Journal of Advanced Biomedical Sciences

https://jabs.fums.ac.ir/
Online ISSN: 2783-1523

Journal of Advanced Biomedical Sciences

Fasa University of Medical Sciences

The Role of TP53 and Associated Pathways in Pancreatic Ductal Adenocarcinoma Progression

Mahdi Mokhtari Tabar[1], **Mohammad Ghorbani**[2], **Abdolmajid Ghasemian**[3]

1. Department of Biochemistry, Faculty of Medicine, Fasa University of Medical Sciences, Fasa, Iran
2. Department of Hematology, School of Medicine, Fasa University of Medical Sciences, Fasa, Iran
3. Noncommunicable Diseases Research Center, Fasa University of Medical Sciences, Fasa, Iran

Article Info

Article Type:
Review Article

Article history:
Received
05 Aug 2024
Received in revised form
25 Aug 2024
Accepted
29 Oct 2024
Published online
20 Jan 2025

Publisher
Fasa University of Medical Sciences

Abstract

The pancreas, an organ integral to both exocrine and endocrine functions, plays a pivotal role in the digestive system and blood glucose regulation. Among the various malignancies affecting this organ, pancreatic cancer—particularly pancreatic ductal adenocarcinoma (PDAC)—stands out as one of the deadliest forms, claiming countless lives annually. The high mortality rate is primarily attributed to the absence of reliable early detection methods, rendering PDAC the third leading cause of cancer-related deaths. Genetic aberrations, such as mutations in *KRAS*, *CDKN2A*, *TP53*, and *SMAD4*, are observed in up to 90% of PDAC cases. These findings underscore the urgent need for advanced research into genetic diagnostics and treatment strategies. This review not only explores the biological significance of the pancreas but also investigates the genetic underpinnings of PDAC. Furthermore, contemporary therapeutic modalities, with a particular emphasis on gene therapy and targeted treatment approaches, are comprehensively analyzed.

Keywords: Pancreatic Ductal Adenocarcinoma, Suppressor Proteins, Gene therapy, Targeted Therapy

Cite this article: Mokhtari Tabar M, Ghorbani M, Ghasemian A. The Role of TP53 and Associated Pathways in Pancreatic Ductal Adenocarcinoma Progression. J Adv Biomed Sci. 2025; 15(1): 1-18.
DOI: 10.18502/jabs.v15i1.17548

Introduction

The pancreas is an organ in the human body situated in the upper abdomen, behind the stomach, and in front of the spine. Positioned on the left side of the body, the pancreas extends from the duodenum, the first part of the small intestine, to the spleen. It comprises three major parts: the head, body, and tail. The pancreatic head lies on the right side of the abdomen, nestled within the curve of the duodenum, while the pancreatic tail reaches toward the left side of the body, near the spleen (1–3).

The pancreas develops from two buds, the dorsal and ventral buds, which form during embryonic development from opposite sides of the distal foregut endoderm. The dorsal bud gives rise to the body and tail of the pancreas, whereas the ventral bud develops into the head of the pancreas. During embryonic development, these two buds fuse to form the mature pancreas (4).

Corresponding Author: Abdolmajid Ghasemian, Noncommunicable Diseases Research Center, Fasa University of Medical Sciences, Fasa, Iran.
Email: majidghasemian86@gmail.com

[DOI: 10.18502/jabs.v15i1.17548]

Within the body, the pancreas performs both exocrine and endocrine functions. Its exocrine functions include producing and secreting enzymes and bicarbonate ions into the duodenum to facilitate food digestion. The majority of the pancreatic glandular mass consists of acinar cells, which secrete digestive enzymes into ducts that empty into the duodenum. Additionally, the pancreatic duct cells produce bicarbonate ions to neutralize the stomach's acidic contents (5).

The endocrine functions of the pancreas involve the secretion of hormones directly into the circulatory system to regulate various physiological processes throughout the body. These hormones are produced by endocrine cells that form the islets of Langerhans—small, island-like structures embedded within the exocrine pancreatic tissue, comprising only 1–2% of the organ. The pancreatic endocrine system works to maintain blood glucose concentrations within a specific range, typically between 4 and 6 mM, thereby regulating carbohydrate metabolism and energy balance. This is primarily achieved through the secretion of hormones, such as glucagon and insulin (6).

Numerous common disorders, including diabetes and cancer, can arise from pancreatic dysfunction (7, 8). According to recent studies, a potential relationship exists between diabetes and pancreatic cancer. Specifically, concurrent diabetes or hyperglycemia—often presenting as a recently diagnosed condition—occurs in up to 80% of individuals with pancreatic cancer. The observation that diabetes and hyperglycemia frequently improve or even resolve after the surgical removal of pancreatic cancer provides further support for this link. These findings have led researchers to hypothesize that screening for pancreatic cancer may be a viable strategy for individuals newly diagnosed with diabetes. However, it is important to emphasize that the precise mechanisms underlying the association between diabetes and pancreatic cancer remain unclear and warrant further investigation (Figure 1) (9).

Overview of Pancreatic Cancer

Pancreatic cancer is a malignant tumor associated with an exceptionally poor prognosis,

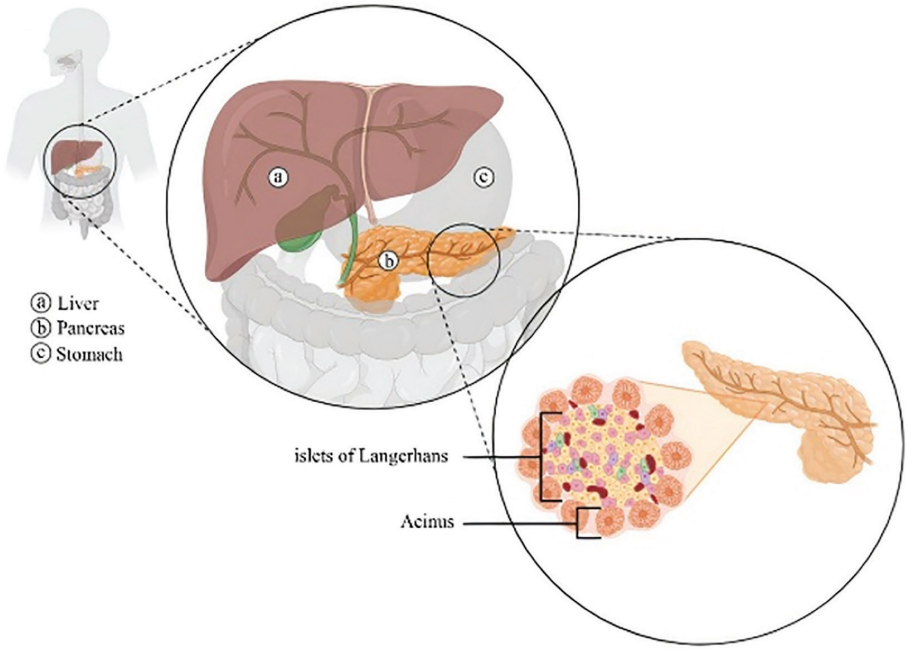

Figure 1. The location of pancreas behind the stomach in the abdominal cavity. It performs both exocrine and endocrine tasks. The pancreas contains clumps of cells called Langerhans islets, which secrete hormones. They perform an endocrine role by directly secreting hormones into the bloodstream.

[DOI: 10.18502/jabs.v15i1.17548]

as evidenced by patients' low five-year survival rates, which remain at approximately 6% in the United States. Despite advancements in surgical techniques, chemotherapy regimens, and the application of neoadjuvant chemoradiotherapy or chemoimmunotherapy, pancreatic cancer still accounts for 3% of all malignancies and 7% of all cancer-related deaths in the U.S. (10).

The dismal survival rate stems from multiple factors, the most significant being that the majority of patients are diagnosed at an advanced stage, making treatment exceedingly challenging. In other words, most individuals with pancreatic cancer exhibit no symptoms until the disease has progressed, with late-stage symptoms complicating early detection and intervention (11).

Screening individuals for pancreatic cancer also presents significant challenges. A primary obstacle is the low prevalence of pancreatic ductal adenocarcinoma (PDAC) in the general population, which reduces the pre-test probability of a positive laboratory result. Even highly specific biomarker assays capable of accurately identifying PDAC could result in many individuals undergoing imaging studies unnecessarily, causing anxiety despite not having the disease. As a result, the U.S. Prevention and Screening Task Force (USPSTF) has assigned a "D" grade to PDAC screening in the general population, indicating that it is ineffective and may even pose harm (12).

Pancreatic cancer can develop in various ways. As mentioned earlier, the pancreas consists of three major parts: the body, head, and tail. These regions do not experience the same incidence rates of cancer. The most common type of pancreatic cancer is PDAC, which is currently the third leading cause of cancer-related deaths in the U.S. By 2030, it is expected to become the second leading cause of cancer deaths, reflecting its increasing prevalence and mortality rates (13).

According to data from the Surveillance, Epidemiology, and End Results (SEER) registries in the U.S., approximately 77.5% of PDAC cases originate near the pancreatic head, which has been the primary focus of discussions on pancreatic cancer. Additionally, the annual incidence of pancreatic head cancer is significantly higher than that of pancreatic body or tail cancer, with a rate of 5.6 per 100,000 compared to 1.6 per 100,000, respectively (14).

Genetic Defects Involved in Pancreatic Cancer

From a genetic perspective, PDAC is characterized by frequently altered genes, including *KRAS* and tumor suppressor genes such as *TP53*, *CDKN2A*, and *SMAD4* (15).

***KRAS* Can Cause Continuous Cell Division**

The *KRAS* gene, a member of the rat sarcoma (*RAS*) viral oncogene family, is one of three isoforms found in humans, with *HRAS* and *NRAS* being the other two. Mutations in the *RAS* family, which play critical roles in regulating cell proliferation and differentiation, are implicated in several cancers, including PDAC, lung adenocarcinoma, and colorectal adenocarcinoma (16).

KRAS mutations are found in approximately 8 out of 10 pancreatic cancer cases, and up to 90% of PDAC patients exhibit alterations in the *KRAS* gene. This prevalence underscores the pivotal role of *KRAS* mutations in pancreatic cancer development (17). Specifically, *KRAS* alterations are thought to contribute significantly to tumor growth and resistance to treatment. Therefore, a thorough understanding of *KRAS* biology and its role in PDAC is critical for developing innovative therapies for this deadly disease.

KRAS proteins are guanosine triphosphatases (*GTPases*) that regulate various cellular processes, including cell cycle progression, actin cytoskeletal organization, and cell motility. *RAS* proteins are located on the inner side of the cell membrane, where they mediate communication between activated transmembrane receptors and cytoplasmic effectors. These proteins act as molecular switches, alternating between guanosine diphosphate (GDP)-bound inactive

[DOI: 10.18502/jabs.v15i1.17548]

states and guanosine triphosphate (GTP)-bound active states (18).

Guanine nucleotide exchange factors (*GEFs*) facilitate the exchange of GDP for GTP on *KRAS*, thereby activating the protein. Once activated, *KRAS* interacts with downstream effectors such as *RAF*, *RAL*, and *PI3K*, initiating signaling cascades that activate pathways like PI3K and MAPK. In the absence of *GEFs*, *KRAS* remains inactive, and downstream signaling pathways are not activated.

Conversely, GTPase-activating proteins (*GAPs*) accelerate the hydrolysis of GTP to GDP, thereby inactivating *KRAS***.** If *GAPs* fail to function properly, *KRAS* remains in its active form, leading to uncontrolled cell growth and proliferation. Maintaining the proper balance between *GEFs* and *GAPs* is essential to regulate *KRAS* activity and ensure appropriate control of downstream signaling pathways (Figure 2) (18, 19).

How Genetic Alterations in Tumor Suppressor Genes Affect PDAC?

Tumor suppressor genes (TSGs) play a critical role in regulating cell growth and proliferation by counteracting the effects of oncogenic driver mutations. Their mechanisms of action include inducing cell cycle arrest, apoptosis, and senescence, which collectively limit the expansion of potentially harmful cells (20).

PDAC represents a malignancy primarily driven by genetic mutations that promote tumorigenesis. Specifically, the loss of function in certain tumor suppressor genes has been identified as a major contributor to this process. Among these *TSGs*, *CDKN2A*, *TP53*, and *SMAD4/DPC4* are the most frequently affected, resulting in an impaired ability to restrain excessive cell growth .

The *CDKN2A* gene encodes two distinct proteins, p16*INK4A* and p14*ARF*, which play pivotal roles in cellular regulation.

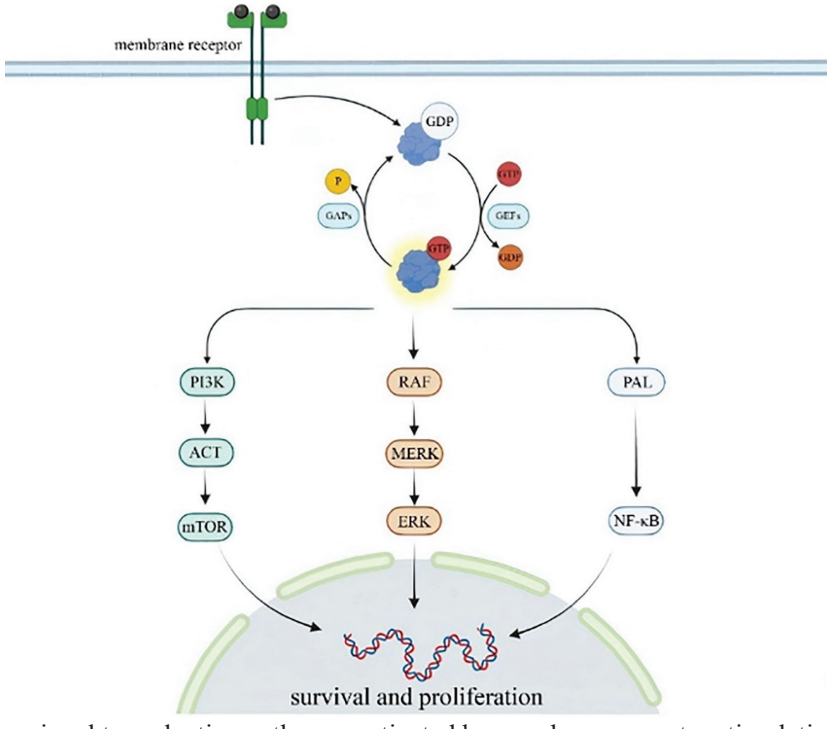

Figure 2. The key signal transduction pathways activated by membrane receptor stimulation, leading to cell survival and proliferation. The cascade is initiated with receptor activation, triggering GDP/GTP cycling regulated by GAPs and GEFs. This activation subsequently branches into three major signaling pathways: the PI3K/AKT/mTOR pathway, the RAF/MERK/ERK cascade, and the PAL/NF-κB pathway. These parallel signaling cascades ultimately converge to regulate gene expression in the nucleus, thereby promoting mechanisms of cellular survival and proliferation.

[DOI: 10.18502/jabs.v15i1.17548]

While p16*INK4A* inhibits cyclin-dependent kinases to promote cellular differentiation, p14*ARF* regulates the activity of the tumor suppressor protein *p53*. *TP53*, in turn, is essential for preventing the propagation of damaged DNA and promoting apoptosis during PDAC progression. Additionally, *SMAD4/DPC4*, which functions as a transcriptional regulator within the transforming growth factor beta (*TGF-β*) signaling pathway, is vital for maintaining a balance between cell proliferation and differentiation. Its suppression significantly contributes to neoplastic transformation by promoting uncontrolled cell proliferation and reducing differentiation efficiency (21).

The functional inactivation of these *TSGs* in PDAC is thought to drive disease progression by enabling cells to bypass critical checkpoints that would otherwise prevent their proliferation. As such, identifying and characterizing alterations in these genes has become essential for enhancing our understanding of the mechanisms underlying PDAC and for developing more effective diagnostic and therapeutic strategies.

Mutated *TP53* Can Cause Metastasis in PDAC Cells

The *TP53* gene is the most significant member of the TSG family, playing a pivotal role in cancer prevention by producing the p53 protein, which regulates cell division and promotes apoptosis, also known as programmed cell death. However, in PDAC, mutations in the *TP53* genetic code result in the disruption of this critical tumor-suppressing mechanism. When the p53 protein becomes dysfunctional, it fails to regulate uncontrolled cellular growth, ultimately leading to the onset of PDAC (22, 23).

Research has demonstrated that mutant p53 cannot activate essential genes such as *CDKN1A/p21*, *BAX*, *NOXA*, and *PUMA*, which are critical for responding to cellular stress or DNA damage. The impaired expression of these vital genes is directly linked to the loss of p53's DNA-binding ability, thereby rendering it ineffective in initiating its target gene responses. Consequently, cells harboring mutant p53 cannot undergo apoptosis or cell cycle arrest, resulting in unchecked proliferation and the formation of cancerous tissue (24). The interaction between p53 and these genes is intricate, involving multiple signaling pathways and regulatory mechanisms:

I. CDKN1A/p21

CDKN1A/p21, a cyclin-dependent kinase inhibitor, mediates cell cycle arrest in response to p53 activation. When p53 is triggered by stress signals, it transcribes *CDKN1A*, which inhibits cyclin-dependent kinases and halts the cell cycle at the G1 phase. This arrest provides an opportunity for DNA repair or initiates apoptosis if the damage is irreparable, thus ensuring genomic stability and preventing tumorigenesis (25-27).

II. BAX

BAX, a pro-apoptotic member of the Bcl-2 family, is upregulated by p53 in response to cellular stress, facilitating apoptosis (28). The activation of BAX promotes mitochondrial outer membrane permeabilization, leading to the release of cytochrome c and the activation of caspases, which execute programmed cell death. This pathway is essential for eliminating damaged cells that could otherwise contribute to cancer progression (29-31).

III. NOXA and PUMA

NOXA and PUMA, also pro-apoptotic proteins regulated by p53, play a pivotal role in the apoptotic response under conditions of severe stress. NOXA interacts with anti-apoptotic proteins such as Mcl-1, while PUMA binds to both pro- and anti-apoptotic members of the Bcl-2 family. Their expression ensures that cells with irreparable damage are driven to apoptosis, preventing their survival with potentially oncogenic mutations (32-35).

The signaling pathways connecting p53 and its target genes are highly diverse and intricately linked. The DNA Damage Response (DDR) pathway is activated upon DNA damage, leading

[DOI: 10.18502/jabs.v15i1.17548]

to the phosphorylation and stabilization of p53, which in turn transcribes its target genes. In addition, hypoxia signaling modulates p53 activity, influencing its interaction with hypoxia-inducible factors (HIFs), thereby impacting tumor progression and metastasis. Furthermore, aberrant signaling within the MAPK/PI3K pathways, which are closely associated with p53 status, can impair p53 functionality and alter the expression of immune checkpoint proteins (36-39).

Molecular Mechanisms of *TP53* Mutations in PDAC

The mutation spectrum of *TP53* in PDAC exhibits distinct patterns, with several notable hotspot regions predominantly located within the DNA-binding domain (DBD), which spans exons 5–8. The most prevalent hotspot mutations in PDAC occur at codons 175, 248, and 273, with arginine residues being particularly prone to mutational events. These specific sites are essential for DNA binding and for preserving the structural integrity of the p53 protein (40–43).

The mutational landscape of *TP53* in PDAC encompasses various genetic alterations, with missense mutations being the most frequently observed type of *TP53* mutation in cancer. For instance, studies have reported 1,297 unique missense somatic mutations out of a total of 29,891 genomic mutations recorded in the *TP53* Database. These missense mutations typically result in single amino acid substitutions, which can lead to either a loss of function or the acquisition of oncogenic properties (gain-of-function mutations). Meanwhile, frameshift and nonsense mutations are less common and generally produce non-functional protein products.

Among the specific hotspot mutations in PDAC, R273H, R248W, and R175H are the most frequently detected within the arginine residues of the DNA-binding domain (44). These sites play a crucial role in DNA binding and in maintaining the structural integrity of the p53 protein, particularly within the L2 and L3 zinc-binding domains and the LSH (loop-sheet-helix) motif (Figure 3) (45).

Scientific studies have demonstrated that mutations in the *TP53* gene play a significant role in facilitating metastasis, a process

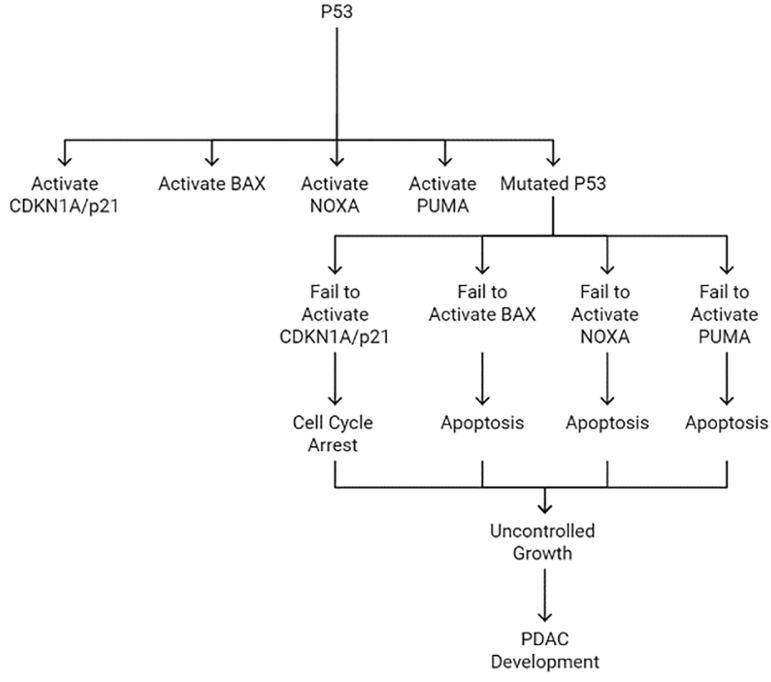

Figure 3. The Relationship between the *TP53* gene and its associated genes (*CDKN1A/p21, BAX, NOXA,* and *PUMA*) and their signaling pathway

[DOI: 10.18502/jabs.v15i1.17548]

characterized by the spread of malignant cells from their primary site to distant organs via blood circulation or lymphatic pathways, ultimately leading to the formation of secondary tumors. The dysregulation induced by these mutations enhances the proliferation rate and invasive potential of pancreatic cancer cells, including those in PDAC, thereby substantially contributing to disease progression (46, 47). On the other hand, the relationship between p53 signaling and metastasis is complex. Functional p53 exerts a tumor-suppressive effect by inducing apoptosis in cells that have acquired aggressive traits. However, mutations in the *TP53* gene can result in gain-of-function activities that not only fail to suppress metastasis but actively promote it. Furthermore, the loss or mutation of *TP53* enhances cellular plasticity, enabling cancer cells to adapt to new microenvironments during metastasis (48–50).

Therefore, understanding the genetically mediated mechanisms underlying *TP53* mutations is crucial for developing targeted therapeutic strategies aimed at preventing uncontrolled solid tumor growth and inhibiting further dissemination across various tissues, thereby mitigating the life-threatening impact of PDAC.

Mutated *CDKN2A* Is Present in Nearly All PDAC Patients

The *CDKN2A* gene is a critical genetic determinant located on chromosome 9p21, encoding two essential proteins: p16^INK4a and p14^ARF. These proteins play a pivotal role in regulating cell proliferation by modulating distinct phases of the cell cycle (51). Specifically, p16^INK4a functions by inducing G1-phase cell cycle arrest, achieved through inhibition of cyclin-dependent kinase 4 (CDK4) activity and prevention of retinoblastoma (Rb) protein phosphorylation. Similarly, p14^ARF, the other protein encoded by *CDKN2A*, maintains cellular homeostasis **by** inhibiting HDM2, thereby stabilizing p53 tumor suppressor levels and preventing damaged cells from progressing through replication (52).

The *CDKN2A* gene is also highly significant in PDAC. Notably, somatic mutations or alterations in *CDKN2A* have been identified in approximately 95% of pancreatic cancer cases. From a molecular standpoint, loss of function in this tumor suppressor gene can occur through several mechanisms, including homozygous deletions (40%), intragenic mutations coupled with loss of the remaining allele (40%), or hypermethylation of the promoter region (~15%) (53). These findings underscore the critical importance of understanding the molecular alterations driving tumorigenesis in PDAC, as such insights may provide a foundation for the development of targeted therapeutic strategies.

Half of Pancreatic Cancers Are Attributable to Defects in SMAD4

SMAD4 (also referred to as DPC4) is a tumor suppressor gene located on chromosome 18q21.1 and belongs to the *SMAD* family of proteins, which play a crucial role in regulating the TGF-β signaling pathway, a mechanism that inhibits epithelial cell proliferation. *SMAD4* was first identified as a tumor suppressor gene in pancreatic cancer by Harn et al. and was designated DPC4 (deleted in pancreatic carcinoma, locus 4) (54).

Functioning as a cofactor, *SMAD4* enhances gene transcription and tumor suppression through this signaling pathway. The *SMAD4* status is considered a key molecular feature that distinguishes the two major subtypes of PDAC: *SMAD4*-positive and *SMAD4*-negative cancers. Loss of *SMAD4* expression has been associated with poor prognostic indicators, including accelerated tumor progression, increased metastatic potential, and reduced survival rates (55).

Mutations in the *SMAD4* gene are detected in approximately 60% of PDAC cases. Additionally, the TGF-β signaling pathway, which is disrupted in nearly 47% of PDAC cases, exhibits diverse oncogenic functions, particularly in PDAC.

[DOI: 10.18502/jabs.v15i1.17548]

Given the pivotal role of *SMAD4* in TGF-β signaling and its frequent inactivation in PDAC, it represents a promising target for the development of novel therapeutic strategies to combat this malignancy (56). Thus, elucidating the intricate relationship between *SMAD4* and PDAC could provide valuable insights into the pathophysiology of this disease.

Diagnosis and Treatment of PDAC

The diagnosis and treatment of pancreatic cancer pose significant challenges due to the disease's complexity. Unfortunately, the vast majority of cases are detected at an advanced stage, with either locally invasive or metastatic disease. Indeed, 80%–85% of pancreatic cancers are deemed incurable at the time of diagnosis, underscoring the urgent need for improved diagnostic tools (57). In other words, most cases are diagnosed at an advanced stage, and recurrence rates remain high.

Currently, various imaging techniques are employed for the early detection of pancreatic cancer, with many of these modalities being used in conjunction with tissue sampling to enhance diagnostic accuracy. The primary imaging techniques for pancreatic cancer diagnosis include computed tomography (CT) and magnetic resonance imaging (MRI), both of which are widely utilized. Another valuable imaging modality is positron emission tomography (PET), which helps detect and assess the extent of pancreatic cancer. Additionally, endoscopic ultrasound (EUS), which provides high-resolution imaging and enables fine-needle aspiration (FNA) biopsy for tissue collection, is frequently employed for both diagnosis and staging (58).

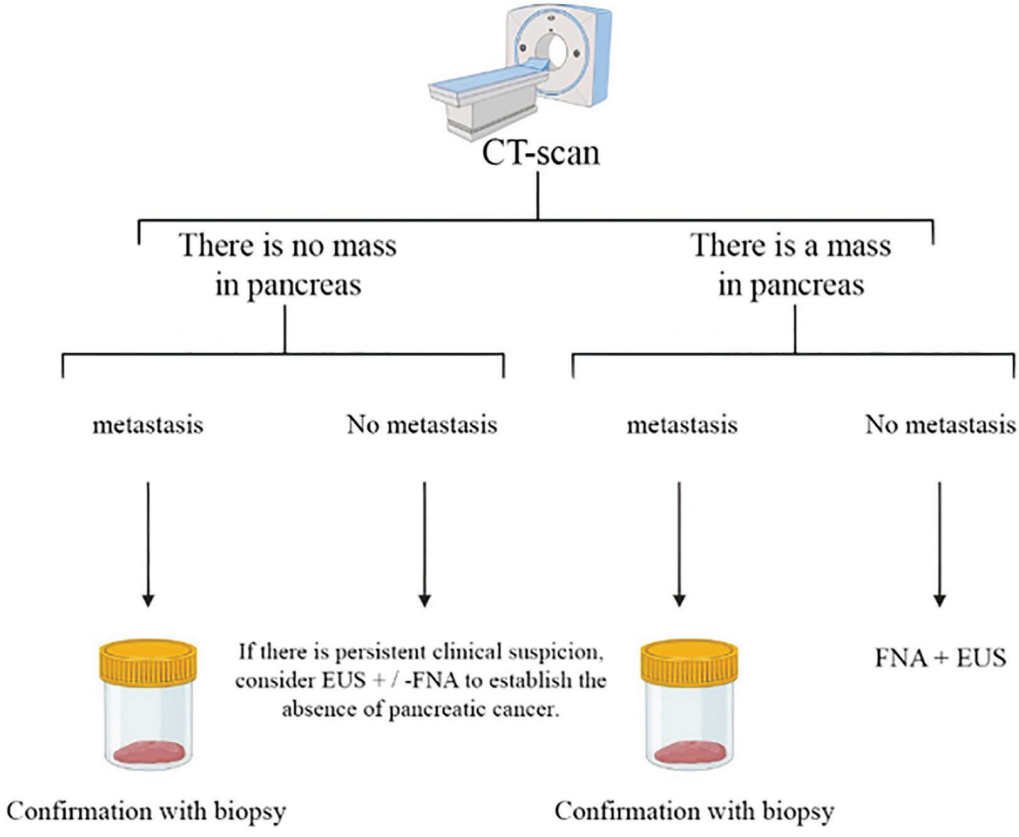

Figure 4. The standard diagnostic approach for pancreatic cancer. In the presence of clinical symptoms, patients should first undergo evaluation through imaging modalities, such as computed tomography (CT) scanning. Abdominal imaging can provide critical insights that inform subsequent diagnostic and treatment strategies.

[DOI: 10.18502/jabs.v15i1.17548]

Among the available imaging modalities, multi-detector computed tomography (MDCT) is the most widely accessible and well-validated technique for evaluating patients with pancreatic cancer. MDCT offers comprehensive anatomical coverage and high spatial resolution through multi-planar imaging, allowing for accurate differentiation between tumors and normal pancreatic parenchyma**.** Several studies have demonstrated that CT imaging is highly effective in diagnosing pancreatic cancer. A large meta-analysis comparing various imaging techniques reported CT's sensitivity and specificity to be 89% and 90%, respectively, which is comparable to MRI (59).

Recent advancements in MDCT have further improved its diagnostic performance, with reported sensitivities reaching 96% for pancreatic cancer detection. These improvements are attributed to thin collimation imaging, enhanced spatial and temporal resolution, and the integration of multi-planar reconstruction and three-dimensional (3D) imaging techniques (Figure 4) (60).

Surgery

The primary treatment options for pancreatic cancer include surgery, chemotherapy, radiation therapy, or a combination of these modalities (61). Among these, surgical resection remains the only potentially curative approach. While the addition of adjuvant chemotherapy to surgical resection has been shown to improve survival rates, long-term prognosis remains poor (62, 63).

Several types of surgical procedures are employed in the treatment of pancreatic cancer, including the Whipple procedure (pancreaticoduodenectomy), distal pancreatectomy, and total pancreatectomy. The choice of procedure depends on the tumor's size and location.

• The Whipple procedure, or pancreaticoduodenectomy, is the most common surgical approach for tumors located in the head of the pancreas. This procedure involves the removal of the pancreatic head, duodenum, gallbladder, and parts of the stomach and bile duct.

• Distal pancreatectomy is performed when the tumor is located in the body or tail of the pancreas.

• Total pancreatectomy involves the complete removal of the pancreas, spleen, gallbladder, and portions of the small intestine and bile duct. Due to its severe long-term consequences, including insulin-dependent diabetes, this procedure is rarely performed (64–67).

Although early-stage surgical resection provides the best opportunity for prolonging survival, the median survival for resected patients is less than 20 months. Only a small percentage of patients achieve long-term survival, while the majority do not (68). This underscores the critical need for continued research into more effective treatment options for pancreatic cancer.

Chemotherapy

Chemotherapy is another key strategy for slowing disease progression. The treatment landscape for pancreatic cancer typically involves two lines of chemotherapy, which are outlined below.

First-Line Chemotherapy

The choice of first-line chemotherapy is influenced by several factors, including disease stage, overall patient health, and individual preferences. However, the two most commonly used first-line chemotherapy regimens for advanced pancreatic cancer are:

1. FOLFIRINOX
2. Gemcitabine/nab-paclitaxel (69)

FOLFIRINOX is a combination chemotherapy regimen consisting of four drugs: 5-fluorouracil (5-FU), irinotecan, leucovorin, and oxaliplatin (70). While highly effective, it is associated with significant toxicity and side effects, including diarrhea, nausea, fatigue, neuropathy, and myelosuppression. These adverse effects can be partially managed with supportive medications such as antiemetics and antidiarrheals (71, 72). Despite its higher toxicity, FOLFIRINOX has been shown to significantly prolong overall survival and progression-free survival compared

[DOI: 10.18502/jabs.v15i1.17548]

to gemcitabine monotherapy (73).

Another widely used first-line chemotherapy regimen is gemcitabine/nab-paclitaxel, which combines gemcitabine with nanoparticle albumin-bound (nab) paclitaxel. Studies have indicated that following the failure of FOLFIRINOX therapy, switching to gemcitabine/nab-paclitaxel is a more viable alternative than other treatment options (74).

Second-Line Chemotherapy

There are no universally established second-line treatments for progressed pancreatic cancer following failure of first-line chemotherapy with FOLFIRINOX or gemcitabine/nab-paclitaxel. However, current clinical practice involves switching to an alternative chemotherapy regimen, depending on the initial treatment:

• Patients previously treated with gemcitabine-based chemotherapy are often transitioned to a 5-FU-based regimen.

• Conversely, those who initially received FOLFIRINOX may be switched to gemcitabine/nab-paclitaxel.

Although no randomized controlled trials have definitively established the optimal second-line regimen, several retrospective single-institution analyses suggest that gemcitabine/nab-paclitaxel remains a reasonable second-line option following FOLFIRINOX failure (75).

Radiotherapy

Radiotherapy is another potential treatment option for patients with pancreatic cancer. However, current evidence suggests that neither radiation therapy alone nor its combination with chemotherapy has led to a significant improvement in patient survival (76). Although it remains uncertain whether radiotherapy enhances survival outcomes, it is the only therapeutic approach—aside from surgery—that has been shown to improve local disease control (63).

One of the major limitations of conventional radiation therapy is its high toxicity. However, the development of intensity-modulated radiotherapy (IMRT) has helped reduce treatment-related toxicity associated with traditional techniques. Despite these advancements, several challenges persist in radiotherapy studies for PDAC, including the lack of consensus on optimal dosage, fractionation, and tumor delineation, which can lead to variability in treatment protocols and potentially impact survival rates (77).

An emerging and promising radiotherapy technique is stereotactic body radiation therapy (SBRT), which has demonstrated encouraging results in the treatment of pancreatic cancer, particularly in improving local tumor control compared to traditional external beam radiation therapy (EBRT). While SBRT may pose a higher risk of late toxicity, continued advancements in treatment planning, delivery techniques, and a deeper understanding of normal tissue tolerance are expected to mitigate these risks over time (78).

Future of Pancreatic Cancer Treatments: The Road Ahead

The evidence presented thus far underscores that conventional treatments, such as surgery and chemotherapy, have not significantly reduced pancreatic cancer-related mortality. Therefore, to effectively lower the death toll associated with this malignancy, it is imperative to develop novel treatment strategies.

One of the most promising emerging therapies is gene therapy, which has garnered global interest among researchers. Gene therapy involves the introduction of foreign genetic material into host cells to modify gene expression or alter cellular functions. Initially designed to treat genetic disorders, gene therapy is now being explored for a wide range of hereditary and acquired diseases, including cancer (79).

The gene therapy process begins with the delivery of therapeutic genes into host cells. In the context of cancer treatment, viral vectors are the most widely used gene delivery method. These vectors come in various forms, including adenoviruses, retroviruses, lentiviruses, and adeno-associated viruses (AAVs). Each type of viral vector has its own set of advantages and

[DOI: 10.18502/jabs.v15i1.17548]

limitations, which influence its suitability for specific therapeutic applications (80).

Oncolytic Virotherapy

Oncolytic virotherapy is an emerging cancer treatment strategy that involves selectively infecting and destroying cancer cells while sparing healthy cells. The viruses used in oncolytic virotherapy may either occur naturally or be genetically modified to enhance their ability to infect and eliminate malignant cells. Once cancer cells are infected, the virus begins to replicate, ultimately causing the host cells to lyse (break open), releasing tumor antigens and other immune-stimulating molecules. This process helps activate the immune system, enabling it to recognize and attack remaining cancer cells, thereby contributing to tumor eradication (81).

Beyond direct tumor cell destruction, the viruses released during lysis can infect neighboring cancer cells, triggering a self-propagating therapeutic effect that enhances treatment efficacy. This chain reaction ensures that all cancerous cells in the affected area are targeted and eliminated. Numerous studies have demonstrated that oncolytic virotherapy holds significant potential for treating a wide range of malignancies, including pancreatic cancer (82). However, further research is necessary to fully elucidate its mechanisms of action and optimize the therapeutic application of oncolytic viruses in cancer treatment.

Adenoviruses (Ads) are a prominent class of viral vectors that, due to their exceptional gene-delivery capabilities both *in vitro* and *in vivo*, serve as a foundation for developing oncolytic therapies (83). To enhance both the safety and efficacy of cancer treatments, conditionally replicative adenoviruses (CRAds) have been engineered. These viruses are designed to replicate exclusively in tumor cells, leaving healthy surrounding tissues unaffected (84).

CRAds are classified into two main categories:

1. Mutation-based CRAds – These can replicate only in specific tumor types that compensate for the loss of function caused by targeted gene alterations or deletions in the E1 region.

2. Cancer-specific promoter-based CRAds – These replicate exclusively in cancer cells where the regulating promoter is active, as they depend on tumor-specific promoter-controlled transcription of the E1 region (85).

Examples of cancer-specific promoter-based CRAds include OBP-301, which expresses E1A under the control of the human telomerase reverse transcriptase (*hTERT*) promoter, and AduPARE1A, which drives E1A expression via the urokinase-type plasminogen activator receptor (*uPAR*) promoter (86). In preclinical pancreatic cancer models, these CRAds have demonstrated selective replication and potent anticancer activity.

In addition to viral-based gene delivery, alternative methods such as physical and chemical vector-based approaches have been developed for transferring therapeutic genes into target cells (87).

Physical Vectors

Physical methods of gene transfer include electroporation, microinjection, and microparticle bombardment (88).

• Microinjection involves the direct insertion of nucleic acids into a single living cell using a micropipette. This technique is highly precise, enabling researchers to target specific cells with exceptional accuracy. Microinjection is commonly employed when only small quantities of nucleic acid need to be transferred in an experiment (89).

• Microparticle bombardment, also known as biolistics or the "gene gun" method, involves propelling nucleic acid-coated microparticles into target cells using high-pressure helium gas. The microparticles, typically composed of gold, tungsten, or silver, act as carriers to deliver genetic material into living cells. This method is particularly useful for transfecting cells that are difficult to modify using conventional techniques (88, 90).

[DOI: 10.18502/jabs.v15i1.17548]

• Electroporation is another widely used gene delivery technique. It involves applying high-voltage electrical pulses to temporarily disrupt the target cell's membrane, allowing plasmid DNA to enter the cytoplasm (91). This technique is frequently used to introduce nucleic acids into various cell types, including mammalian, bacterial, and plant cells (92, 93). Due to its high efficiency, electroporation has become a preferred method in industrial applications, such as biomanufacturing (94).

Chemical Vectors

Chemical vectors are a class of gene delivery systems that utilize non-viral carriers to introduce genetic material into cells. These vectors have been extensively studied due to their unique advantages, including safety, scalability, low toxicity, cost-effectiveness, and ease of preparation (95).

One of the most widely used chemical vectors for gene transfer is cationic lipids, commonly referred to as liposomes (96, 97). Liposomes are vesicular structures composed of an aqueous core enclosed by a phospholipid bilayer. In addition to phospholipids, these vectors also contain sterols such as cholesterol, which contribute to their structural stability. The phospholipid bilayer consists of two layers of phospholipid molecules: one with hydrophilic heads facing the aqueous environment and the other with hydrophobic tails oriented toward the bilayer's interior (98).

Cationic lipids interact electrostatically with negatively charged nucleic acids, such as DNA and RNA, due to their positively charged head groups. When combined with nucleic acids, lipoplexes—lipid-nucleic acid complexes—are formed. These lipoplexes can then be delivered into cells, where they are internalized via endocytosis (99, 100). Once inside the cell, the lipoplexes are transported to endosomes, where the acidic environment facilitates fusion between the lipids and the endosomal membrane, enabling the release of nucleic acids into the cytoplasm. The delivered genetic material can then enter the nucleus and integrate into the host genome, leading to gene expression (101).

Cationic lipids offer several key advantages as chemical vectors for gene transfer. They are cost-effective, non-toxic, and elicit minimal immune responses, making them relatively safe for therapeutic applications. However, they also present certain limitations, including low transfection efficiency and a tendency to aggregate in the presence of serum or other proteins, which may compromise their effectiveness (102–104).

Genetic Biomarkers for Early Detection: Current Advances and Clinical Applications

Recent advancements in molecular diagnostics have transformed cancer detection through the use of genetic biomarkers, offering unprecedented opportunities for early diagnosis and real-time tumor monitoring. One of the most promising non-invasive biomarkers is circulating tumor DNA (ctDNA), which allows for continuous tracking of tumor dynamics and molecular evolution. Studies have shown that ctDNA analysis can detect cancer-specific genetic alterations months before they become apparent on conventional imaging, with sensitivity rates reaching up to 87.18% in certain malignancies (105–110).

In the case of PDAC, research has demonstrated that ctDNA analysis can identify key genetic mutations associated with the disease, particularly KRAS mutations, which are present in more than 80% of PDAC cases. This approach not only facilitates early detection but also provides critical insights into tumor heterogeneity and its evolutionary trajectory over time (111, 112).

Beyond ctDNA, gene expression markers have been explored as potential diagnostic tools for PDAC. Using high-throughput technologies, such as RNA sequencing and microarray analysis, researchers have identified distinct gene expression patterns associated with pancreatic cancer. These molecular signatures

[DOI: 10.18502/jabs.v15i1.17548]

can differentiate PDAC from benign pancreatic conditions, offering a valuable complement to traditional imaging techniques. Furthermore, certain gene expression markers have been correlated with clinical outcomes, providing prognostic insights that may inform therapeutic decision-making (112).

Another emerging avenue of research involves mutation detection in body fluids beyond blood, such as urine and saliva. The presence of tumor-derived genetic material in these biofluids offers the potential for non-invasive diagnostic testing, further enhancing early detection efforts. Studies have confirmed that KRAS mutations can be identified in pancreatic juice and urine samples from PDAC patients, reinforcing their potential as biomarkers for early diagnosis (111, 113).

Finally, novel molecular diagnostic techniques are being developed to increase the sensitivity and specificity of PDAC detection. Liquid biopsy technologies, which analyze ctDNA and other tumor-derived biomarkers from blood samples, are gaining traction due to their ability to provide real-time insights into tumor biology without requiring invasive procedures. Additionally, next-generation sequencing (NGS) technologies now enable comprehensive profiling of genetic alterations in PDAC, facilitating the identification of novel biomarkers that could be leveraged for earlier and more precise cancer detection (114–116).

Treatments Based on Gene Therapy

In the field of oncology, ongoing clinical trials are actively exploring innovative gene therapy-based approaches for the treatment of PDAC. These investigations primarily focus on two promising modalities: RNA interference (RNAi)-based therapeutics delivered via nanomedicine platforms and Chimeric Antigen Receptor T-cell (CAR-T) therapy.

RNAi-based therapeutics, when administered through nanomedicine platforms, offer a highly targeted strategy for PDAC treatment by selectively silencing oncogenes. This approach utilizes small interfering RNA (siRNA) molecules, which bind to and degrade specific messenger RNA (mRNA) sequences within cancer cells, thereby inhibiting the translation of oncogenic proteins and suppressing tumor growth and progression. The incorporation of nanocarriers, such as nanoparticles and liposomes, enhances siRNA delivery, improving its stability, cellular uptake, and tumor specificity, while simultaneously minimizing off-target effects. This method is particularly valuable in overcoming PDAC's inherent resistance to conventional therapies by disrupting key molecular pathways that drive tumor aggressiveness (117).

Concurrently, CAR-T therapy represents another promising gene therapy approach for treatment-refractory PDAC. This technique involves genetically engineering a patient's T-cells to express CARs that specifically recognize tumor-associated antigens on PDAC cells. By enhancing T-cell cytotoxicity, CAR-T therapy circumvents the immune evasion mechanisms that make pancreatic cancer particularly challenging to treat. This personalized immunotherapy offers a potentially transformative treatment option for patients with refractory PDAC who have not responded to standard therapeutic regimens (118, 119).

These emerging gene therapy strategies, currently under evaluation in clinical trials, represent substantial advancements in the search for more effective PDAC treatments. Given that pancreatic cancer has historically exhibited resistance to conventional therapies, these approaches hold considerable promise in addressing the disease's genetic underpinnings and improving patient outcomes.

Conclusion

Pancreatic cancer is an aggressive malignancy with a grim prognosis, primarily due to its asymptomatic nature in early stages and its resistance to standard treatments. Despite notable advancements in surgical techniques and chemotherapy regimens, therapeutic outcomes

[DOI: 10.18502/jabs.v15i1.17548]

remain suboptimal. Consequently, there is an urgent need to develop novel targeted therapies that can improve survival rates and provide more effective treatment alternatives.

Gene therapy-based strategies, such as oncolytic virotherapy and other molecularly targeted approaches, hold significant potential by directly addressing the genetic alterations that drive pancreatic tumorigenesis. However, further research is required to optimize these techniques, assess their safety and efficacy, and translate preclinical findings into clinically viable treatments.

A comprehensive understanding of the genetic landscape of pancreatic cancer, combined with the development of personalized treatment regimens, may pave the way for substantial progress against this lethal disease. Future studies should prioritize the refinement of gene therapy approaches, leveraging promising preclinical data to develop clinically effective treatments for pancreatic cancer patients.

Acknowledgements

This manuscript has been written by the authors.

Conflict of Interest

The authors declare no conflicts of interest.

Authors' Contribution

M.M.M.T conceptualized the study. M.M.M.T and A.G. drafted the draft. A.G. and M.Gh. edited the manuscript.

References

1 Migita K, Matsumoto S, Wakatsuki K, Ito M, Kunishige T, Nakade H, et al. The anatomical location of the pancreas is associated with the incidence of pancreatic fistula after laparoscopic gastrectomy. Surg Endosc. 2016;30:5481-9.
2 Mahadevan V. Anatomy of the pancreas and spleen. Surgery (Oxford). 2019;37(6):297-301.
3 Yuan Q, Pan A, Fu Y, Dai Y. Anatomy and physiology of the pancreas. Integrative pancreatic intervention therapy: Elsevier; 2021. p. 3-21.
4 Jennings RE, Berry AA, Strutt JP, Gerrard DT, Hanley NA. Human pancreas development. Development. 2015;142(18):3126-37.
5 Chandra R, Liddle RA. Neural and hormonal regulation of pancreatic secretion. Curr Opin Gastroenterol. 2009;25(5):441.
6 Röder PV, Wu B, Liu Y, Han W. Pancreatic regulation of glucose homeostasis. Experiment Mol Med. 2016;48(3):e219-e.
7 Zimmet PZ, Magliano DJ, Herman WH, Shaw JE. Diabetes: a 21st century challenge. Lancet Diabetes Endocrinol. 2014;2(1):56-64.
8 Cooperman AM. Pancreatic cancer: the bigger picture. Surg Clin. 2001;81(3):557-74.
9 Chari ST, Leibson CL, Rabe KG, Ransom J, De Andrade M, Petersen GM. Probability of pancreatic cancer following diabetes: a population-based study. Gastroenterol. 2005;129(2):504-11.
10 Liu J, Kang R, Tang D. The art of war: ferroptosis and pancreatic cancer. Front Pharmacol. 2021;12:3697.
11 Kamisawa T, Wood LD, Itoi T, Takaori K. Pancreatic cancer. Lancet. 2016;388(10039):73-85.
12 Singhi AD, Koay EJ, Chari ST, Maitra A. Early detection of pancreatic cancer: opportunities and challenges. Gastroenterol. 2019;156(7):2024-40.
13 Chandana S, Babiker HM, Mahadevan D. Therapeutic trends in pancreatic ductal adenocarcinoma (PDAC). Expert Opin Investig Drugs. 2019;28(2):161-77.
14 Ling Q, Xu X, Zheng S-S, Kalthoff H. The diversity between pancreatic head and body/tail cancers: clinical parameters and in vitro models. Hepatobiliary Pancreat Dis Int. 2013;12(5):480-7.
15 Chen X, Kang R, Kroemer G, Tang D. Targeting ferroptosis in pancreatic cancer: a double-edged sword. Trends Cancer. 2021;7(10):891-901.
16 Huang L, Guo Z, Wang F, Fu L. KRAS mutation: from undruggable to druggable in cancer. Signal transduct Target Ther. 2021;6(1):386.
17 He Q, Liu Z, Wang J. Targeting KRAS in PDAC: A New Way to Cure It? Cancers. 2022;14(20):4982.
18 Tomasini P, Walia P, Labbe C, Jao K, Leighl NB. Targeting the KRAS pathway in non-small cell lung cancer. Oncologist. 2016;21(12):1450-60.
19 Bos JL, Rehmann H, Wittinghofer A. GEFs and GAPs: critical elements in the control of small G proteins. Cell. 2007;129(5):865-77.
20 Marshall CJ. Tumor suppressor genes. Cell. 1991;64(2):313-26.
21 Ying H, Dey P, Yao W, Kimmelman AC, Draetta GF, Maitra A, et al. Genetics and biology of

[DOI: 10.18502/jabs.v15i1.17548]

pancreatic ductal adenocarcinoma. Gen Develop. 2016;30(4):355-85.
22 Rozenblum E, Schutte M, Goggins M, Hahn SA, Panzer S, Zahurak M, et al. Tumor-suppressive pathways in pancreatic carcinoma. Cancer Res. 1997;57(9):1731-4.
23 Barton C, Staddon S, Hughes C, Hall P, O'sullivan C, Klöppel G, et al. Abnormalities of the p53 tumour suppressor gene in human pancreatic cancer. Br J Cancer. 1991;64(6):1076-82.
24 Khoo KH, Verma CS, Lane DP. Drugging the p53 pathway: understanding the route to clinical efficacy. Nat Rev Drug discov. 2014;13(3):217-36.
25 Abbas T, Dutta A. p21 in cancer: intricate networks and multiple activities. Nat Rev Cancer. 2009;9(6):400-14.
26 Kreis NN, Louwen F, Yuan J. The Multifaceted p21 (Cip1/Waf1/CDKN1A) in Cell Differentiation, Migration and Cancer Therapy. Cancers (Basel). 2019;11(9):1220.
27 Karimian A, Ahmadi Y, Yousefi B. Multiple functions of p21 in cell cycle, apoptosis and transcriptional regulation after DNA damage. DNA Repair. 2016;42:63-71.
28 Hao Q, Chen J, Lu H, Zhou X. The ARTS of p53-dependent mitochondrial apoptosis. J Mol Cell Biol. 2023;14(10):mjac074.
29 Dewson G, Kluck RM. Mechanisms by which Bak and Bax permeabilise mitochondria during apoptosis. J Cell Sci. 2009;122(Pt 16):2801-8.
30 Aubrey BJ, Kelly GL, Janic A, Herold MJ, Strasser A. How does p53 induce apoptosis and how does this relate to p53-mediated tumour suppression? Cell Death Different. 2018;25(1):104-13.
31 Chipuk JE, Maurer U, Green DR, Schuler M. Pharmacologic activation of p53 elicits Bax-dependent apoptosis in the absence of transcription. Cancer Cell. 2003;4(5):371-81.
32 Roufayel R, Younes K, Al-Sabi A, Murshid N. BH3-Only Proteins Noxa and Puma Are Key Regulators of Induced Apoptosis. Life (Basel, Switzerland). 2022;12(2):256.
33 Akhtar RS, Geng Y, Klocke BJ, Latham CB, Villunger A, Michalak EM, et al. BH3-only proapoptotic Bcl-2 family members Noxa and Puma mediate neural precursor cell death. The Journal of neuroscience. J Neusci. 2006;26(27):7257-64.
34 van Delft MF, Huang DCS. How the Bcl-2 family of proteins interact to regulate apoptosis. Cell Res. 2006;16(2):203-13.
35 Dumont A, Lohard S, Maillet LA, Juin P, Barillé-Nion SC. NOXA the BCL-2 family member behind the scenes in cancer treatment. J Cell Signal. 2020;1(4):127.
36 Zhang C, Liu J, Wang J, Zhang T, Xu D, Hu W, et al. The interplay between tumor suppressor p53 and hypoxia signaling pathways in cancer. Front Cell Develop Biol. 2021;9:648808.
37 Kciuk M, Kołat D, Kałuzińska-Kołat Ż, Gawrysiak M, Drozda R, Celik I, et al. PD-1/PD-L1 and DNA Damage Response in Cancer. Cells. 2023;12(4):530.
38 Chen Z, Han F, Du Y, Shi H, Zhou W. Hypoxic microenvironment in cancer: molecular mechanisms and therapeutic interventions. Signal Transduct Target Ther. 2023;8(1):70.
39 Sun Q, Hong Z, Zhang C, Wang L, Han Z, Ma D. Immune checkpoint therapy for solid tumours: clinical dilemmas and future trends. Signal Transduct Target Ther. 2023;8(1):320.
40 Saha G, Singh R, Mandal A, Das S, Chattopadhyay E, Panja P, et al. A novel hotspot and rare somatic mutation p.A138V, at TP53 is associated with poor survival of pancreatic ductal and periampullary adenocarcinoma patients. Mol Med (Cambridge, Mass). 2020;26(1):59.
41 Butz J, Wickstrom E, Edwards J. Characterization of mutations and loss of heterozygosity of p53 and K-ras2 in pancreatic cancer cell lines by immobilized polymerase chain reaction. BMC Biotechnol. 2003;3:11.
42 Katkoori VR, Chatla C, Jia X, Grizzle WE, Manne U. Mutations in the L3 loop of zinc-binding domain of p53 are poor prognostic indicators of Caucasians with colorectal adenocarcinomas. Cancer Res. 2005; 65 (9_Supplement): 598.
43 Hill RA, Liu Z, Liu YY. Small ceramide tames big p53 mutant beast. Oncotarget. 2020;11(37):3418-9.
44 Pan M, Jiang C, Zhang Z, Achacoso N, Alexeeff S, Solorzano AV, et al. TP53 Gain-of-Function and Non-Gain-of-Function Mutations Are Associated With Differential Prognosis in Advanced Pancreatic Ductal Adenocarcinoma. JCO Precision Oncol. 2023;7:e2200570.
45 McCubrey JA, Yang LV, Abrams SL, Steelman LS, Follo MY, Cocco L, et al. Effects of TP53 Mutations and miRs on Immune Responses in the Tumor Microenvironment Important in Pancreatic Cancer Progression. Cells. 2022;11(14): 2155.
46 Morton JP, Timpson P, Karim SA, Ridgway RA, Athineos D, Doyle B, et al. Mutant p53 drives metastasis and overcomes growth arrest/senescence in pancreatic cancer. Proc Natl Acad Sci U S A. 2010;107(1):246-51.
47 Grant TJ, Hua K, Singh A. Molecular pathogenesis

[DOI: 10.18502/jabs.v15i1.17548]

of pancreatic cancer. Prog Mol Biol Transl Sci. 2016;144:241-75.
48 Marvalim C, Datta A, Lee SC. Role of p53 in breast cancer progression: An insight into p53 targeted therapy. Theranost. 2023;13(4):1421-42.
49 Rivlin N, Brosh R, Oren M, Rotter V. Mutations in the p53 Tumor Suppressor Gene: Important Milestones at the Various Steps of Tumorigenesis. Gen Cancer. 2011;2(4):466-74.
50 Kastenhuber ER, Lowe SW. Putting p53 in Context. Cell. 2017;170(6):1062-78.
51 Serrano M, Hannon GJ, Beach D. A new regulatory motif in cell-cycle control causing specific inhibition of cyclin D/CDK4. Nature. 1993;366(6456):704-7.
52 Kimura H, Klein AP, Hruban RH, Roberts NJ. The role of inherited pathogenic CDKN2A variants in susceptibility to pancreatic cancer. Pancreas. 2021;50(8):1123-30.
53 Schutte M, Hruban RH, Geradts J, Maynard R, Hilgers W, Rabindran SK, et al. Abrogation of the Rb/p16 tumor-suppressive pathway in virtually all pancreatic carcinomas. Cancer Res. 1997;57(15):3126-30.
54 Miyaki M, Kuroki T. Role of Smad4 (DPC4) inactivation in human cancer. Biochem Biophys Res Communic. 2003;306(4):799-804.
55 Kassardjian A, Wang HL. SMAD4-expressing pancreatic ductal adenocarcinomas have better response to neoadjuvant therapy and significantly lower lymph node metastasis rates. Pancreas. 2020;49(9):1153-60.
56 Dardare J, Witz A, Merlin J-L, Gilson P, Harlé A. SMAD4 and the TGFβ pathway in patients with pancreatic ductal adenocarcinoma. Int J Mol Sci. 2020;21(10):3534.
57 McGuigan A, Kelly P, Turkington RC, Jones C, Coleman HG, McCain RS. Pancreatic cancer: A review of clinical diagnosis, epidemiology, treatment and outcomes. World J Gastroenterol. 2018;24(43):4846.
58 Zhang L, Sanagapalli S, Stoita A. Challenges in diagnosis of pancreatic cancer. World J Gastroenterol. 2018;24(19):2047.
59 Treadwell JR, Zafar HM, Mitchell MD, Tipton K, Teitelbaum U, Jue J. Imaging tests for the diagnosis and staging of pancreatic adenocarcinoma: a meta-analysis. Pancreas. 2016;45(6):789-95.
60 Raman SP, Horton KM, Fishman EK. Multimodality imaging of pancreatic cancer—computed tomography, magnetic resonance imaging, and positron emission tomography. Cancer J. 2012;18(6):511-22.
61 Gemenetzis G, Groot VP, Blair AB, Laheru DA, Zheng L, Narang AK, et al. Survival in locally advanced pancreatic cancer after neoadjuvant therapy and surgical resection. Ann Surg. 2019;270(2):340.
62 Versteijne E, Vogel JA, Besselink M, Busch O, Wilmink J, Daams J, et al. Meta-analysis comparing upfront surgery with neoadjuvant treatment in patients with resectable or borderline resectable pancreatic cancer. J Brit Surg. 2018;105(8):946-58.
63 Hammel P, Huguet F, van Laethem J-L, Goldstein D, Glimelius B, Artru P, et al. Effect of chemoradiotherapy vs chemotherapy on survival in patients with locally advanced pancreatic cancer controlled after 4 months of gemcitabine with or without erlotinib: the LAP07 randomized clinical trial. JAMA. 2016;315(17):1844-53.
64 Yang F, Jin C, Warshaw AL, You L, Mao Y, Fu D. Total pancreatectomy for pancreatic malignancy with preservation of the spleen. J Surg Oncol. 2019;119(6):784-93.
65 Canto MI, Kerdsirichairat T, Yeo CJ, Hruban RH, Shin EJ, Almario JA, et al. Surgical outcomes after pancreatic resection of screening-detected lesions in individuals at high risk for developing pancreatic cancer. J Gastrointest Surg. 2020;24:1101-10.
66 Eberlin LS, Margulis K, Planell-Mendez I, Zare RN, Tibshirani R, Longacre TA, et al. Pancreatic cancer surgical resection margins: Molecular assessment by mass spectrometry imaging. PLoS Med. 2016;13(8):e1002108.
67 Griffin JF, Poruk KE, Wolfgang CL. Pancreatic cancer surgery: past, present, and future. Chin J Cancer Res. 2015;27(4):332.
68 Dal Molin M, Zhang M, De Wilde RF, Ottenhof NA, Rezaee N, Wolfgang CL, et al. Very Long-term Survival Following Resection for Pancreatic Cancer Is Not Explained by Commonly Mutated Genes: Results of Whole-Exome Sequencing AnalysisVery Long-term Survival in Pancreatic Cancer. Clin Cancer Res. 2015;21(8):1944-50.
69 Springfeld C, Jäger D, Büchler MW, Strobel O, Hackert T, Palmer DH, et al. Chemotherapy for pancreatic cancer. Presse Med. 2019;48(3):e159-e74.
70 Suker M, Beumer BR, Sadot E, Marthey L, Faris JE, Mellon EA, et al. FOLFIRINOX for locally advanced pancreatic cancer: a systematic review and patient-level meta-analysis. The Lancet Oncology. 2016;17(6):801-10.
71 Kleeff J, Korc M, Apte M, La Vecchia C, Johnson CD, Biankin AV, et al. Pancreatic cancer. Nat Rev Dis Prim. 2016;2(1):1-22.
72 Tong H, Fan Z, Liu B, Lu T. The benefits of modified FOLFIRINOX for advanced pancreatic cancer and its induced adverse events: a systematic review and meta-analysis. Sci Rep. 2018;8(1):8666.

[DOI: 10.18502/jabs.v15i1.17548]

73 Conroy T, Hammel P, Hebbar M, Ben Abdelghani M, Wei AC, Raoul J-L, et al. FOLFIRINOX or gemcitabine as adjuvant therapy for pancreatic cancer. New Eng J Med. 2018;379(25):2395-406.
74 Williet N, Saint A, Pointet A-L, Tougeron D, Pernot S, Pozet A, et al. Folfirinox versus gemcitabine/nab-paclitaxel as first-line therapy in patients with metastatic pancreatic cancer: a comparative propensity score study. Therapeut Adv Gastroenterol. 2019;12:1756284819878660.
75 Saung MT, Zheng L. Current standards of chemotherapy for pancreatic cancer. Clin Therapeut. 2017;39(11):2125-34.
76 Chin V, Nagrial A, Sjoquist K, O'Connor CA, Chantrill L, Biankin AV, et al. Chemotherapy and radiotherapy for advanced pancreatic cancer. Cochrane Database Syst Rev. 2018(3):1-68.
77 Bouchart C, Navez J, Closset J, Hendlisz A, Van Gestel D, Moretti L, et al. Novel strategies using modern radiotherapy to improve pancreatic cancer outcomes: toward a new standard? Therapeut Adv Med Oncol. 2020;12:1758835920936093.
78 Trakul N, Koong AC, Chang DT. Stereotactic body radiotherapy in the treatment of pancreatic cancer. Semin Radiat Oncol; 2014;24(2):140-147.
79 Belete TM. The current status of gene therapy for the treatment of cancer. Biologic Target Ther. 2021;18(15):67-77.
80 Wirth T, Ylä-Herttuala S. Gene Therapy Used in Cancer Treatment. Biomedicines. 2014;2(2):149-62.
81 Chen J, Weihs D, Vermolen FJ. A cellular automata model of oncolytic virotherapy in pancreatic cancer. Bulletin Mathematic Biol. 2020;82(8):103.
82 Wennier S, Li S, McFadden G. Oncolytic virotherapy for pancreatic cancer. Expert Rev Mol Med. 2011;13:e18.
83 Hallenbeck PL, Chang Y-N, Hay C, Golightly D, Stewart D, Lin J, et al. A novel tumor-specific replication-restricted adenoviral vector for gene therapy of hepatocellular carcinoma. Human Gen Ther. 2004;10(10): :1721-33.
84 Zhang J, Ding M, Xu K, Mao L, Zheng J. shRNA-armed conditionally replicative adenoviruses: a promising approach for cancer therapy. Oncotarget. 2016;7(20):29824.
85 Sato-Dahlman M, Roach BL, Yamamoto M. The role of adenovirus in cancer therapy. Cancers. 2020; 12(11): 3121.
86 Maliandi MV, Mato-Berciano A, Sobrevals L, Roué G, José A, Fillat C. AduPARE1A and gemcitabine combined treatment trigger synergistic antitumor effects in pancreatic cancer through NF-κB mediated uPAR activation. Mol Cancer. 2015;14(1):1-12.
87 Ramamoorth M, Narvekar A. Non viral vectors in gene therapy-an overview. J Clin Diagnost Res. 2015;9(1):GE01.
88 Liu SX, Xia ZS, Zhong YQ. Gene therapy in pancreatic cancer. World J Gastroenterol. 2014;20(37):13343-68.
89 Chi Z, Xu Q, Zhu L. A review of recent advances in robotic cell microinjection. IEEE Access. 2020;8:8520-32.
90 Yahya EB, Alqadhi AM. Recent trends in cancer therapy: A review on the current state of gene delivery. Life Sci. 2021;269:119087.
91 Gehl J. Electroporation: theory and methods, perspectives for drug delivery, gene therapy and research. Acta Physiolo Scand. 2003;177(4):437-47.
92 Rols M-P, Teissié J. Electropermeabilization of mammalian cells to macromolecules: control by pulse duration. Biophys J. 1998;75(3):1415-23.
93 Mir LM. Application of electroporation gene therapy: past, current, and future. Methods Mol Biol. 2008; 423:3-17.
94 Sayed N, Allawadhi P, Khurana A, Singh V, Navik U, Pasumarthi SK, et al. Gene therapy: Comprehensive overview and therapeutic applications. Life Sci. 2022:120375.
95 Hosseinkhani H, Abedini F, Ou KL, Domb AJ. Polymers in gene therapy technology. Polymer Adv Technol. 2015;26(2):198-211.
96 Felgner PL, Tsai YJ, Sukhu L, Wheeler CJ, Manthorpe M, Marshall J, et al. Improved cationic lipid formulations for in vivo gene therapy. Ann New York Acad Sci. 1995;772:126-39.
97 Ewert K, Slack NL, Ahmad A, Evans HM, Lin AJ, Samuel CE, et al. Cationic lipid-DNA complexes for gene therapy: understanding the relationship between complex structure and gene delivery pathways at the molecular level. Curr Med Chem. 2004;11(2):133-49.
98 Lasic DD, Templeton N. Liposomes in gene therapy. Adv Drug Deliv Rev. 1996;20(2-3):221-66.
99 Liu C, Zhang L, Zhu W, Guo R, Sun H, Chen X, et al. Barriers and strategies of cationic liposomes for cancer gene therapy. Mol Ther Methods Clin Develop. 2020;18:751-64.
100 Zu H, Gao D. Non-viral vectors in gene therapy: Recent development, challenges, and prospects. AAPS J. 2021;23(4):78.
101 Hart SL. Lipid carriers for gene therapy. Curr Drug Deliv. 2005;2(4):423-8.
102 Lv H, Zhang S, Wang B, Cui S, Yan J. Toxicity of cationic lipids and cationic polymers in gene delivery. J Controlled Release. 2006;114(1):100-9.

[DOI: 10.18502/jabs.v15i1.17548]

103 Kumar V, Singh R, Chaudhuri A. Cationic transfection lipids in gene therapy: successes, set-backs, challenges and promises. Curr Medicin Chem. 2003;10(14):1297-306.
104 Dodds E, Dunckley M, Naujoks K, Michaelis U, Dickson G. Lipofection of cultured mouse muscle cells: a direct comparison of Lipofectamine and DOSPER. Gen Ther. 1998;5(4):542-51.
105 Wan JC, Massie C, Garcia-Corbacho J, Mouliere F, Brenton JD, Caldas C, et al. Liquid biopsies come of age: towards implementation of circulating tumour DNA. Nat Rev Cancer. 2017;17(4):223-38.
106 Bae M, Kim G, Lee TR, Ahn JM, Park H, Park SR, et al. Integrative modeling of tumor genomes and epigenomes for enhanced cancer diagnosis by cell-free DNA. Nat Communicat. 2023;14(1):2017.
107 Zhao J, Reuther J, Scozzaro K, Hawley M, Metzger E, Emery M, et al. Personalized Cancer Monitoring Assay for the Detection of ctDNA in Patients with Solid Tumors. Mol Diagn Ther. 2023;27(6):753-68.
108 Liu Y, Liu Y, Wang Y, Li L, Yao W, Song Y, et al. Increased detection of circulating tumor DNA by short fragment enrichment. Translat Lung Cancer Res. 2021;10(3):1501-11.
109 Li K, Hu Y, Li L, Ren Z, Jiang L. Progress of circulating-tumor DNA in the precise medicine for advanced-stage cancer. Chin J Laborator Med. 2019;42:176-80.
110 Cao M, Shi J, Xia C, Li H, Cai W, Qi X, et al. Efficacy of ctDNA methylation combined with traditional detection modality to detect liver cancer among high-risk patients: A multicenter diagnostic trial. Chin J cancer Res. 2023;35(1):58-65.
111 Singhi AD, George B, Greenbowe JR, Chung J, Suh J, Maitra A, et al. Real-Time Targeted Genome Profile Analysis of Pancreatic Ductal Adenocarcinomas Identifies Genetic Alterations That Might Be Targeted With Existing Drugs or Used as Biomarkers. Gastroenterol. 2019;156(8):2242-53.e4.
112 Hong TS, Deshpande V, Ancukiewicz M, Yeap BY, Borger DR, Wo JY, et al. Genetic, tissue, and plasma biomarkers of outcomes from a prospective study of neoadjuvant short course proton-based chemoradiation for resectable pancreatic ductal adenocarcinoma (PDAC). J Clin Oncol. 2013;31:4047.
113 Lee DKC, Loke R, Chow JT, Gabra M, Salmena L. Phosphoinositide-focused CRISPR screen identifies novel genetic vulnerabilities in PDAC and AML cells. bioRxiv. 2024:2024-09.
114 Workman S, Jabbour SK, Deek MP. A narrative review of genetic biomarkers in non-small cell lung cancer: an update and future perspectives. AME Med J. 2023;8:6.
115 Mohler MJ, Fain MJ, Wertheimer AM, Najafi B, Nikolich-Žugich J. The Frailty syndrome: clinical measurements and basic underpinnings in humans and animals. Experiment Gerontol. 2014;54:6-13.
116 Chen X, Zhang H, Xiao B. C9orf16 represents the aberrant genetic programs and drives the progression of PDAC. BMC Cancer. 2022;22(1):1102.
117 Won EJ, Park H, Yoon TJ, Cho YS. Gene Therapy Using Nanocarriers for Pancreatic Ductal Adenocarcinoma: Applications and Challenges in Cancer Therapeutics. Pharmaceutics. 2022;14(1):137.
118 DeSelm CJ, Tano ZE, Varghese AM, Adusumilli PS. CAR T-cell therapy for pancreatic cancer. J Surg Oncol. 2017;116(1):63-74.
119 Akce M, Zaidi MY, Waller EK, El-Rayes BF, Lesinski GB. The Potential of CAR T Cell Therapy in Pancreatic Cancer. Front Immunol. 2018;9:2166.

[DOI: 10.18502/jabs.v15i1.17548]