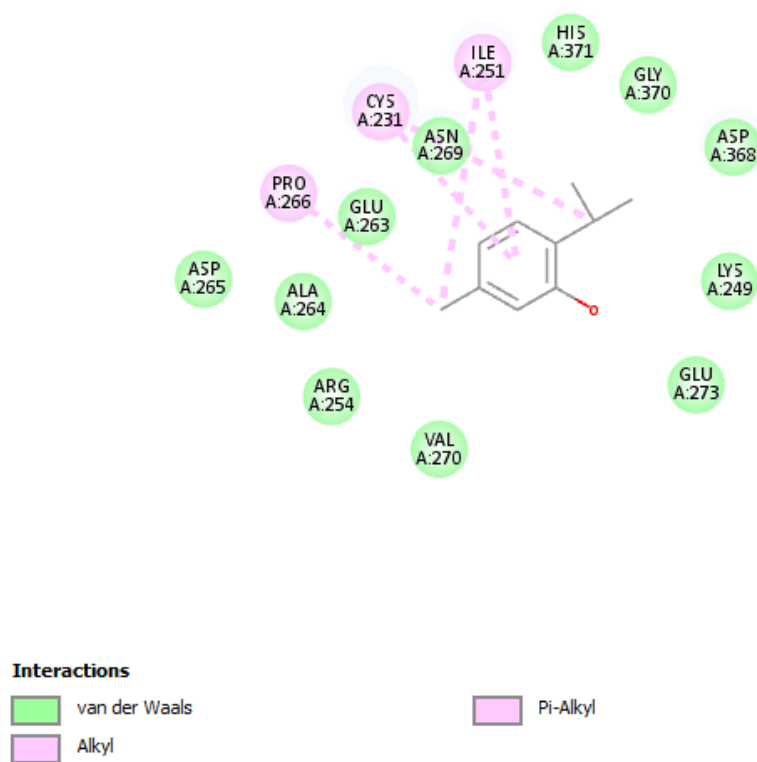


Fig 5.5.4.2 2D structure of CHEK2 binding with thymoquinone

5.5.5 Interaction of BRCA2- 3EU7 with CARVONE

The 3 dimensional structure of BRCA2 binding with carvone

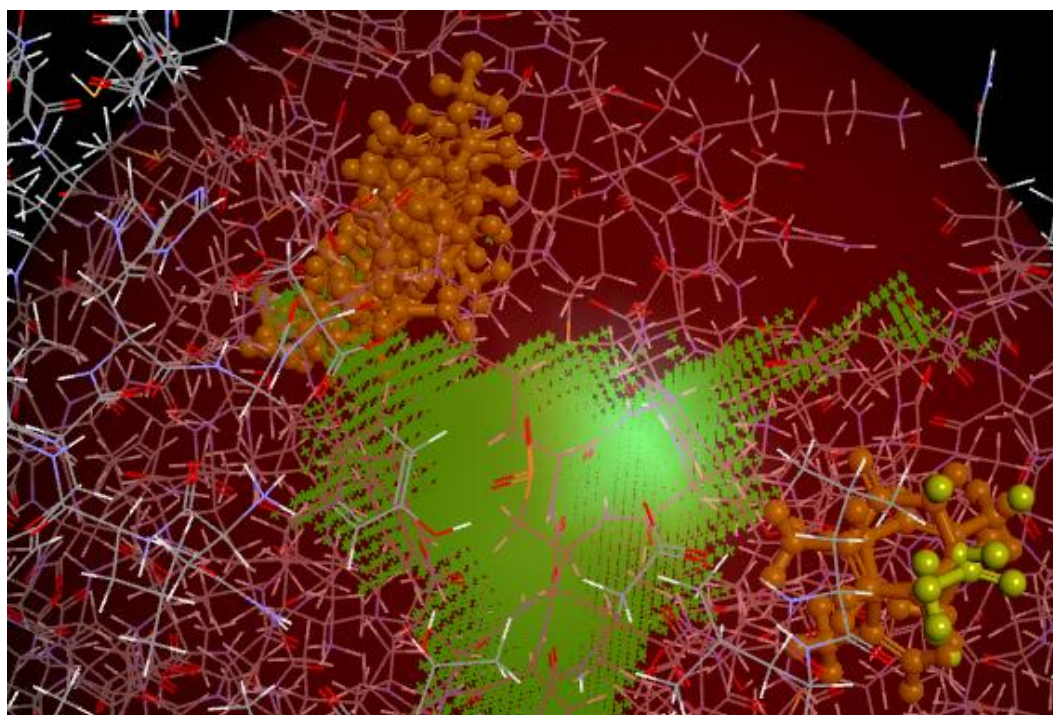


Fig 5.5.5.1 Interaction of BRCA2- 3EU7 with CARVONE

The 2 dimensional structure of BRCA2 binding with carvone

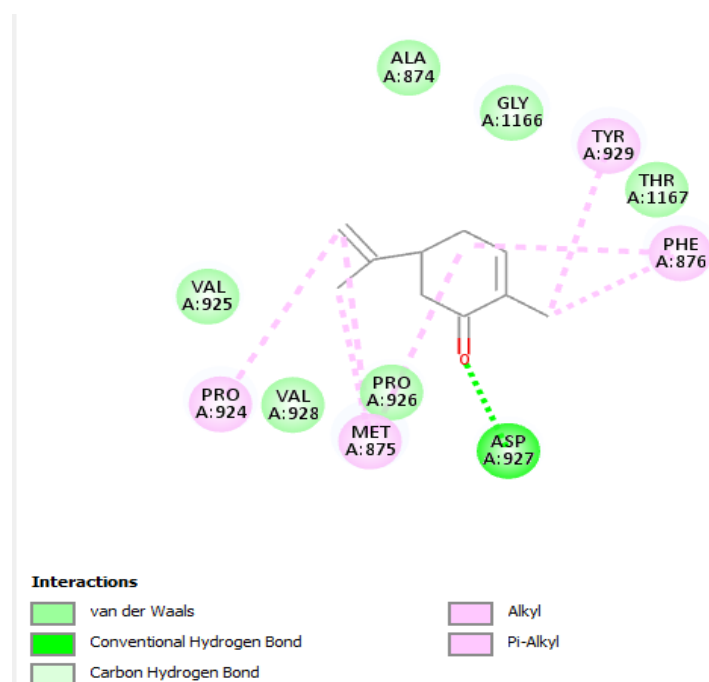


Fig 5.5.5.2 2D structure of BRCA2 binding with carvone

5.5.6 Interaction of RAD50- 5GOX with THYMOL

The 3 dimensional structure of RAD50 binding with thymol

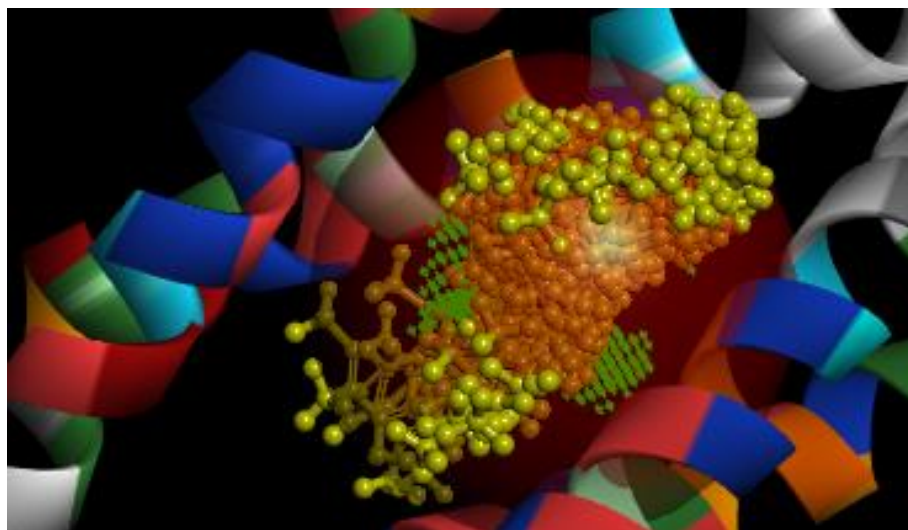


Fig 5.5.6.1 Interaction of RAD50- 5GOX with THYMOL

The 2 dimensional structure of RAD50 binding with thymol

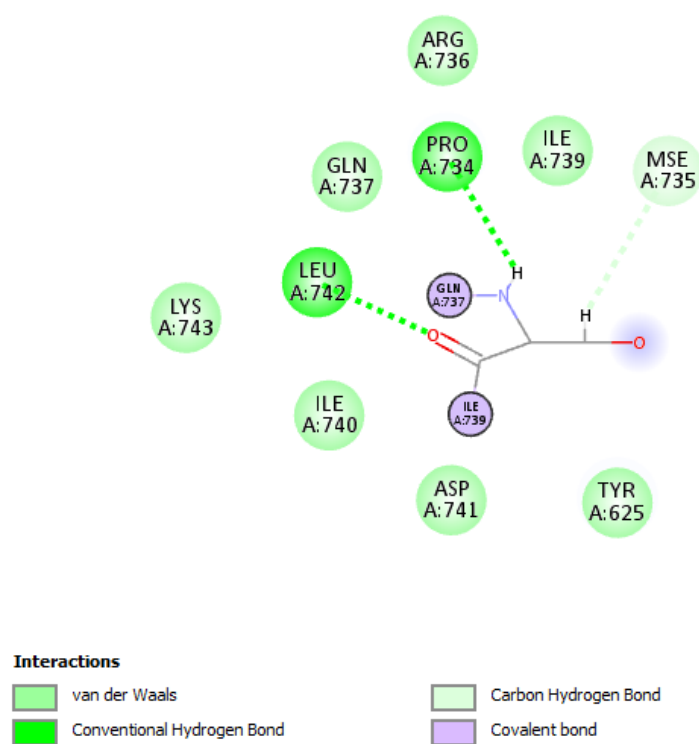


Fig 5.5.6.2 2D structure of RAD50 binding with thymol

5.6.7. BEST DOCKING RESULTS

TABLE 5.6.7.1 BEST DOCKING RESULTS

S.NO	TARGET	LIGAND	LIP DOCK SCORE	HYDROGEN BONDING	DISTANCE
1.	BRCA1	CARVACROL	76.64	764	1.965 A
2.	TP53	CARVACROL	66.25	1201	1.695 A
3.	MMR	THYMOQUINONE	74.65	1081	1.925 A
4.	CKEK2	THYMOQUINONE	67.54	599	1.982 A
5.	BRCA2	CARVONE	78.78	569	2.705 A
6.	RAD50	THYMOL	64.569	748	2.798 A

Molecular docking is carried out for all the possible targets with the lead compound. The highest dock score is obtained for each target and they discussed in detail with the interaction of the lead compound on that target, bond length and interacting residues.

6. SUMMARY AND CONCLUSION

The study of ovarian cancer against *Nigella sativa* is carried through in silico approach which has shown effective results. Based on the literature survey the genes and the phytochemicals of *Nigella sativa* were obtained. All possible and comparative prediction tools and study was carried out to know better about the ligands. PreADMET tool was used for further details of the lead compound such as ADME, drug likeness and Toxicity. Pharmacodynamics study was carried out by using pass online server which showed the effects of biological activity of the lead compound. Molecular docking of the target with the ligand showed, libdock score, and relative energy, bond length and 2D prediction.

CONCLUSION

Overall, study revealed that the binding activity of the lead compound and the drug compound has shown significant results whereas the lead compound can act as drug molecule for the given targets which has good activity

Among all the targets BRCA1 and BRCA2 with these phytochemicals therefore Carvone Thymoquinone and carvacol has shown good results. From the above results we can conclude that the plant *nigella sativa* can cure the ovarian cancer. Also invitro and in vivo studies should be carried out the for effective therapeutic drug. Computational approach has been evident that it can act as better drug

7.REFERENCE

1. Ahmad A, Husain A, Mujeeb M, Khan SA, Najmi AK, Siddique NA, et al. A review on therapeutic potential of *Nigella sativa*: a miracle herb. *Asian Pac J Trop Biomed*. 2013;337–52.
2. Alireza Tavakkoli, Vahid Mahdian, Bibi Marjan Razavi and Hossein Hosseinzadeh; Review on Clinical Trials of Black Seed (*Nigella sativa*) and Its Active Constituent, Thymoquinone; 2017 Sep 30: 179-193.
3. Angela Toss, Chiara Tomasello, Elisabetta Razzaboni, Giannina Contu, Giovanni Grandi, Angelo Cagnacci, Russell J. Schilder and Laura Cortesi ; Hereditary Ovarian Cancer: Not Only BRCA 1 and 2 Genes 2015 May 17.
4. Badgwell D, Bast JRC. Early detection of ovarian cancer. *Dis Markers*. 2007;397–410.
5. Bali A, O'Brien PM, Edwards LS, et al. Cyclin D1, p53, and p21Waf1/Cip1 expression is predictive of poor clinical outcome in serous epithelial ovarian cancer. 2004;10:5168–77.
6. Banerjee S, Padhye S, Azmi A, Wang Z, Philip PA, Kucuk O, et al. Review on molecular and therapeutic potential of thymoquinone in cancer. *Nutr Cancer*. 2010;938–46.
7. BL. Understanding oral cancer in the genome era. *Head & neck*. 2010;32(9):1246–68.
8. Brose M. S., Rebbeck T. R., Calzone K. A., Stopfer J. E., Nathanson K. L., Weber B. L. Cancer risk estimates for BCRA1 mutation carriers identified in a risk evaluation program. *Journal of the National Cancer Institute*. 2002;1365–1372.

9. Butt MS, Sultan MT. *Nigella sativa*: reduces the risk of various maladies. *Crit Rev Food Sci Nutr*. 2010;654–65.
10. Casadei S., Norquist B. M., Walsh T., et al. Contribution of inherited mutations in the BRCA2-interacting protein PALB2 to familial breast cancer. *Cancer Research*. 2011;2222–2229.
11. Darakhshan S, Bidmeshki Pour A, Hosseinzadeh Colagar A, Sisakhtnezhad S. Thymoquinone and its therapeutic potentials. *Pharmacol Res*. 2015;95–96:138–58.
12. Donna S. Shewach and Robert D. Kuchta Michigan; Introduction to Cancer Chemotherapeutics University of Michigan School of Medicine; 2009 Jul; 109(7): 2859–2861.
13. Easton D. Cancer risks in BRCA2 mutation carriers: the breast cancer linkage consortium. *Journal of the National Cancer Institute*. 1999;1310–1316.
14. Ferlay J., Shin H.-R., Bray F., Forman D., Mathers C., Parkin D. M. Estimates of worldwide burden of cancer in 2008: GLOBOCAN 2008. *International Journal of Cancer*. 2010;2893–2917.
15. Ford D., Easton D. F., Bishop D. T., Narod S. A., Goldgar D. E. Risks of cancer in BRCA1-mutation carriers. *The Lancet*. 1994:692–695.
16. Giuseppina D’Andrilli, Antonio Giordano, and Alessandro Bovicelli, Epithelial Ovarian Cancer: The Role of Cell Cycle Genes in the Different Histotypes ;2008 Feb 6; 7: 7–12.
17. Haseena S, Manjunath Aithal, Kusal K Das, Shaik Hussain Saheb; Phytochemical Analysis of *Nigella sativa* and its Effect on Reproductive System, 2015;514-517.

18. Helen M. Berman, John Westbrook, Zukang Feng, Gary Gilliland, T. N. Bhat, Helge Weissig, Ilya N. Shindyalov, Philip E. Bourne; The Protein Data Bank: 1 January 2000, Pages 235–242.
19. Ingolf Cascorbi, Prof. Dr. med. Dr. rer. Nat; Drug Interactions—Principles, Examples and Clinical Consequences; 2012 Aug 20.
20. J. P. van den Berg, H. E. M. Vereecke, J. H. Proost D. J. Eleveld, J. K. G. Wietasch, A. R. Absalom and M. M. R. F. Struys; Pharmacokinetic and pharmacodynamic interactions in anaesthesia. A review of current knowledge and how it can be used to optimize anaesthetic drug administration (2017).
21. Khan MA, Chen HC, Tania M, Zhang DZ. Anticancer activities of *Nigella sativa* (black cumin) Afr J Tradit Complement Altern Med. 2011;226–32.
22. Leita MM, Soslow RA, Baergen RN, et al. Mutation and expression of the TP53 gene in early stage epithelial ovarian carcinoma. Gynecol Oncol. 2004;93:301–6.
23. M. Iskar, G. Zeller, Zhao XM, V. Van Noort, P. Bork, “Drug discovery in the age of systems biology: the rise of computational approaches for data integration”, Pp.609–616, 2012.
24. Mariam, A. and A. Al-Basal, In vitro and In vivo AntiMicrobial Effects of *Nigella sativa* Linn. Seed Extracts against Clinical Isolates from Skin Wound Infections. Amer. J; 1440–1447;2009.
25. Mazumder A, Palma AJF, Wang Y. Validation and integration of gene-expression signatures in cancer. 2008.
26. Milner BJ, Allan LA, Eccles DM, et al. p53 mutation is a common genetic event in ovarian carcinoma. Cancer Res. 1993;53:2128–32
27. Mollazadeh H, Hosseinzadeh H. The protective effect of *Nigella sativa* against liver injury: a review. Iran J Basic Med Sci. 2014;958–66.

28. Pei-Wei Zhang, Lei Chen, Tao Huang, Ning Zhang, Xiang-Yin Kong, Yu-Dong Cai; Classifying Ten Types of Major Cancers Based on Reverse Phase Protein Array Profiles March 30, 2015. Viet CT, Schmidt
29. S. Hemadri Reddy, Adhari Said Al-Kalbani and Afrah Said Al-Rawahi; Studies on phytochemical screening GC-MS characterization, antimicrobial and antioxidant assay of black cumin seeds (*Nigella sativa*) 2018.
30. S.S. Ortega, L.C. Cara, M.K. Salvador, "In silico pharmacology for a multidisciplinary drug discovery process; 199–207, 2012. C.M. Song, S.J. Lim, J.C. Tong, "Recent advances in computer aided drug design", 579–591, 2009.
31. Shafi, G., A. Munshi, T.N. Hasan, A.A. Alshatwi, A. Jyothy and K.Y. David, Anti cancer activity of *Nigella sativa*; 2009.
32. Shafiq H, Ahmad A, Masud T, Kaleem M. Cardio-protective and anti-cancer therapeutic potential of *Nigella sativa*. Iran J Basic Med Sci. 2014;967–79.
33. Siegel R., Ma J., Zou Z., Jemal A. Cancer statistics, 2014. CA—Cancer Journal for Clinicians. 2014;9-29.
34. Sunghwan Kim, Paul A Thiessen, Evan E Bolton, Jie Chen, Gang Fu, Asta Gindulyte, Lianyi Han, Jane He, et al., PubChem Substance and Compound databases; 2016 Jan 4.
35. Toss A., Laura C. Molecular mechanisms of PARP inhibitors In BRCA-related ovarian cancer. Journal of Cancer Science and Therapy. 2013;409–416.
36. Walsh T., Casadei S., Coats K. H., et al. Spectrum of mutations in BRCA1, BRCA2, CHEK2, and TP53 in families at high risk of breast cancer. Journal of the American Medical Association. 2006;1379–1388.
37. Walsh T., Casadei S., Lee M. K., et al. Mutations in 12 genes for inherited ovarian, fallopian tube, and peritoneal carcinoma identified by massively parallel sequencing. Proceedings of the National Academy of Sciences of the United States of America. 2011;18032–18037.

38. Wooster R., Weber B. L. Breast and ovarian cancer. The New England Journal of Medicine. 2003.
39. Ziaee T, Moharreri N, Hosseinzadeh H. Review of pharmacological and toxicological effects of *Nigella sativa* and its active constituents. J Med Plants. 2012;16–42.
40. Zohre Momenimovahed, Azita Tiznobaik, Safoura Taheri, and Hamid Salehiniya; Ovarian cancer in the world: epidemiology and risk factors: 2019 Apr 30. 287–299.