Survivin – prognostic tumor biomarker in human neoplasms – review

Surwiwina – czynnik prognostyczny w nowotworach u ludzi – praca poglądowa

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Abstract

Accurate diagnosis and proper monitoring of cancer patients remain important obstacles for successful cancer treatment. The search for cancer biomarkers is carried out in order to quickly identify tumor cells and predict treatment response, ultimately leading to a favorable therapeutic outcome.

One such prognostic marker seems to be survivin. Many studies have shown that survivin is strongly expressed in a vast majority of cancers. Its overexpression was demonstrated in breast and lung cancer, prostate, gastric, colon, bladder, and esophageal carcinomas, osteosarcomas, and lymphomas. In many of those tumors, high activity of the survivin gene was associated with a poor prognosis and worse survival rates. Moreover, survivin expression was correlated with resistance to chemotherapy and radiotherapy-induced apoptosis. Since survivin may be identified as an independent prognostic factor and inhibitor of apoptosis, it may prove to be a useful therapeutic target in cancer therapy.

Key words: **survivin** / **tumor biomarker** / **neoplasms** /

Streszczenie

Trafne rozpoznanie i właściwa kontrola pacjentów z chorobami nowotworowymi są nadal jednymi z najistotniejszych czynników skutecznego leczenia nowotworów złośliwych u ludzi.

Pomocnymi w szybkiej identyfikacji komórek nowotworowych i przewidywaniu ich odpowiedzi na leczenie, a w konsekwencji mającymi korzystny wpływ na wyniki leczenia mogą być badania skupiające się na poszukiwaniu markerów specyficznych dla guzów. Jednym z takich markerów prognostycznych wydaje się być surwiwina.

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Liczne badania wykazały między innymi, iż ulega ona silnej ekspresji w większości nowotworów złośliwych. Jej nadmierną ekspresję wykazano w nowotworach piersi, prostaty, żołądka, jelita grubego, pęcherza moczowego, raku przełyku, płuc, kostniakomięsakach oraz chłoniakach. W większości badanych przypadków wysoka aktywność genu surwiwiny wiązała się ze złym rokowaniem i krótszym czasem przeżycia pacjentów.
Wykazano dodatkowo, że ekspresja surwiwiny korelowała z opornością na chemio i radioterapię, apoptozozależną. Od czasu kiedy surwiwina została zidentyfikowana jako niezależny czynnik prognostyczny i inhibitor apoptozy, stała się ona ważnym celem w terapii przeciwnowotworowej.

Słowa kluczowe: **surwiwina** / **marker biologiczny guza** / **nowotwory** /

Survivin's structure and function

Survivin (also called IAP 4) is a protein with the ability to regulate the cell cycle and programmed cell death. It is the smallest member of the IAP (inhibitor of apoptosis protein) family, controlling chromosome compaction, mitotic spindle formation, and microtubule dynamics. At the molecular level, survivin is a multifunctional protein which not only plays a central role in cell division, but also suppresses apoptosis and enhances angiogenesis [1].

Survivin was discovered in 1997 by Ambrosini and colleagues. The 16.5 kDa protein contains a single 70 amino acid BIR (baculovirus repeat) domain and an extended α-helical coiled-coil C-terminus. It is encoded by a single gene *BIRC5* mapped to chromosome 17q25 [2].

One of survivin's basic roles is the inhibition of apoptosis. It acts like other proteins belonging to the IAP family and blocks both the extrinsic and intrinsic apoptotic pathways by direct or indirect interactions with capsase-3 and -7 [1, 2, 3]. Another mechanism of apoptosis inhibition involves preventing the release of Smac/DIABLO from the mitochondria or binding directly to XIAP (X-linked inhibitor of apoptosis protein), also known as inhibitor of apoptosis protein 3 (IAP3) [7, 8].

Survivin is also involved in the regulation of mitosis. This protein acts during all stages of cell division, including mitotic spindle formation *via* chromosome separation and cytokinesis. Subcellular localization of survivin has shown that the protein is a member of the chromosomal passenger complex, which is associated with centromeres, but during the metaphase-anaphase transition it leaves the centromeres and remains in the spindle mid-zone [4].

Survivin has been shown to be expressed only during mitosis. Its expression increases in G2/M phase and decreases rapidly in G1 and its expression is regulated by a number of factors such as: E2F, Sp1, TCF, and heat shock protein (Hsp) 90 [9, 10]. Survivin is also regulated by p53. Wild type p53 suppresses survivin's expression at the level of mRNA, and down-regulation of survivin by p53 is important for inducing apoptosis. Additionally, post-transcriptional phosphorylation has been proven to play an important regulatory role in survivin activation [12].

Results of immunohistochemical studies have demonstrated that in human cancer cells survivin may exist in distinct nuclear and cytoplasmic subcellular pools. Its expression in the nuclei of tumor cells appears to be associated with unfavorable clinical outcomes [32-34].

In addition to the full-length transcript, four alternative splice variants of the survivin gene product have been described: survivin-ΔEx3, survivin-3B, survivin-2ß and survivin-2α [4].

Previous studies showed that an imbalance in the alternative transcript ratio may influence the cell's apoptosis resistance or sensitivity [4].

It has been demonstrated that the survivin transcript ratio is also informative in predicting the response to chemotherapy. Many reports suggest that survivin-ΔEx3 and survivin-3ß are cytoprotective, while survivin-2α and survivin-2ß are pro-apoptotic. Survivin-ΔEx3 has been associated with higher tumor staging, increased tumor aggressiveness, as well as poor prognosis [13]. The localization of the transcription variants of survivin, namely survivin-2ß and survivin-ΔEx3, differs. While survivin-2ß was localized in both nuclear and cytoplasmic compartments, survivin-ΔEx3 was found only in the nucleus [14].

Survivin expression in premalignant lesions

Survivin expression in normal physiological conditions is limited to embryogenesis, hematopoietic, epithelial, and gonadal cell lines [11]. The protein is also detected in mature tissues with high proliferation potential such as placenta, thymus, endothelium, and CD34+ hematopoietic progenitor cells [11, 24, 25].

However the level of survivin in normal adult tissues was shown to be much lower than in tumor tissues [2]. Survivin transcripts are also commonly found in pre-cancerous lesions, including: breast adenomas, Bowen disease, and colon polyps. Survivin was also detected in breast ductal carcinoma *in situ* and cervical intraepithelial neoplasia (CIN) [26, 27].

In addition to the increased expression of survivin observed in premalignant lesions, its subcellular localization may be an important prognostic factor as well. Subcellular localization of survivin may change during malignant transformation like in the case of survivin's cytoplasmic staining in non-malignant hepatocytes and its nuclear staining in hepatocellular carcinoma [28]. Similarly in malignant melanoma cells, staining for survivin was found only within the nuclei of those cells [29].

The increased expression of survivin in a number of pre-invasive lesions with a high predisposition of progressing to malignancy is one of the reasons that survivin is considered to be a tumor marker.

Survivin expression in cancers

Survivin overexpression is observed in a variety of cancers and correlates with an unfavorable clinical outcome. Survivin overexpression was found in 96% of lung cancer specimens, 100% of colon adenocarcinomas, 71% of prostate adenocarcinomas, 80% of glioblastomas and 100% of laryngeal carcinomas [5,6]. The activity of its gene is also high in patients with hematologic malignancies, including acute leukemias and lymphomas [31].

Elevated survivin expression is associated with clinicopathologic variables of aggressive disease and correlates with shorter survival time. A large study analyzing 275 patients with breast cancer demonstrated that survivin is a significant prognostic factor and predicted the outcome, independent of patients' age, tumor size and histologic grade [32]. In the case of ovarian cancers, survivin expression correlates with the histological grading of the tumor [32]. Moreover, ovarian carcinomas revealed that nuclear localization of survivin correlated with poor prognostic factors such as: high histologic grade, mutant p53, and poor histologic type [35].

Many reports suggest that the differential localization of survivin - nuclear or cytoplasmic - may reflect different protein functions. It is suggested that the nuclear pool of survivin controls cell division, whereas cytoplasmic survivin works as a cytoprotective agent. Nuclear survivin has been shown to be an unfavorable prognostic indicator in esophageal, hepatocellular, non-small cell lung, and ovarian cancers, as well in lymphomas and endometrial cancers [33, 34]. In contrast, immunohistochemical analysis has revealed that cytoplasmic survivin linked to an anti-apoptotic effect has no prognostic significance. In view of the data published thus far, survivin's role in various cancers is contradictive. There is no definite explanation for the differences observed. However the simplest one is based on the presumption that different techniques were used to study survivin's presence and function. Thus, the search for a new method is carried out in order to establish the protein's expression, mechanism of action, and correlation with clinical parameters.

Recently the prognostic value of survivin's different splice variants has been considered. There is conservation of anti-apoptotic properties of survivin-ΔEx3 and a markedly reduced anti-apoptotic potential of survivin-2B [36].

Loss of survivin-2B expression was found in the later stages of cancer development, while survivin and survivin-ΔEx3 were not detected, suggesting different roles of these variants in tumor development. In addition, the different subcellular localization of survivin-ΔEx3 observed in the nucleus, and survivin-2B detected in cytoplasm seems to confirm their potential for different roles. These observations indicate that survivin-ΔEx3 and survivin-2B may play a role in tumor progression and/or tumorigenesis [37].

Analysis of gastric cancers demonstrated that levels of survivin-2B and survivin- ΔEx3 measured at the level of mRNA possess an important prognostic value. These findings may imply different properties of survivin splice variants: where the anti-apoptotic potential of survivin-ΔEx3 is preserved, survivin-2B has lost its anti-apoptotic potential and may act as a naturally occurring antagonist of the full-length survivin [38].

Previous studies focused on benign brain and pituitary tumors demonstrated the presence of survivin in these tissues [15, 16]. Overexpression of survivin was found in primary tumors of the nervous system, meningiomas, and benign peripheral nerve tumors [30]. It was found to be a characteristic feature of pituitary tumors, though survivin was also present in normal pituitary tissue. However the level of the gene expression was 6-fold higher in tumors than in normal pituitary tissue [16]. All these findings suggest that an increase in survivin expression might be one of the factors involved in pituitary neoplastic transformation. It was confirmed by further correlation between survivin overexpression and cell proliferation using the proliferation ratio (using the proliferation marker – PCNA) [15]. Such a correlation was demonstrated in different tumors, and there was a close association between survivin expression and a high proliferation index, low apoptotic rate, resistance to chemio- and radiotherapy, and an increased percentage of cancer relapse [20, 21, 22, 23].

Thus, survivin represents a unique target for biologic therapy in many human malignancies. A number of survivin-directed anticancer therapies, including transcriptional repressors, antisense oligonucleotides (ASOs), hammerhead ribozymes, small interfering RNAs (siRNAs), dominant-negative mutants, small molecular inhibitors, cyclin-dependent kinase (CDK) inhibitors, and immunotherapies are currently in the development stage [41]. Popular pharmacological agents consist of histone deacetylase inhibitors, mitogen-activated protein kinase and cyclin-dependent kinase inhibitors [39].

It has already been reported that suppression of survivin expression using antisense oligonucleotides (ASO) induces tumor cell apoptosis *in vitro* and *in vivo* [17]. Survivin ASOs were first used against malignant melanoma cell lines. Transfection with the ASOs triggered spontaneous apoptosis linked to decreased endogenous survivin expression [41]. In another study intratumoral injection of plasmids blocking survivin's expression stimulated the generation of tumor-specific CTLs (cytotoxic T lymphocytes). This may be beneficial for the treatment of large lymphomas [40].

Many survivin-based vaccines are in the clinical trials and peptide vaccinations have been used in the treatment of colorectal and breast cancers. Moreover, they have been well tolerated [41].

Concluding remarks

Recent studies have identified survivin as a factor responsible for cancer progression and tumor angiogenesis. The prognostic value of survivin for many human cancers has shown it to be correlated with an unfavorable clinical outcome. All these finding suggest that survivin expression has the potential for use as a predictive biomarker to identify cancer.

The high expression of survivin in cancer cells, with little expression in most normal tissues, makes survivin an attractive anticancer molecular therapeutic target. A wide variety of strategies have been employed to reduce survivin activity, and strong pre-clinical data have led to early-phase clinical trials in which its effectiveness has been established. Further investigations concerning the regulatory mechanisms of survivin's expression and function in normal and cancerous cells will help to elucidate survivin's biology and consequently to develop innovative strategies for selective survivin inhibition.

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