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DOI: 10.14670/HH-11-981
Article type: REVIEW
Accepted: 2018-03-06
Epub ahead of print: 2018-03-06

This article has been peer reviewed and published immediately upon acceptance. Articles in “Histology and Histopathology” are listed in Pubmed. Pre-print author’s version
Review article

The Expression of CD Markers in Solid Tumors: Significance in Metastasis and Prognostic Value

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Running title: Significance of CD marker in solid tumors
Text word count: 2556
Abstract word count: 186
Tables: 3
Figures: 0

Highlights

← The importance of epigenetic changes in the expression of CD markers on tumor cells
← The role of CD markers in the proliferation and metastasis of tumor cells
← The role of CD markers in induction and mobilization of tumor cells to different organs of the body
← The prognostic importance of CD markers in response to various treatments
Abstract

Objective

The clusters of differentiations (CDs) are among the surface markers expressed on different cells in the body, which are involved in the communication of cells with each other and the induction of signaling. Moreover, the evaluation of the ectopic expression of these markers in solid tumors has led to the detection of disease in early stages. In this paper, we have examined the effect of CD markers expression on the function of cancer cells, as well as their importance as the diagnostic and prognostic factors for monitoring the progression of solid tumors.

Materials and methods

Relevant literature was identified by a PubMed search (1988-2017) of English language papers using the terms “CD markers”, “diagnostic”, “prognostic”, “predictive marker” and “solid tumors.”

Discussion

Finally, it can be stated that the evaluation of CDs is not only of diagnostic value at disease onset, but these markers can be used as prognostic and predictive markers to contribute to the treatment of disease and predict its relapse.

Conclusion

Monitoring of tumors progression through CDs expressed on circulating tumor cells could be a new diagnostic and prognostic factor in the future.

Keywords: CD Marker, Solid Tumors, Diagnosis, Prognosis, Predictive
**Introduction**

The most important factor increasing the risk of death in cancer patients is the lack of early diagnosis of initial stages of metastasis or relapse. Furthermore, the failure to identify a specific tumor marker for the detection and monitoring of solid tumors could lead to the invisibility of tumor in the early stages (1). Tumor markers are factors in plasma or even in tissues that indicate the presence of tumor cells. These markers should be capable of detecting tumor cells much earlier than common diagnostic methods, and on the other hand, the response of cancer cells to treatments can be evaluated by measuring tumor marker levels in the body (2). Therefore, to diagnose tumors in the early stages, it is essential to identify a suitable marker for the evaluation and treatment of patients.

CD markers seems to be better diagnosis and prognosis markers in compare to other markers in cancer. CDs comprise a series of surface antigens (Ags) of different types (3), which are predominantly expressed on leukocytes and other cells and contribute to various processes, including cell-cell communication and the elicitation of immune responses against foreign agents. In addition, it has recently been noted that these markers are also expressed on tumors and are involved in molecular processes (e.g. self-renewal) in these cells (4, 5). Moreover, since the expression of CD markers is associated with physiologic changes, their expression on tumor cells is different from normal cells. Hence, the evaluation of these markers as the factors indicative of physiological and pathological changes occurring in the body can be effective in tracing and detecting the tumor cells in the body (2). Several studies have been conducted on the expression of different CD markers on tumor cells and their involvement in molecular mechanisms. However, there have been few studies to date on the assessment of these CD markers as prognostic and predictive markers. Through the identification of biological functions of these markers on cancer cells, they are likely to be used as prognostic factors, as well as for the development of appropriate treatment protocols according to their molecular mechanisms. In this paper, we assessed the expression importance of a number of CD markers as effective factors for disease diagnosis and prognosis of solid tumor cells [e.g. colorectal cancer...
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Epigenetic changes regulate CD markers expression

The expression of some CD markers on tumor cells due to epigenetic changes results in the aggressiveness and resistance of these cells to therapeutic agents (6). Epigenetic processes involve a series of changes without substitution in the DNA sequence, which lead to the expression activation or silencing of genes. DNA methylation and histone modification are major epigenetic changes that lead to the formation of a tumor phenotype and the proliferation of tumor (7, 8). The hyper- and hypo-methylation of CpG islands in the promoters of genes triggers the silencing and activation of gene expression, respectively. Hypomethylation of these regions in the promoter of many CD marker genes (including CD133) results in the increased expression of these markers on tumors, which eventually increases tumor survival. In addition, hypomethylation in the CpG islands has been shown to increase the transcription factor binding to these regions, which enhances the expression of CD markers (9). Reduced activity of DNMT1 and DNMT3B enzymes, which methylate CpG islands in gene promoters, is another mechanism that leads to the expression of CD markers on tumor cells. The expression of CD133 has been shown to increase as a result of DNMT1 and DNMT3B inhibition by transformation growth factor-β (TGF-β) (10).

Enhancer of zester homolog 2 (EZH2) is a component of polycom repressive catalytic complex (PRC2). It has methyltransferase activity, and through the methylation of histone H3, suppresses the expression of tumor suppressor genes and CD markers, which results in solid tumors (such as in glioblastoma) (11). In tumor cells, EZH2 suppresses and increases the expression of miR-218 and CD133 on tumor cells, respectively. Besides miR-218, there are other miRs suppressing the expression of CD markers on tumor cells, which in fact play the role of tumor suppressors by binding the 3′-untranslated region (3′-UTR) of CD marker mRNAs and prevent their expression (12). However, in methylated tumor cells, binding of inhibitory factors to the promoter regions of these miRs reduces their expression and in turn increases the expression of their target genes (CD markers) (Table 1). Therefore, the CD markers expressed on tumor
cells could be used as diagnostic and prognostic factors and might also detect the signaling pathways inactivating the tumor suppressors.

Table 1. Effect of epigenetic changes on the expression of CD markers.

<table>
<thead>
<tr>
<th>MiRNA</th>
<th>Expression</th>
<th>Disease</th>
<th>CD marker</th>
<th>Mechanisms</th>
<th>Prognosis</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>miR-199-5b</td>
<td>Down</td>
<td>Medulloblastoma</td>
<td>CD15</td>
<td>- Methylation in the CpG islands at the upstream promotor</td>
<td>Poor</td>
<td>(13)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>- Binding Hes1 factor to promoter</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>- miR-199-5b binding site in the 3'-UTR of CD15</td>
<td></td>
<td></td>
</tr>
<tr>
<td>miR-199a</td>
<td>Down</td>
<td>Ovarian cancer</td>
<td>CD44</td>
<td>_ Binding site in the 3'-UTR</td>
<td>Poor</td>
<td>(14)</td>
</tr>
<tr>
<td>miR-193a</td>
<td>Down</td>
<td>Ovarian cancer</td>
<td>CD117</td>
<td>- Binding of E2F6 to polycomb complex of miR-193a</td>
<td>Poor</td>
<td>(15)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>- Binding site in the 3'-UTR of CD117</td>
<td></td>
<td></td>
</tr>
<tr>
<td>miR-370</td>
<td>Down</td>
<td>Lung cancer</td>
<td>CD105</td>
<td>- Binding TRAF4 in the 3'-UTR of miR-370</td>
<td>Poor</td>
<td>(16, 7)</td>
</tr>
<tr>
<td>miR-142-3p</td>
<td>Down</td>
<td>Hepatic and colon cancer</td>
<td>CD133</td>
<td>- Binding OCT4 to promoter of miR-142-3p</td>
<td>Poor</td>
<td>(18, 19)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>- Binding site in the 3'-UTR of CD133</td>
<td></td>
<td></td>
</tr>
<tr>
<td>miR-34a</td>
<td>Down</td>
<td>Prostate cancer</td>
<td>CD44</td>
<td>Expression of miR-34a is repressed by p53</td>
<td>Poor</td>
<td>(20)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>- Binding site in the 3'-UTR of CD44</td>
<td></td>
<td></td>
</tr>
<tr>
<td>miR-224</td>
<td>Down</td>
<td>Pancreatic ductal adenocarcinomas</td>
<td>CD40</td>
<td>- Binding site in the 3'-UTR of CD40</td>
<td>Poor</td>
<td>(21)</td>
</tr>
<tr>
<td>miR-486</td>
<td>Down</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>miR-200</td>
<td>Down</td>
<td>Lung cancer</td>
<td>CD274</td>
<td>- Binding site in the 3'-UTR of CD274</td>
<td>Poor</td>
<td>(22)</td>
</tr>
</tbody>
</table>

Abbreviation: 3'-UTR: 3'-untranslated region; TRAF4: TNF receptor-associated factor 4.

**CD markers in solid tumors**

**Liver cancer:** Liver cancer is one of the most lethal and invasive malignancies that causes a large number of deaths every year worldwide (23). Liver cancer is divided into different types based on the affected liver cells, and HCC is the most common form of this cancer (24). Due to the lack of a specific factor, as well as delayed diagnosis of liver cancer, the metastasis and relapse rates of HCC cells are increased in patients, which lead to a reduction in the survival of patients (25, 26). Today, it is known that cancer stem cells (CSCs) are sparsely distributed among HCC tumor cells, leading to disease relapse and lack of response to treatment. Several CD markers are expressed on CSCs, including CD13 and CD24 (27). However, CD133, CD44, and CD90 have been indicated as the most common markers differentiating CSCs from other HCCs. CD133 is a transmembrane glycoprotein that plays an important role in the self-renewal of HCCS, and the tumorigenic potential of CD133+ CSCs is higher than CD133−
ones (28). CD44 is another surface marker of CSCs, which causes HCC metastasis through the induction of Epithelial-Mesenchymal Transition (EMT), so that the knockdown of CD44 prevents the metastasis of HCCs (29). Moreover, considering the involvement of Wnt/β-catenin pathway in the induction of EMT process and self-renewal of tumor cells, this pathway probably leads to the proliferation of tumor cells via induction of CD44 expression (30). CD34 and CD105 (endoglin) are other markers used to evaluate the cellular metastasis of HCC cells, and the angiogenesis rate in cancers such as HCCs can be evaluated as microvascular density (MVD) by these markers. However, recent studies have shown that CD105 is a specific factor to assess angiogenesis and disease progression in HCC due to its expression in new vessels and its distinction between immature and mature vessels (31, 32). CD151 is another marker used to evaluate MVD. Considering that the increased CD151 expression is associated with increased release of matrix metalloproteinase 9 (MMP9), the expression of this marker in HCC patients is associated with an unfavorable prognosis (Table 2) (33).

Macrophages are divided into two types of M1 (CD68+) and M2 (CD206+) based on function and expression of a series of markers on their surface. The M1 type plays an important role in suppressing the growth of cancers by stimulating immune responses, while the M2 type increases tumor cell proliferation by suppressing immune responses (34). Accordingly, it has been shown that the increased expression of CD68 and CD206 markers could be used to evaluate prognosis in patients, so that the overexpression of CD68 and CD206 has been associated with favorable and poor prognosis, respectively (Table 2). In addition to prognostic value, the CDs expressed on HCC cells can be used to analyze the response rate of patients to treatments. So, it was investigated that cancer cells which express CD279 and CD152 suppress immune responses so that target them can induce immune responses against cancer cells (Table 3).
Table 2. The expression of CD makers in cancers and their diagnostic and prognostic value.

<table>
<thead>
<tr>
<th>Type of cancer</th>
<th>Commonly identified CD markerS</th>
<th>Prognostic CD markerS</th>
<th>Diagnostic CD markerS</th>
<th>Type of sample</th>
<th>Type of diagnostic technique</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>liver</td>
<td>CD133, CD44, CD90, CD105, CD34, CD151, CD206, CD68</td>
<td>CD151, CD68, CD206</td>
<td>--------</td>
<td>Tissue, Cell line</td>
<td>IHC, Flow cytometry, RT-PCR</td>
<td>(35-38)</td>
</tr>
<tr>
<td>colorectal</td>
<td>CD66, CD110, CD133, CD44, CD2, CD89, CD200</td>
<td>CD110, CD133, CD44, CD200</td>
<td>CD66</td>
<td>Tissue, Cell line, peripheral blood</td>
<td>IHC, Flow cytometry, RT-PCR</td>
<td>(39-42)</td>
</tr>
<tr>
<td>lung</td>
<td>CD117, CD176, CD166, CD88, CD103CD66</td>
<td>CD88, CD103,</td>
<td>CD66</td>
<td>Tissue, Cell line, peripheral blood</td>
<td>IHC, Flow cytometry, RT-PCR</td>
<td>(43-48)</td>
</tr>
<tr>
<td>Breast</td>
<td>CD44, CD24, CD133, CD14, CD200, CD4 CD8, CD4 CD25</td>
<td>CD44, CD133, CD14</td>
<td>--------</td>
<td>Tissue, Cell line, peripheral blood</td>
<td>IHC, Flow cytometry, ELISA</td>
<td>(49-53)</td>
</tr>
</tbody>
</table>

Abbreviation: IHC: Immunohistochemistry; RT-PCR: Reverse transcription polymerase chain reaction; ELISA: enzyme-linked immunosorbent assay.

Table 3. Expression of CD makers and their response to treatment.

<table>
<thead>
<tr>
<th>Cancer</th>
<th>Predictive CD markers</th>
<th>Function</th>
<th>Type of treatment</th>
<th>Response</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Liver</td>
<td>CD133</td>
<td>Activation of ERK pathway</td>
<td>Sorafenib</td>
<td>Ineffective</td>
<td>(54-56)</td>
</tr>
<tr>
<td></td>
<td>CD279</td>
<td>Phosphorylation of ERK</td>
<td>Sorafenib+anti-PD-1</td>
<td>Good</td>
<td></td>
</tr>
<tr>
<td></td>
<td>CD152</td>
<td>Suppression of T-cells</td>
<td>Tremelimumab+TACE</td>
<td>Good</td>
<td></td>
</tr>
<tr>
<td>Colorectal</td>
<td>CD66</td>
<td>Apoptosis</td>
<td>Chemotherapy-5-FU</td>
<td>Good</td>
<td></td>
</tr>
<tr>
<td></td>
<td>CD47</td>
<td>Mediated phagocytosis by macrophage</td>
<td>mAb</td>
<td>Good</td>
<td>(57-60)</td>
</tr>
<tr>
<td></td>
<td>CD147</td>
<td>Apoptosis and arrested cell cycle</td>
<td>5-FU</td>
<td>Good</td>
<td></td>
</tr>
<tr>
<td>Lung</td>
<td>CD133</td>
<td>Expression ABCG2 and CXCR4</td>
<td>Cisplatin</td>
<td>Ineffective</td>
<td>(61-63)</td>
</tr>
<tr>
<td></td>
<td>CD317</td>
<td>Induced ADCC and CDC</td>
<td>mAb</td>
<td>Good</td>
<td></td>
</tr>
<tr>
<td></td>
<td>CD66</td>
<td>Sensitive to anoikis</td>
<td>8F5m Ab+Paclitaxel</td>
<td>Good</td>
<td></td>
</tr>
<tr>
<td>Breast</td>
<td>CD166</td>
<td></td>
<td>Taxane-free adjuvant chemotherapy</td>
<td>Good</td>
<td>(64-66)</td>
</tr>
<tr>
<td></td>
<td>HER2 with CD3</td>
<td></td>
<td>Specific Ab</td>
<td>Good</td>
<td></td>
</tr>
</tbody>
</table>
Colorectal cancer: This malignancy is ranked third and second in terms of prevalence in males and females, respectively. CRC cells are metastable to liver in 70-80% of cases (67). Liver is the organ producing thrombopoietin (TPO) and CD110 as the TPO receptor is expressed by CRC cells. Hence, the chemotactic property between TPO and its receptor probably leads to the metastasis of CRC cells to the liver (39). On the other hand, CRC cells can metastasize to lymph nodes if CD133 and CXCR4 are expressed on their surface (68). However, if this is the case, the Wnt/β-catenin signaling pathway will be disrupted and CRC metastasize to the liver (41, 69). Besides, the expression of CD200 (which leads to the suppression of immune responses) is increased on CD133⁺ CD44⁺ cells relative to CRC CD133⁻ CD44⁻ cells, which is associated with the activation of Wnt/β-catenin pathway that ultimately results in the suppression of the immune system (70). Conversely, the 90K glycoprotein (a type of tumor suppressor) has a number of domains to which CD9 and CD82 are bound to inactivate the Wnt/β-catenin signaling pathway (40). Thus, the CDs expressed on CRCs can activate or suppress the Wntβ-catenin pathway according to their physiological functions and the activation of this pathway is associated with a poor prognosis.

The expression of CD66 by CRC cells is reduced in the early stages of CRC but is increased with disease progression and expansion, which is associated with resistance against anoikis (a form of programmed cell death) (42). There are long (L) and short (S) domains on the cytoplasmic side of CD66 and the L domain shows a good response to treatment. A number of other CDs are expressed on CRC cells, which are known as predictive markers. CD47 is an example of mentioned CD markers which activates the anti-dependent cell mediated cytotoxicity phenomenon (ADCC) (Table 3).

Lung cancer: Lung cancer is a malignancy with a high mortality rate in patients (71). CD133 is not only expressed on CSCs of lung cancer, but it is known as a diagnostic marker for circulatory tumor cells (CTCs) in this type of cancer, so
that the detection of CTCs in peripheral blood (PB) can indicate the metastasis of cancer cells. Therefore, CD133 could be used as a diagnostic marker for lung cancer CTCs in the early stages of metastasis (72). Therefore, targeting the CD133 may be effective in preventing the progression of cancer cells (Table 3). Other markers expressed on CSCs are listed in Table 2, among which the CD66 is expressed to a higher extent in malignant cells than in normal cells. Therefore, monitoring of CD66 in patients can lead to disease diagnosis (43). On the other hand, CD66 targeting is also associated with increased activity of the immune system and improved survival of patients (Table 3).

In normal conditions, C5a interaction with its receptor (CD88) leads to inflammation via activation of immune cells (44). Recently, however, it has been shown that such an interaction in lung cancer leads to the decreased expression of CD62E (E-selectin) and eventually an increase in EMT and metastasis. Furthermore, the overexpression of CD9 can increase the apoptosis of tumor cells due to calretinin expression that ultimately reduces metastasis (73, 74). It has been shown that CD317 (H.M1.24) targeting can lead to increased complement-mediated tumor lysis (Table 3). CD103 is another marker expressed on immune cells such as CD8+ T-cells and tumor infiltrating lymphocytes (TILs) but CD103 expression is decreased in patients with lung cancer, leading to increased proliferation of cancer cells. Therefore, the reduction in CD103 expression may be associated with an unfavorable prognosis (45). Finally, we can say that the evaluation of immune markers as prognostic factors may prove helpful in monitoring the progression of tumor cells.

Breast cancer: This is one of the most common cancers among women. Human epidermal growth factor receptor (HER2) is an important marker in breast cancer, which is used as a prognostic and diagnostic factor (75). CD24 and CD44 are two markers expressed on breast CSCs. However, the expression of CD24 and CD44 in HER2 positive CSC cells and CER HER2 negative cells has been indicated, respectively. Moreover, HER2 positive CSCs are more aggressive than HER2 negative ones probably due to CD24 expression by the former (49). Given the expression induction of hypoxia inducible factor (HIF) through the activation of PI3K/AKT pathway, HIF is likely to enhance the aggressiveness of HER2 positive CSC cells due to increased angiogenesis (76,
On the other hand, the expression of CD24 reduces the activity of MAPK signaling pathway, which reduces the proliferation of cancer cells. Moreover, it increases the apoptosis of the cancer cells by suppressing the CD44-induced NF-kB pathway (78). On the other hand, CD44⁺ALDH<sup>high</sup> breast cells can metastasize toward this organ through the interaction with factors released by lung such as osteopontin (79,80). CD133 is expressed on CSCs as well as CD44. Recent research has shown that CD133⁺ ALDH⁺ cells cause increased angiogenesis (50). CD146 is another marker expressed on CSCs, leading to the increased expression of Ki-67 (which increases cell proliferation) as well as enhancing angiogenesis as a co-receptor with VEGFR2. Therefore, the expression of CD146 is associated with metastasis and disease relapse (51).

T regulatory (Treg) and T helper (Th) cells are characterized by the expression of CD4⁺/CD25⁺ and CD4⁺/CD8⁺ markers, respectively. Th/Treg ratio plays a crucial role in the control of tumors. Recent research has indicated that the Th/Treg ratio is reduced in breast cancer patients relative to healthy people due to increasing Treg cells in advanced (IV stage) breast cancer (52). The expression of CD200 on immune cells and interaction with its receptor (CD200R) results in immune system suppression and increased proliferation of tumor cells (81). However, recent results have shown that the soluble CD200 leads to the release of inflammatory cytokines (e.g. IL-1β), preventing metastasis and relapse of tumor cells (53). It is interesting to note that in other malignancies like chronic lymphocytic leukemia (CLL), the secretion of soluble CD200 leads to the production of a number of CD markers in addition to those mentioned on tumor cells, the targeting of which may affect the course of disease improvement. Finally, targeting CD24 and CD3 additionally to HER2 would prevent the proliferation of cancer cells and stimulate the immune system against cancer cells, respectively (Table 3).

**Discussion and future perspective**

Cancer is a disease that is difficult and even impossible to control and treat due to the lack of diagnosis at disease onset. The absence of a specific or common marker to detect tumor cells is the main factor for uncontrolled progression of this disease (82). Although a variety of markers have been raised to identify and evaluate the progression of cancer cells, it seems that the identification of CD markers expressed on CTCs in PB has a higher clinical prognostic value due
to straightforward measurement and cost-effectiveness compared to other markers (72, 83). In addition to prognostic value of CD markers expression, these markers could be used as predictive markers to evaluate patients responses to chemotherapy and even radiotherapy. For example, CD133 is a marker expressed in CSCs of various cancers, the expression of which is associated with an unfavorable prognosis and shows variable responses to different therapies (Table 2 & 3). Also, given that CSCs from different solid tumors possess a series of common marker like CD133 and CD44, these markers may be not only be used to detect CSCs in different cancers but to design a common integrated therapeutic approach to target these cells (Table 3). In addition, the expression of CD44 and other CSC markers can be due to the activation of Wnt/β-catenin signaling pathway. Finally, tracing the activation of this pathway in solid tumors can be considered as a prognostic factor with respect to the markers expressed via this pathway (30).

Finally, it is understood that the evaluation of CD markers in solid tumors might be useful as a diagnostic factor in monitoring the disease development and treatment response.

Acknowledgement

We wish to thank all our colleagues in Shafa Hospital and Allied Health Sciences School, Ahvaz Jundishapur University of Medical Sciences.

Authors' Contributions

Najmaldin Saki conceived the manuscript and revised it. Hadi Rezaeeyan, Saeid Shahrabi, Trevor D. McKee and Najmaldin Saki wrote the manuscript.

Ethical approval

This article does not contain any studies with human participants or animals performed by any of the authors.

Conflict of Interest

The authors declare no conflict of interest.
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