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Unravelling Multifaceted p73: From Cell Kinetics to Therapeutics

Dentistry Section

ANITHA DAYAKAR[1], PUSHPARAJA SHETTY[2]

ABSTRACT

The p73 gene is an important member of the p53 gene family, which includes p53, p63, and p73 genes. The p73 gene was identified by McKeon's group in 1997 and localised to 1p36 region. The p73 shows a complex interplay with both, p53 and p63 genes. Similar to p63, but unlike p53, p73 has an important role in development especially neuronal development, apart from tumourigenesis. The p73 gene plays a vital role in the development of tumours and exhibits both tumour suppressor and oncogenic behaviour. This dual function is attributed to the presence of the two different variants: TAp73 and ΔNp73 with mutually divergent functions. There exists a dynamic relationship between the multiple p73 isoforms. The p73 has a major role in regulating apoptosis, and Deoxyribonucleic Acid (DNA) damage related cell cycle arrest. It can replace p53 in various apoptotic pathways, as it is capable of transcription of p53 responsive genes. The p73 also participates in myriad processes including angiogenesis, epithelial-mesenchymal transformation, senescence, and maintaining genomic stability. The p73 overexpression has been detected in a number of human cancers. Since it is frequently overexpressed rather than mutated, it is a promising potential candidate for developing therapeutic strategies in various cancers as well as, for predicting the prognosis. It is also implicated in the lack of response to chemotherapeutic drugs. However, the entire spectrum of cellular mechanisms with respect to p73 is still not understood clearly. This article explores the multiple roles of p73, especially in tumour development processes, and also, its recent novel applications in therapeutics.

Keywords: Apoptosis, Immunohistochemistry, p53 family, Tumour suppressor

INTRODUCTION

The p53 family includes p53, p63, and p73 genes. These genes show considerable structural homology and also share common functions like apoptosis and cell cycle arrest [1]. Although the p53 gene has been reported as mutated in 50% of the cancers, p73 seldom shows mutation, instead, it may show overexpression in many tumours [2]. The p73 is mapped to chromosome 1p36.2-3, a region that is reported to be lost frequently in many human neoplasms including neuroblastoma [3].

The p73 gene was initially reported in 1997, as a p53-like gene since it showed considerable homology with p53, especially in the DNA binding domain. It was observed that the p53 gene could regulate many of the p53 target genes, thereby triggering apoptosis. The p73 was subsequently localised to the chromosome 1p36.3, locus which was known to harbour a tumour suppressor gene related to neuroblastoma, melanoma, breast, and prostate cancers [3,4].

Kaghad M et al., in 1997, reported the identification of a new gene p73 with remarkable similarity to the DNA binding domain and oligomerisation domain of p53. The investigators noted that p73 was capable of inducing p21 and also, the presence of two splice variants, p73α and p73β [3].

De Laurenzie V et al., identified two novel p73 splice variants namely p73ϒ and p73δ showing varied transcriptional functions. Simultaneously, De Laurenzie V et al., also reported two isoforms differing in the C-terminal region due to alternative splicing, one isoform was similar to p73 ϒ, while the other was labelled as p73ε [5].

The p73 gene is approximately 60 kb in length, with 14 exons [6]. It encodes two different proteins, which show mutually antagonistic functions:

1. Full-length Transcriptionally Active TAp73 isoforms.
2. NH_2 terminal truncated dominant-negative ΔNp73 isoforms [4].

As a member of the p53 family, the p73 structure shows the presence of an Amino-terminal Transactivation Domain (TAD), a central DNA Binding Domain (DBD), and a carboxy-terminal Oligomerisation Domain (OD). The p73 also contains a carboxy-terminal Inhibitory region and an additional Sterile α Motif (SAM) [7].

The p73 protein sequencing showed a 63% similarity with p53 in the DNA binding domain, 29% similarity in the TAD, and 42% similarity in the OD [7].

Like other p53 family members, p73 has two promoters, P1, which is located upstream of exon 1, and P2, which is present within exon 3. The presence of alternative promoters and splicing leads to the encoding of different isoforms. Transcribing from the P1 promoter produces TA isoforms of p73. The amino terminal truncated Np73 isoforms are obtained using the alternative P2 promoter [6,7].

Studies have identified various TAp73 isoforms (α, β, ϒ, δ, ε, θ, φ, η, and η1) generated by splicing at the C-terminus site, TAp73α and TAp73β being considered most responsive to p53 targets [7,8]. Splicing at the NH_2 terminal gives rise to six other p73 isoforms which include ΔNp73α, ΔNp73β, ΔN'p73α, ΔN'p73β, Ex2Delp73, and Ex2/3Delp73 [7]. The ΔN isoforms are generated by the truncation of the transactivation domain at the N terminus end, these isoforms, exhibit inhibitory functions with respect to p53 and TAp73 isoforms [4,7,9].

The TAp73 shares many functions native to p53, like inhibition of cell cycle progression, apoptosis, cell metabolism, and ageing. This is made possible by the fact that TAp73 is capable of triggering the transcriptional targets of the p53 gene, like p21/waf1 gene, involved in cell cycle regulation, p53 Upregulated Modulator of Apoptosis (PUMA), NOXA, Bcl-2 Associated X protein (BAX), and p53 Inducible Gene 3 (PIG3) associated with apoptosis, and GADD45, 14-3-3-σ, and p53 AIP1 [9].

Studies on reporter and gel shift assays concluded that TAp73, as well as TAp63 activate the p53 amenable targets to mediate apoptosis [9,10].

In contrast, ΔNp73 isoforms function as a powerful dominant-negative inhibitor of TAp73, TAp63, and p53. The ultimate outcome of this interplay depends on the TA/ΔN p73 ratio [9]. Two mechanisms of ΔNp73 dominance over TA isoforms have been put forward:

1. **Promoter competition:** This involves blocking the target gene promoters by ΔNp73 since they share a similar DNA binding domain. This process prevents TA isoform transcription.
2. **Heterocomplex formation:** ΔNp73 isoforms generate hetero-oligomeric complexes, which are incapable of inducing transcription themselves, and also block the TA isoforms [7,11].

The p53 and TAp73 control ΔNp73 levels via transcription from the P2 promoter of p73 gene.

Although p73 was considered a tumour suppressor gene, paradoxically, mouse knockout models for p73, failed to show increased tumour formation or early mortality as observed in p53 null animal models. Rather, the p73 knockout mice showed brain development abnormalities like hippocampus dysgenesis, hydrocephalus due to hypersecretion from choroid plexus, and mucous hypersecretion from respiratory mucosa leading to inflammation and infection [12].

ROLE OF p73 GENE

p73 and Development

p73 plays a pivotal role in development and differentiation with a distinct biological role. It is necessary for neuronal differentiation, nervous system, and olfactory system development [11].

p73 has been proposed as the guardian of the male germline. TAp73 null mice show increased DNA damage in spermatogonia, which results in male infertility because of derangement of spermatogenesis [13].

p73 and Genomic Stability

p73 performs an important role in spindle assembly during mitosis. The absence of TpA73, leads to the inability to initiate or maintain cell cycle arrest, triggering genomic instability. TAp73 loss increases spontaneous or carcinogen-induced tumour formation and also infertility because of genomic instability of the oocyte [11].

The p73 gene, especially TAp73 performs a paramount role in maintaining genomic stability during cell division. Loss of p73 leads to aneuploidy and polyploidy due to aberrant cyclin/Cyclin Dependent Kinases (CDK) complex activation. p73 interaction with BubR1 is necessary for Spindle Assembly Checkpoint (SAC) functioning. Loss of TAp73 leads to defective SAC, which fails to assist the mitotic cells culminating in aneuploidy, cancer, and infertility [7].

The p73 gene also acts as a safeguard mechanism in the prevention of abnormal mitosis by regulating cell death during prolonged mitotic arrest or dysfunctional spindle assembly checkpoint activity [7].

p73 and Apoptosis

The mainstay of p73 as a tumour suppressor gene is because of its contribution toward apoptosis. p73 mRNA levels are upregulated as a consequence of E2F1 mediated transcription. The Tp73 gene possesses multiple E2F1 binding sites in its promoter region which facilitate E2F1 binding and transcription. p73 activation further leads to transactivation of downstream p53 responsive genes and apoptosis.

Conversely, the activity of ΔNp73 isoforms in p73 mutants leads to an inhibition of E2F1 mediated apoptosis, in p53-null Mouse Embryo Fibroblasts (MEF) and p53- defective tumour cell lines [9].

p73 induced apoptosis can be broadly segregated as:

1. Transcription-dependent p73- related apoptosis.
2. Transcription-independent p73-related apoptosis.

Transcription-dependent p73 related apoptosis: It Involves p73 modulated transactivation of various pro-apoptotic genes. Apoptotic activity of p73 is usually attributed to its transactivation of pro-apoptotic genes, which include PUMA, NOXA, Bax, GRAMD4, Apoptin, and CD95. The role of p73 is to transactivate the effector genes, thus, indirectly inducing them to initiate apoptosis in the cytoplasm or mitochondria. p73 directly transactivates PUMA following DNA damage and facilitates Bax translocation to the mitochondria, leading to the liberation of cytochrome c, which in turn, results in apoptosis [2,14,15].

GRAMD4: It is another pro-apoptotic gene transactivated by p73 and undergoes mitochondrial translocation. GRAMD4, itself, is responsible for Bax upregulation, promoting apoptosis. GRAMD4 responds only to p73, and not to p53, thus, making it a selective mechanism restricted to the p73 gene [15].

Apoptin: Apoptin overexpression leads to TAp73 stablilisation via the p73 Induced RING 2 protein (PIR2) pathway followed by transactivation of the downstream pro-apoptotic genes like PUMA. PIR2 is also capable of ΔNp73 degradation, further stabilising pro-apoptotic TAp73 [16].

CD95: TAp73 transactivates CD95 causing apoptosis through a caspase-dependent mechanism via death receptor CD95 through the extrinsic pathway [15].

Transcription-independent p73-related apoptosis: The mechanism is not fully understood. It is associated with cytoplasmic localisation of p73. Predominantly, this type of apoptosis is modulated by WW domain-containing Oxidoreductase (WWOX). Another mechanism involves Ran-binding Protein 9 (RanBP9) in the mitochondria, leading to stabilisation of p73 levels at both transcriptional and post-transcriptional stages [15].

Bcl family proteins: p73 is capable of modulating apoptosis by a novel pathway directly interacting with the Bcl-2 family proteins in the mitochondria [15].

Tumour Necrosis Factor-related Ligand (TRAIL): In Death receptor-ligand TRAIL-mediated apoptosis, TAp73 undergoes sequestration by caspases 3 and 8, and the recombinant products induce cytochrome c release from the mitochondria [7,17,18].

p73 Reaction to DNA Damage and Genotoxic Stress

DNA damage leads to p73 multi-site phosphorylation by non-receptor kinase c-abl, ultimately culminating in p73 protein stabilisation and triggering apoptosis [19]. Ionising radiation leads to activation of c-abl tyrosine kinase in the presence of a serine protein kinase Ataxia Telangiectasia Mutated (ATM) [14]. Following administration of genotoxic drugs, checkpoint kinases Chk2 and Chk1, which are downstream genes in the ATM pathway, modulate p73 activation via the E2F1 transcription factor [2,18]. Chk2 is also capable of direct p73 phosphorylation, thereby enhancing the apoptotic pathway. Another mechanism that has been suggested for p73 activation during cisplatin treatment is the nuclear collection of the IKK-α protein, ultimately leading to apoptosis. As a result of DNA damage, different stress activated mechanisms including JNK/SAPK as well as p38MAPK are involved in p73 regulation [9].

The p73 upregulation via the JNK pathway is modulated through the c-jun transcription factor, which promotes p73 stabilisation by inhibiting proteasome-mediated p73 degradation. Following cisplatin treatment, JNK is capable of direct TAp73 phosphorylation, stabilising TAp73 with a proapoptotic effect [20].

The p38 protein also facilitates p73 activation and stabilisation via c-abl protein. This process involves direct p73 phosphorylation [20]. Another mechanism of p73 modulation involves isomerisation changes induced by Pin-1, which increases p73 acetylation leading to p73 stabilisation/following DNA damage. YAP protein has an important role in p73 stabilising following DNA damage, thus enhancing p73-dependent apoptosis. p73 acetylation is mediated by p300 and enhanced by Yes Associated Protein (YAP). Also, YAP1 competes with ITCH protein (an E3 ubiquitin ligase responsible for p73 breakdown) for p73 binding, thus preventing p73 degradation [21].

The p73 gene interacts with Apoptosis Stimulating Proteins of p53 (ASPP) family members inducing downstream activation of apoptotic genes like PIG3, Bax, and PUMA. Another ASPP family member, iASSP has been found to have oncogenic inhibitory functions [22]. Blocking iASSP-TAp73 with the help of a small peptide leads to p73-induced transcription and apoptosis [23]. Protein kinase C δ (PKCδ) along with active catalytic fragment Protein Kinase C Catalytic Fragment (PKCCF) also, activates p73 dependent transcription and stabilises p73 protein following DNA damage [18].

In some instances, inhibitory phosphorylation of p73 has been observed with certain proteins like Haematopoietic Cell Kinase (HCK), c-src, Protein Kinase A (PKA), Polo-Like Kinase 1 (Plk-1) as well as CDKs. The effect of these molecules is to directly phosphorylate p73, leading to its inhibition [9].

p73 Protein Degradation

The p73 degradation occurs in a proteasome-dependent manner. The p73 protein undergoes degradation following ubiquitination by several E3 ligases. ITCH protein, which is a HECT family ubiquitin ligase, acts upon both TA and ΔN forms of p73. The NQO1 and UFD2 molecules participate in the proteasomal TAp73 degradation in a ubiquitin independent manner following DNA damage [20]. Calcium-dependent protease calpain-1 also leads to proteolytic degradation of p73 [24,25]. The degradation of p73 is isoform-specific. ITCH mediated degradation is restricted to α and β isoforms [9]. The TA isoforms undergo stabilisation following DNA damage through downregulation of ITCH. In contrast genotoxic stress destabilises ΔNp73α [26].

p73 in Ageing and Senescence

TAp73 induces BRCA1, MRE11, and RAD50 transcription, which participate in homologous recombination. Experiments on TAp73 null mice showed premature ageing and senescence due to metabolic derangement. The TAp73 promotes ATG5 expression, thus preventing ageing by the maintenance of homeostatic control and regulation of autophagy [13,27].

The p73 overexpression is associated with the upregulation of a number of DNA repair proteins including DNA PK [11].

p73 and Epithelial-mesenchymal Transformation (EMT)

Abrus Agglutinin (AGG) prevents invasion of tumour cells by Snail protein degradation via p73 dependent pathway in oral cancer cells [28]. Also, studies show that deficiency of p73 in pancreatic duct adenocarcinoma promoted an increase in the EMT, driven by TGF-β activation and suppression of SMAD-dependent pathway [7,29,30]. Restin (a Melanoma Antigen Gene superfamily molecule) prevents EMT and metastasis by regulating tumour metastasis suppressor mir-200a/b in breast cancer cell lines in a p73-dependent pathway [31].

Role of p73 in Angiogenesis

A majority of the studies have concluded that ΔNp73 promotes angiogenesis in tumours; however, the role of TAp73 remains unclear [30,32]. There are two schools of thought:

1. TAp73 has a negative regulatory effect on Hypoxia Inducing Factor 1α (HIF-1α) via the MDM2 degradation pathway with a net antiangiogenic effect [30].
2. As an angiogenesis promoter, TAp73 is stabilised by hypoxia and subsequently leads to the activation of pro-angiogenic targets like VEGF-A. There was a correlation between tumour size and blood vessel density with TAp73 expression in Xenograft models [30].

Conflicting TAp73 behaviour in angiogenesis might be explained either based on temporal effects or the intensity of TAp73 activation. Transient TAp73 overexpression supported angiogenesis while long-term TAp73 induction resulted in antigenic suppression [32].

The majority of studies support a direct association between TAp73 and VEGF-A expression [27]. However, a few studies have reported an inverse relationship between TAp73 and VEGF-A [32].

Role of p73 in Oral Cancer

Immunohistochemistry studies, cell line studies, and Reverse Transcription Polymerase Chain Reaction (RT-PCR) studies have been conducted in head and neck cancer including oral cancer with a number of utilities including prediction of outcome or survival, targeting the gene for therapeutic purpose, predicting the drug response and/or chemoresistance, as well as understanding the neoplastic process.

p73-induced apoptosis is an important tumour-suppressor mechanism in head and neck cancer. Suppression of p73-induced apoptosis by ΔNp63α was found to be critical to the survival of Head and Neck Squamous Cell Carcinoma (HNSCC) cell lines, thus, perpetuating the tumour cells [33].

Faridoni-Laurens et al., demonstrated the p73 expression in the undifferentiated basal cells of the normal oral epithelium and attributed a role for the p73 gene in the maintenance of the undifferentiated cells of the mucosa. An RT-PCR study also suggested an important role for p73 in head and neck tumourigenesis. Yet another RT-PCR study in oral cancer, suggested a dominant oncogenic role for ΔNp73. Reduction in the Δ Np73 levels conferred a small trend of better overall survival status in HNSCC. The TAp73, which has a tumour suppressor function, was also weakly expressed but appeared to be functionally irrelevant [34].

An immunohistochemical study by Chen et al., suggested that p73 expression in buccal carcinomas may represent an initial event in oral cancer and could be a predictor for oral epithelial dysplasia transformation into frank cancer [34].

A number of anti-cancer drugs primarily function by triggering apoptosis through DNA damage resulting in cytotoxicity. The HNSCC cell lines subjected to cytotoxic drugs like cisplatin, taxol, doxorubicin, and etoposide lead to an increase in the p73 levels triggering apoptosis. Conversely, p73 inhibition by p53 mutants promoted chemoresistance to the anti-cancer drugs in the clinical scenario. Thus, p73 modulates chemosensitivity as well as drug resistance in oral cancer and other tumours. An immunohistochemical study also supported a predictive value for p53 and p73 expression regarding the probability of drug response [33].

p73 IN ANTI-CANCER TREATMENT

Knowledge regarding the molecular structure, various isoforms, and their behaviour in different cellular processes and neoplasms of different sites, has helped evolve various therapeutic strategies.

The p73 gene, like p53, is upregulated at both transcriptional and post-transcriptional levels by chemotherapeutic agents and ϒ irradiation [7].

The p73 gene has an important role in modulating cellular response in radiotherapy, gamma irradiation, and chemotherapy involving various drugs like cisplatin, bleomycin etoposide, doxorubicin, camptothecin, mitoxantrone, taxol, cytosine analogs like gemcitabine, Ara-C and T-ara-C. The overall effect depends on the type of drug as well as the cellular response. Cisplatin activates and stabilises p73 but does not induce p73 mRNA. Doxorubicin and taxol, on the other hand, induce p73 proteins as well as p73 mRNA simultaneously. In contrast, ϒ irradiation promotes p73 protein activity without changing the protein levels or mRNA levels, according to one study [19]. Yet another study reported an increased p73 accumulation in response to ϒ irradiation [9].

Specific p73 Targeted Therapies in Cancer

Various pharmacological agents acting through the p73 pathway are summarised in [Table/Fig-1] [7,9,17,20,23,25,35-49].

Name of drug/ Active component	Chemical name or class	Target pathway/Mechanism of action	Biological effects/Clinical applications	Related studies
Nutlin-3a	Cis-Imidazoline analogue	MDM2 inhibitors, disrupts Mdm2/EPF1/ p73 complex, promotes apoptosis.	Induces apoptosis in doxorubicin resistant neuroblastoma cells. Induces apoptosis in neuroblastoma and colon cancer cells with mutant p53.	Ramos H et al., [35] Pierce SK and Findley HW [36] Ray RM et al., [37]
RETRA-2	Small molecule	Disrupts mutant p53-p73 inhibitory complex, prevents tumour growth.	In vitro and in vivo studies in tumour cell lines and nude mouse xenograft show delayed tumour growth.	Kravchenko JE et al., [38] Bisso A et al., [20] Vilgelm A et al., [9]
37AA peptide	Small peptide inhibitor of iASPP, Hybrid peptide derived from p53	Binds to iASPP, releases p73 leading to activation of p73 target genes.	In vitro studies show the occurrence of cell death in various tumour types.	Vilgelm A. et al., [9] Bell HS et al., [23] Slade N and Horwat A, [17]
PRIMA1, PRIMA 1 Met	Quinuclidinone	PRIMA-1 is converted to methyl quinuclidinone which complexes with mutant p53. Induces apoptosis.	Inhibits migration and colony formation in multiple myeloma cells. Synergistic action with doxorubicin was observed in multiple myeloma cell culture. Tumour growth delayed with enhanced survival in mouse model studies with multiple myeloma.	Zhang S et al., [39] Ramos H et al., [35] Saha MN et al., [40]
NSC 59984	Lead compound	Salvages wild type p53 signalling, through p73 activation and promotes mutant p53 degradation.	Promotes apoptotic cell death in colon cancer cell lines, inhibits the tumour growth in colon tumour xenograft.	Zhang S et al., [41]
Prodigiosin	Heterocyclic tripyrrole compound, bacterial secondary metabolite	Disrupts mutant p53-p73 binding complex.	Induces apoptosis and cell cycle arrest in mutant p53 and null p53 human colon cancer cell lines. Restriction of colonosphere formation in colorectal cancer stem cells.	Hong B et al., [42] Prabhu W et al., [43]
Metformin	1,1 dimethyl biguanide hydrochloride	mTOR inhibitor p73 stabilisation through AMPK activation, which leads to apoptosis. Metformin is capable of promoting cellular autophagy by inducing p73 protein expression.	Tumour suppression was observed in various cancers including breast, liver, lungs, and haematological cancers.	Slade N and Horwat A, [17] Yi Y et al., [44]
Forodesine	Transitional state analogue inhibitor of purine nucleoside phosphorylase	Upregulates transcription of p73	Induces mitochondrial apoptosis in CLL cell lines via p73 and Bcl-2-like Protein (BIM).	Bisso A et al., [20]
MLN 8054	Aurora kinase inhibitors	Upregulates TAp73β levels, aiding apoptosis	Promotion of apoptosis in p53 deficient cancer cell lines.	Bisso A et al., [20]
Retinoids	Vitamin A derivatives	Regulate target gene expression including p73, enhance TAp73 levels	Reverses potentially malignant lesions of oral cavity and head and neck malignancies. Retinoic acid is considered a standard treatment for neuroblastoma.	Rufini A et al., [7] Wagner LM and Danks MK, [45]
Rapamycin	mTOR inhibitor	Upregulates TAp73 expression	Synergistic effect with cisplatin in breast cancer	Wong SW et al., [46]
Celecoxib	Cox-2 inhibitors	Increases TAp73 β levels	Promotes cell death and apoptosis in p53 deficient cell lines	Slade N and Howart A, [17] Lau LM et al., [47]
Cisplatin	Alkylating agent Platinum coordination complex	p73 phosphorylation by SH2 binding domain of c-abl, c-Jun N- terminal kinase activation, which binds and phosphorylates p73 alpha at several sites	p73 mediated apoptosis modulates chemoresistance in ovarian cancers, triple-negative breast cancer. Synergistic role for p73 with cisplatin in nonfunctional-p53 Hela cells.	Al-Bahlani S et al., [25] Leong CO et al., [48] Kim KC et al., [49]

[Table/Fig-1]: Summarising various p73 related treatment modalities for cancer [7,9,17,20,23,25,35-49].
CLL: Chronic lymphocytic leukaemia

Brief Description of p73-related Pharmacological Agents in the Treatment of Various Cancers

1. **Nutlins:** Nutlins are non-peptide, small molecule, imidazoline compounds that belong to the family of MDM2 inhibitors. Nutlin-3 upregulates TAp73 expression via the E2F pathway, inducing apoptosis. These bind to the p53 pockets of MDM2 and increase p53 activity with nanomolar potency. Nutlins, along with Reactivating p53 and Inducing Tumour Apoptosis (RITA) are capable of displacing the MDM2/EPF1/p73 complex, promoting apoptosis. It may be used in combination with doxorubicin [7,20,50].
2. **Reactivation of Transcriptional Reporter Activity (RETRA) molecules:** RETRA are small molecules that are capable of displacement of p73 from mutant p53/p73 complex, thereby increasing p73 levels and resulting in growth arrest of the tumour cells. It acts selectively in mutant p53-p73 expressing cells [20,51].
3. **PRIMA:** PRIMA 1 (AR-246) gets converted to reactive electrophile Methylene Quinuclidine (MQ) and binds at the core domain of mutant p53, leading to refolding of mutant-p53 and restoration of wild-type p53 functions and further interactions with p73 [20,39].
4. **Metformin:** Metformin targets Adenosine Monophosphate-activated Protein Kinase (AMPK), augments β-oxidation, and obstructs oxidative phosphorylation, ultimately increasing TAp73β levels [20,51].
5. **Prodigiosin:** It acts by direct p73 activation bypassing mutant p53, producing tumour-suppressive effects [39].
6. **NSC59984:** It is also a promising molecule capable of p73 pathway activation via mutant p53 degradation induced by a ROS-ERK2-MDM2 pathway [39].
7. **Forodesine (Immucilin H):** A transition-state analog inhibitor of purine nucleoside phosphorylase, it is a highly potent orally active compound. It has been effective in CLL, as well as cutaneous T cell lymphoma and NK cell lymphoma. Forodesine has high antitumour activity in CLL without functional p53 and activates p53-independent mitochondrial apoptosis by induction of p73 and BIM [20].
8. **Aurora Kinase (AURK) inhibitors:** AURK A overexpression or amplification is a common finding in various epithelial malignancies like head and neck cancer, colon, pancreas, gastric cancers, liver, bladder, and breast cancers. AURK A

overexpression is known to cause centrosomal amplification and abnormalities during cell division. It suppresses TAp73 expression in p53 null cancer cells [20,52,53].

Small molecule MLN8054, is a novel AURK A inhibitor that induces TAp73β expression in p53-null cells leading to the downstream pro-apoptotic gene activation [20].

9. **Retinoids:** Retinoids are capable of inhibiting or reversing the oncogenic process in haematologic and other malignancies in head and neck region, breast, skin, and liver as well as premalignant lesions of the oral cavity. Retinoic acid inhibits cell growth and promotes the differentiation of cells by regulating a number of genes, including p73 [7].
10. **Mammalian Target of Rapamycin (mTOR) inhibitor:** Serine threonine kinase mTOR is associated with cell metabolism, growth, senescence, and neoplastic mechanism. mTOR inhibitors are suggested as positive regulators of p73, leading to the stabilisation and accumulation of TAp73 to induce programmed cell death in tumour cells. mTOR inhibitors are observed to be beneficial in alveolar rhabdomyosarcoma treatment [7,20].
11. **Celecoxib:** A cyclooxygenase inhibitor, it is a positive regulator of TAp73, while it selectively downregulates the expression of ΔNp73 inhibition in tumours like neuroblastoma resulting in cell cycle arrest and apoptosis. The mechanism of action is through suppression of the cyclooxygenase pathway. A study reported an association between oral cancer prevention and administration of high dosage celecoxib over 5 years [20,54].
12. **37AA peptide:** It acts by binding with iASPP and disrupting the iASPP-p73 complex, leading to cell death [23].

CONCLUSION(S)

Considerable progress has been achieved through research regarding p73 function since its discovery. To this date, novel findings are being deciphered regarding the multifaceted p73 gene and its interactions with other genes in the path of tumour development and its control. The ultimate aim in this regard, whether it is the researcher, clinician, diagnostician, or drug developer, is to make available a lasting and affordable cure for the patient, which is still elusive but may well be achieved through concerted and focused efforts.

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PARTICULARS OF CONTRIBUTORS:
1. PhD Scholar, A B Shetty Memorial Institute of Dental Sciences, Nitte University, Deralakatte, Mangalore, Karnataka, India; Reader, Department of Oral Pathology and Microbiology, KVG Dental College, Sullia, Karnataka, India.
2. Professor, Department of Oral Pathology and Microbiology, A B Shetty Memorial Institute of Dental Sciences, Nitte University, Mangalore, Karnataka, India.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:
Dr. Anitha Dayakar,
PhD Scholar, A B Shetty Memorial Institute of Dental Sciences, Nitte University,
Deralakatte, Mangalore-575018, Karnataka, India.
E-mail: anithadayakar99@gmail.com

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