Cyclin D1 and P27KIP1: The Gatekeepers of Dysplasia

Mahmoud M. Bakr1, Simon Guan2, Norman Firth3, Robert M. Love1*

1School of Dentistry and Oral Health, Griffith University, Australia
2School of Dentistry, University of Otago, New Zealand
3University of Queensland, Brisbane, Australia

ABSTRACT

There is increasing evidence suggesting that cyclins, cyclin-dependent kinases (CDKs), and cyclin-dependent kinase inhibitors (CDIs) either are themselves targets for genetic change in cancer or are disrupted secondarily by other oncogenic events. Cyclin D1 and p27KIP1 are two important regulators at the G1/S checkpoint. Cyclin D1 is an oncogene of cell cycle regulation with positive effect. Normally, cyclin D1 at G1 is constant or at a very low level and its excessive expression may be associated with the disordered proliferation of cells leading to malignant change. On the other hand, p27KIP1 is an anti-oncogene for cell cycle regulation, which functions as a negative regulator. Under the regulation of TGF-β, p27KIP1 inhibits the activity of oncogenes and controls the transition of the G1/S phase mainly by the interaction with CDK and CDK-Cyclin in order to inhibit cell proliferation and give cells opportunities to repair DNA. In addition, p27KIP1 not only acts as CDK inhibitor, but also promotes cell differentiation and induces the apoptosis of cells. In this article we review studies that have explored the effects of cyclin D1 and P27KIP1 on cancer progression and dysplasia with a specific focus on oral dysplasia and oral squamous cell carcinoma (OSCC). We also aim to shed some light on the different means of evaluating the interaction between Cyclin D1 and P27KIP1 as well as the immunohistochemical reactions associated with different forms of cyclin D1.

CELL CYCLE

The cell cycle is the series of events that are required to create two daughter cells from a progenitor cell. The cell division cycle consists of four phases, i.e. G1, S, G2, and M. A cell that is not in the cell cycle is in a quiescent state named G0. G1 is the interval before DNA replication, S is the DNA replication phase, G2 is the interval after DNA replication, M is the mitotic phase. Progression through the cell cycle is governed by a family of cyclin-dependent kinases (CDKs), the activity of which is regulated by phosphorylation, activated by binding of cyclins, and inhibited by CDK inhibitors. The orderly progression through the different phases is assured by proteins that regulate critical checkpoints. Several checkpoints have been identified such as the late G1 phase restriction point, the G1/S phase transition, and the G2/M phase transition. These checkpoints normally verify that the preceding events have been completed before progressing to the next monitor completion of DNA replication and produce signals that interrupt the cell cycle in the event of an error or damage to the genome. Abnormal functioning of checkpoints, such as incapacity in detecting damaged DNA, may play a significant role in tumour progression by permitting cells to progress through the cell cycle with damaged or abnormal DNA.
The orderly progression of cells through the cell cycle is carefully orchestrated by CDK after they bind to a group of proteins called cyclins. The cyclin/CDK complexes form heterodimers that phosphorylate many proteins involved in major cell cycle events. The activity of cyclin/CDK complexes is modulated by the phosphorylation of threonine residues on the CDK, and by interactions with cyclin dependent kinase inhibitors (CDI). The proper and timely action of all these mechanisms is the basis of normal function, and consequently, dysregulation at many levels of the cell cycle has been implicated in tumourigenesis.

**CYCLINS AND CYCLIN D1**

Two major classes of cyclins are recognized according to the phase of action and to their pattern of degradation; these are the mitotic cyclins (Cyclin A, B) and the G1 cyclins (Cyclin A, D, E). Mitotic cyclins are essential for the control of the cell cycle at the G2/M transition (mitosis). They usually accumulate steadily during G2 and are abruptly destroyed as cell exit from mitosis. On the other hand, the G1 cyclins are essential for the control of the cell cycle at the G1/S transition. For example, cyclin A/CDK2 complex is active in S phase. Cyclin D/CDK4, cyclin D/CDK6, and cyclin E/CDK2 regulate the transition from G1 to S phase.

There are three isoforms of cyclin D (Cyclin D1, D2, D3) in humans; cyclin D1 is perhaps the most studied D-type cyclin in human cancers. In mice, cyclin D1 is located on chromosome 7. Human cyclin D1 is localized at chromosome 11q13. It is a nuclear protein that has been shown to be a key regulator of G1-S phase transition, and elevated levels of cyclin D1 induce apoptosis. Cyclin D1 protein binds and activates CDK4 and CDK6, leading to phosphorylation of retinoblastoma protein (RB – a tumour suppressor protein) that results in release of transcriptional activator E2F, leading to transcription and activation of proteins associated with passage through the G1 check point and progression into the S phase.

A review article states that cyclin D1 also acts as a transcriptional modulator by regulating the activity of several transcription factors and histone deacetylase. This function is independent of the CDK4 activity. Cyclin D1 protein is unstable with a short half-life, about 24 minutes. It is degraded mainly by the 26S proteasome in an ubiquitin-dependent pathway. However, cyclin D1 is an important proto-oncogene. Overexpression of cyclin D1 leads to shortening of the G1 phase and to less dependency on exogenous mitogens, resulting in abnormal cell proliferation that in turn may favour the occurrence of additional genetic lesions.

Cyclin D1 is reported as being overexpressed or amplified in a number of primary human cancers supporting its role as an oncogene. In many tumours, genetic alterations affecting the cyclin D1 gene frequently result in overexpression of cyclin D1 protein. Several studies have shown overexpression of cyclin D1 protein is associated with at least half of all invasive breast cancers. Many studies of mantle cell lymphoma have demonstrated increased activity in cyclin D1 and it has been shown that overexpression of cyclin D1 by lymphocytes in the mantle zone impairs the capacity of these cells to exit the cell cycle and to differentiate into mature plasma cells. Studies of oesophageal cancer also showed amplification and overexpression of cyclin D1 protein in 30% of the cases. Amplification and increase expression of cyclin D1 protein have been observed in 10% of hepatocellular carcinoma. Overexpression of cyclin D1 protein has also been associated with decreased survival and worse prognosis in different types of cancer including oesophageal squamous cell carcinoma, breast carcinoma and colonic adenocarcinoma.

Furthermore, several studies have demonstrated that increased levels of cyclin D1 mRNA may be associated with decreased survival rate of patients with head and neck cancers. These observations suggest that cyclin D1 might play an important role in malignant transformation and disease progression. In addition to the above, overexpression of cyclin D1 protein may be the consequence of gene rearrangement. Therefore, amplification of this gene often appears in malignant lesions. Amplification of cyclin D1 gene has been demonstrated in 17%-55% of head and neck squamous cell carcinoma (HNSCC) in several studies. Over expression of cyclin D1 protein has also been shown in 21%-64% of HNSCC and associated with a poor prognosis, more frequent recurrence, and shorter time to recurrence. Moreover, overexpression has been shown to be associated with lymph node metastasis. Little is known of the frequency and the timing of this change in oral epithelial dysplasia. Overexpression of cyclin D1 in oral epithelial dysplasia has been reported in some studies. There are some studies on cyclin D1 degradation in vitro by a number of therapeutic agents. They showed that induction of cyclin D1 degradation might offer a useful avenue for therapeutic intervention. A summary of studies (1990-2018) on expression of cyclin D1 and CDKs in normal tissues, oral epithelial dysplasia and squamous cell carcinoma as well as other different types of cancer is shown in Table 1.

**CYCLIN DEPENDENT KINASES and P27**

Cyclin-dependent kinases (CDKs) are a family of protein kinases that are involved in regulating the cell cycle. Their activity is controlled by a complex network of regulatory subunits and phosphorylation events. In the cell, these regulatory mechanisms generate an interlinked series of CDK oscillators that trigger the events of cell division. Therefore, they may be considered as the engines that drive the events of the cell cycle. There are at least nine
different cyclin-dependent kinases in eukaryotic cells, four of which, CDK1, 2, 3, and 4, have been shown to play an important role in the regulation of the eukaryotic cell division cycle and have also been suggested in the control of gene transcription and other processes\(^{59}\).

Cyclin-dependent kinase inhibitors (CDIs) are proteins that inhibit cyclin-dependent kinase. Cell cycle progression is negatively controlled by CDIs. They are involved in the cell cycle arrest at the G1 phase. Two families of CDIs negatively regulate CDK activities and mediate cell cycle arrest following growth inhibitory stimuli\(^{51}\). The inhibitors of CDK4, also known as the INK4 family members involve p15\(^{INK4B}\), p16\(^{INK4A}\), p18\(^{INK4C}\), and p19\(^{INK4D}\). They specifically inhibit cyclin D1 associated kinases\(^{52,51}\). Members of the kinase inhibitor protein (KIP) family involve P21\(^{CIP1}\) or CDK-interacting protein 1, also known as P21\(^{WAF1}\), P21\(^{SDPI}\) or senescence DNA synthesis inhibitor 1, P27\(^{KIP1}\) or kinase inhibitor protein 1, and P57\(^{KIP2}\) or kinase inhibitor protein 2. It has been suggested that they bind and inhibit cyclin D/CDK4, cyclin E/CDK2 and cyclin A/CDK2 complexes\(^{51}\). P21 is a potent CDI. It is a major element in cell cycle control and it is mainly regulated at the transcriptional level. It binds to and inhibits the activity of cyclin E/CDK2, cyclin B/CDK1 and cyclin D/CDK4/6 complexes and thus acts as a regulator of cell cycle progression at G1/S phase\(^{54}\).

P27\(^{KIP1}\) is a cyclin-dependent kinase inhibitor and a tumour suppressor that regulates G0 to S phase transitions\(^{55}\). It has been identified as an inhibitor in cells arrested by transforming growth factor – β (TGF-β) and is regulated by growth inhibitory cytokines and by contact inhibition\(^{56-58}\). P27\(^{KIP1}\) protein is strongly expressed in non-proliferating cells and plays an important role in the regulation of both quiescence and G1 progression\(^{55}\).

P27\(^{KIP1}\)'s inhibitory activity is mainly controlled by its concentration, subcellular localization and phosphorylation status\(^{55}\). The levels and activity of p27\(^{KIP1}\) protein increase in response to a number of factors, including cell density, differentiation signals, following loss of adhesion to the extracellular matrix, and in response to growth inhibitory signaling by TGF-β or the drug lovastatin\(^{56-63}\). P27\(^{KIP1}\) is not a classic tumour suppressor like P53, but loss of p27\(^{KIP1}\) protein could result in growth to inhibitory factors, deregulation of cell proliferation, and oncogenic change\(^{64}\).

In quiescent normal epithelia of breast, prostate, ovary, lung and other sites, p27\(^{KIP1}\) protein is expressed at high level, and loss or reduction of the level of this protein may be seen in carcinomas\(^{59}\). When both a non-invasive and invasive component coexist in the tumour, loss of p27\(^{KIP1}\) protein is detected in both carcinoma in situ and invasive tumour, suggesting that events leading to deregulation of p27\(^{KIP1}\) protein may precede invasion\(^{59}\). For example, p27\(^{KIP1}\) protein is reduced in benign prostatic hypertrophy, a hyperplastic premalignant prostatic neoplasm\(^{65-67}\). Study of prostate carcinoma also showed variable degrees of reduction in p27\(^{KIP1}\) staining was frequently observed in the prostatic intraepithelial neoplasia adjacent to invasive carcinoma\(^{66}\). Studies of breast cancer also have similar results; p27\(^{KIP1}\) protein is reduced in premalignant and non-invasive malignant lesions, including ductal carcinoma in situ of the breast\(^{66-71}\). In addition, a comparison of p27\(^{KIP1}\) protein levels in primary colon carcinoma and metastatic tumour demonstrated a reduction of p27\(^{KIP1}\) staining in the metastatic tumour\(^{72,73}\). Therefore, reduction in p27\(^{KIP1}\) protein level may contribute to cancer progression in the transitions from carcinoma in situ to invasive tumour, and from localized primary tumour to metastatic tumour\(^{74}\).

Moreover, decreased levels of p27\(^{KIP1}\) protein may be related to high tumour grade and stage in human colorectal\(^{64}\), gastric\(^{75}\), breast\(^{76}\), prostate\(^{66,77}\) and other cancers. Reduction of p27\(^{KIP1}\) protein in tuumours correlates significantly with decreased survival in colorectal\(^{78}\), gastric\(^{79,80}\), breast\(^{76,81,82}\) and esophageal\(^{83}\) squamous cell carcinoma patients, among others. These studies have suggested that p27\(^{KIP1}\) plays an important role in tumour suppression. In fact, identification of the p27\(^{KIP1}\) proteolysis pathways has opened new avenues for therapeutic intervention in cancer\(^{84}\). A summary of studies (1990-2018) on expression of cyclin D1 and CDKs in normal tissues, oral epithelial dysplasia and squamous cell carcinoma as well as other different types of cancer is shown in Table 1.

**IMMUNOHISTOCHEMICAL EVALUATION OF THE INTERACTION BETWEEN CYCLIN D1 AND P27\(^{KIP1}\)**

In most studies, the immunohistochemical reactivity for cyclin D1 and p27\(^{KIP1}\) was evaluated on the basis of presence or absence of nuclear and/or cytoplasmic staining\(^{60,104,107-109}\). However, Gillett et al. excluded cyclin D1 cytoplasmic staining alone in their cases and considered it as negative\(^{110}\). It has been shown that cyclin D1 plays an important role in cell proliferation and differentiation and can be transported between nuclear and cytoplasmic compartments via nuclear pores during different phases of the cell cycle\(^{111}\).

Most of the previous studies suggest cyclin D1 is a nuclear protein\(^{112-114}\). However, results from Guan et al., 2018\(^{109}\) (strong nuclear and cytoplasmic staining) imply that for proteins such as cyclin D1, it is more accurate to apply to nuclear/cytoplasmic distribution ratio than a single cellular localization. Moreover, the capacity of the cell to direct nuclear import and export during cell cycle suggested that movement of cyclin D1 must be considered bi-directional\(^{115}\). The redistribution of cyclin D1 protein correlates with its phosphorylation on Thr-286 by...
Table 1: Illustrates a summary of studies (1990-2018) that investigated the expression of cyclin D1 and CDKs in normal tissues, oral epithelial dysplasia and squamous cell carcinoma as well as other different types of cancer.

<table>
<thead>
<tr>
<th>Bibliographic citation</th>
<th>Study type</th>
<th>Number of patients</th>
<th>Case/setting</th>
<th>Marker protein</th>
<th>Effect size</th>
</tr>
</thead>
<tbody>
<tr>
<td>(Michalides et al., 1995)</td>
<td>Retrospective case-control pathology slides</td>
<td>47 Head and Neck Squamous Cell Carcinomas</td>
<td>Random selection of cases. Single centre study. Netherlands</td>
<td>Cyclin D1</td>
<td>Overexpression of cyclin D1 associated with poor prognosis and a shortened overall survival of these patients (P=0.0095).</td>
</tr>
<tr>
<td>(Akervall et al., 1997)</td>
<td>Retrospective case-control pathology slides</td>
<td>75 Head and Neck Squamous Cell Carcinomas</td>
<td>Random selection. Matched for site. Dual centre. Sweden and Netherlands</td>
<td>Cyclin D1</td>
<td>Patients with tumors strongly positive for Cyclin D1 had poorer survival (P=0.047).</td>
</tr>
<tr>
<td>(Xu et al., 1998)</td>
<td>Retrospective case-control pathology slides</td>
<td>34 Oral Squamous Cell Carcinomas 2 Oral Epithelial Dysplasia</td>
<td>Random selection. Matched for site and Grade. Dual centre. Texas and Argentina</td>
<td>P53, Cyclin D1, Rb and H-ras</td>
<td>High frequency (41%) of Cyclin D1 Overexpression</td>
</tr>
<tr>
<td>(Castle et al., 1999)</td>
<td>Retrospective case-control pathology slides</td>
<td>25 Oral Dysplasia 47 Oral Squamous Cell Carcinomas</td>
<td>Random selection Matched for site, grade, age, gender. Single centre. Florida.</td>
<td>P53 and Cyclin D1</td>
<td>Overexpression of Cyclin D1 was not significant different between 2 age groups studies either for dysplasia or carcinomas.</td>
</tr>
<tr>
<td>(Bova et al., 1999)</td>
<td>Retrospective case-control pathology slides</td>
<td>147 Carcinoma of the Anterior Tongue</td>
<td>Random selection Matched for site, grade, age, gender. Single centre. Australia.</td>
<td>Cyclin D1 and P16INK4A</td>
<td>Overexpression of Cyclin D1 occurred in 68% of tumors and was associated with increased lymph node stage (P=0.014), increased tumor grade (P=0.003), and reduced disease-free (P=0.006) and overall (P=0.01).</td>
</tr>
<tr>
<td>(Lam et al., 2000)</td>
<td>Retrospective case-control pathology slides</td>
<td>56 Oral Squamous Cell Carcinomas</td>
<td>Random selection Matched for site, grade, age, gender. Single centre. Hong Kong.</td>
<td>P53 and Cyclin D1</td>
<td>Cyclin D1 expression was found in 63% of Oral Squamous Cell Carcinomas and was more frequently positive in high-grade lesions (P=0.019).</td>
</tr>
<tr>
<td>(Nakahara et al., 2000)</td>
<td>Retrospective case-control pathology slides</td>
<td>78 Oral Squamous Cell Carcinomas 46 Leukoplakia 20 Normal mucosa</td>
<td>Random selection Matched for site, grade, age, gender. Single centre. Japan.</td>
<td>Cyclin D1 and P16INK4A</td>
<td>The Overexpression of Cyclin D1 was not Observed in normal mucosa and was observed in 35.9% of Squamous Cell Carcinomas</td>
</tr>
<tr>
<td>(Mineta et al., 2000)</td>
<td>Retrospective case-control pathology slides</td>
<td>94 Tongue Squamous Cell Carcinomas</td>
<td>Random selection Matched for Age, Gender, Smoking Alcohol and grade. Dual centre. Japan and Sweden.</td>
<td>Cyclin D1</td>
<td>19% of patients showed Cyclin D1 overexpression. The 5- year survival rate of high Cyclin expressors was 39% (P=0.04).</td>
</tr>
<tr>
<td>(Rousseau et al., 2001)</td>
<td>Retrospective case-control pathology slides</td>
<td>20 Normal Mucosa, 22 Mild Epithelial Dysplasia, 20 Moderate Epithelial Dysplasia, 17 Severe Epithelial Dysplasia and 25 Oral Squamous Cell Carcinoma</td>
<td>Random selection Matched for grade. Dual centre. Canada and USA.</td>
<td>Cyclin D1</td>
<td>Overexpression of Cyclin D1 was identified in 29% of mild, 47% moderate, 29% of severe Oral epithelial dysplasia. There were statistically significant correlations identified between gene and protein levels in all categories of disease.</td>
</tr>
<tr>
<td>(Ronaldo et al., 2001)</td>
<td>Retrospective case-control pathology slides</td>
<td>112 Carcinoma of the Anterior Tongue</td>
<td>Random selection. Matched for site, grade, age, gender. Single centre. Australia.</td>
<td>Cyclin D1</td>
<td>Overexpression of cyclin D1 (65% of the cases) associated with poor prognosis.</td>
</tr>
<tr>
<td>(Sathyan et al., 2006)</td>
<td>Retrospective case-control pathology slides</td>
<td>147 Buccal Squamous Cell Carcinoma and 94 Tongue Squamous Cell Carcinoma</td>
<td>Random selection. Matched for site, grade, age, gender. Single centre. India</td>
<td>P53, Rb, P16, Cyclin D1, CDK4 and PCNA</td>
<td>Among the biological markers, the Overexpression of CyclinD1 (P=0.007) showed significant association with shorter disease-free survival in these cases.</td>
</tr>
<tr>
<td>Study</td>
<td>Design</td>
<td>Control Group</td>
<td>Cases/Slides</td>
<td>Matching Criteria</td>
<td>Cyclins Assayed</td>
</tr>
<tr>
<td>-------------------------------</td>
<td>--------------------</td>
<td>--------------------------------</td>
<td>--------------</td>
<td>-------------------</td>
<td>----------------</td>
</tr>
<tr>
<td>Kövesi &amp; Szende, 2006</td>
<td>Retrospective</td>
<td>18 Oral Leukoplakia</td>
<td>90 Retrospective case control pathology slides</td>
<td>Smoking, alcohol, grade, age and gender. Single centre. Hungary</td>
<td>Cyclin D1, P27KIP1, and P63</td>
</tr>
<tr>
<td>Huang et al., 2012</td>
<td>Retrospective</td>
<td>264 Oral Squamous Cell Carcinoma</td>
<td>28 Mild Epithelial Dysplasia, 28 Mild Moderate Epithelial Dysplasia, and 28 Moderate Epithelial Dysplasia</td>
<td>Random selection. Single Centre, Taiwan</td>
<td>Cyclin D1</td>
</tr>
<tr>
<td>Uma et al., 2012</td>
<td>Retrospective</td>
<td>20 Oral Squamous Cell Carcinoma and 10 Normal Mucosa</td>
<td>20 Oral Squamous Cell Carcinoma and 10 Normal Mucosa</td>
<td>Random selection. Single Centre, India</td>
<td>P53 and Cyclin D1</td>
</tr>
<tr>
<td>Liu et al., 2013</td>
<td>Retrospective</td>
<td>130 Primary Nasopharyngeal Carcinoma</td>
<td>130 Primary Nasopharyngeal Carcinoma</td>
<td>Random selection. Single Centre, China</td>
<td>P27KIP1</td>
</tr>
<tr>
<td>Li et al., 2014</td>
<td>Meta-analysis</td>
<td>4150 cases of colorectal cancer from 22 studies</td>
<td>4150 cases of colorectal cancer from 22 studies</td>
<td>Comprehensive literature search for relevant studies published was performed using PubMed, EMBASE, and ISI Web of Science.</td>
<td>Cyclin D1</td>
</tr>
<tr>
<td>Pereira et al., 2014</td>
<td>Retrospective</td>
<td>85 Prostate Carcinoma 10 Normal prostate tissue</td>
<td>85 Prostate Carcinoma 10 Normal prostate tissue</td>
<td>Random selection. Single Centre, Brazil.</td>
<td>Cyclin D1</td>
</tr>
<tr>
<td>Ravikumar and Ananthamurthy, 2014</td>
<td>Retrospective</td>
<td>35 Ductal Carcinoma of the breast</td>
<td>35 Ductal Carcinoma of the breast</td>
<td>Random selection. Single Centre, India.</td>
<td>Cyclin D1</td>
</tr>
<tr>
<td>Moreno-Galido et al., 2014</td>
<td>Retrospective</td>
<td>41 Primary Laryngeal / Hypopharyngeal Squamous Cell Carcinoma</td>
<td>41 Primary Laryngeal / Hypopharyngeal Squamous Cell Carcinoma</td>
<td>Random selection. Matched for induction chemotherapy treatment. Single Centre, Spain.</td>
<td>EGFR, p53, Cyclin D1, p16, p21, p27KIP1, p-AKT, HIF-1a, Caspase 3 and BCL2</td>
</tr>
</tbody>
</table>
(Malgaonkar et al., 2016) Retrospective case control pathology slides 19 Pleomorphic Adenoma, 8 Mucoepidermoid Carcinoma and 7 Adenoid Cystic Carcinoma Random selection. Single Centre, Saudi Arabia. P27<sup>KIP1</sup> 52.6% of pleomorphic adenoma cases, 25% of mucoepidermoid carcinoma cases and only 14.2% of adenoid cystic carcinoma cases showed strong expression suggesting variable p27<sup>KIP1</sup> expression in both malignant neoplasms.

(Khabaz et al., 2016) Retrospective case control pathology slides 124 Bladder Cancer and 24 Normal Bladder Tissue Random selection. Single Centre, Saudi Arabia. Cyclin D1 Strong cyclin D1 immunohistochemical staining has been significantly linked with low grades (P=0.001), low stages (P=0.005). Cyclin D1 maybe a valuable tissue biomarker for presaging grade, stage, and poor prognosis in bladder cancer.

(Dhingra et al., 2017) Retrospective case control pathology slides 48 Head and Neck Squamous Cell Carcinoma (HNSCC) Random selection. Single Centre, India. Cyclin D1 A significant association was seen between Cyclin D1 expression with tumour stage and with lymph node metastasis but not with grade.

(Barić et al., 2017) Retrospective case control pathology slides 70 Papillary Thyroid Microcarcinoma Random selection. Single Centre, Croatia. Cyclin D1, P27<sup>KIP1</sup> and RET Cyclin D1 and RET expression is not crucial for the development of metastases in lymph nodes. In contrast, p27<sup>KIP1</sup> expression is significantly associated with lymph node metastasis.

(Patel et al., 2017) Retrospective case control pathology slides 30 Oral Squamous Cell Carcinoma and 30 Leukoplakia Random selection. Single Centre, India. Cyclin D1 and P63 The overall expression of cyclin D1 and p63 correlated with tumor differentiation, poor histological grades, from well-differentiated to poorly-differentiated SCC and the severity of leukoplakia.

(Guan et al., 2018) Retrospective case control pathology slides 10 Normal Mucosa, 12 Mild to Moderate Epithelial Dysplasia, and 11 Oral Squamous Cell Carcinoma Random selection. Single Centre, Dunedin, New Zealand Cyclin D1 and P27<sup>KIP1</sup> A significant increase in expression of cyclin D1 and a decrease in expression of p27<sup>KIP1</sup> proteins were observed in oral epithelial dysplasia and less differentiated OSCC. The characteristic expression of both cyclin D1 and p27<sup>KIP1</sup> correlate with the grade of oral epithelial dysplasia and degree of OSCC differentiation.

(Ramos-Garcia et al., 2018) Retrospective case control pathology slides 69 Oral Squamous Cell Carcinoma Random selection. Single Centre, Spain. Cyclin D1 and Ki-67 Cytoplasmic cyclin D1 expression was associated with advanced tumor stage, poor differentiation, elevated Ki-67 expression, and the presence of invasive cell morphology, indicators of a poor prognosis.

(Filipits et al., 2018) Retrospective case control pathology slides 862 Early Breast Cancer Random selection. Matched for Trastuzumab treatment. Multiple Centres. Austria, Canada, Germany, Australia and Greece. TOP2A, Ki67, Cyclin D1 and P27<sup>KIP1</sup> A significant interaction was detected between p27<sup>KIP1</sup> and treatment (p=0.0049). Trastuzumab effect was significant in the p27<sup>KIP1</sup> low subgroup (p<0.001). No trastuzumab effect was observed in the p27<sup>KIP1</sup> high (p=0.89).
glycogen synthase kinase-3 β (GSK-3β). On the other hand, GSK-3β might phosphorylate cyclin D1 protein in the cytoplasm, preventing its association with protein required for nuclear import. However, overexpression of canonical cyclin D1 protein alone is not sufficient to induce cancer transformation.

It has been suggested that cyclin D1 is a multifaceted regulator that exists in two distinct isoforms, cyclin D1a and D1b. Cyclin D1a protein is rarely overexpressed in cancers, because it can be phosphorylated by GSK-3β. However, cyclin D1b protein retains the ability to bind to and active CDK4, but the cyclin D1b protein is refractory to GSK-3β nuclear export, and is thus, constitutively nuclear. Expression of cyclin D1b mutant, cyclin D1b protein, which is not subject to GSK-3β-dependent redistribution and remains in the nucleus during the cell cycle suggesting that deregulation of cyclin D1 nuclear export results in increased cyclin D1 oncogenic activity. In addition, p27KIP1 protein levels are reduced in cyclin D1b-expressing cells and that a non-phosphorylatable cyclin D1b protein may be the oncogenic mechanism that allows overexpression of cyclin D1 protein alone to induce cancer transformation.

Furthermore, it has been suggested that cyclin D1a may merely be a marker of increased proliferation in cancer cells, but it may represent a key driver of oncogenesis in those who express cyclin D1a as well. Therefore, cyclin D1b may be the oncogenic mechanism that allows overexpression of cyclin D1a to function in an oncogenic capacity. These studies and observations suggest that overexpression of cyclin D1b and/or cyclin D1a can lead to strong nuclear cyclin D1 staining in OSCC cells. Finally, cyclin D1a retains the ability to bind to GSK-3β and therefore it can be transported from nucleus to cytoplasm. However, with a reduction in p27KIP1 protein expression, cyclin D1a cannot be degraded in OSCC cells, therefore a strong cytoplasmic staining was observed in this study. Perhaps these could explain why we could observe strong nuclear and cytoplasmic cyclin D1 staining in OSCC specimen.

CONCLUSION

The mechanism of action of cyclins and cyclin-dependent kinases were discussed in this article. Cyclin D1 and p27KIP1 have been identified as important regulators of cell cycle. The consensus amongst different research studies in literature is that cyclin D1 protein overexpression is associated with epithelial dysplasia and tumor progression. On the contrary, p27KIP1 protein expression is decreased in cases with epithelial dysplasia and less differentiated grades of tumors. Therefore, both markers could be used as predictors for tumors’ aggressiveness, prognosis, response to treatment and survival rates.

CONFlicts of interest

None declared

References


