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# Target Therapy in Neuroblastoma

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## Abstract

Neuroblastoma is an embryonal malignancy that originates in the sympathetic nervous system. It is the most common solid tumor in infants and the most frequent extracranial solid tumor in children. Neuroblastoma accounts for 10% of childhood malignancies with 75% occurring in children <4 years. Stage, age, clinical and tumor genomic features are the principal criteria for determining treatment policy. Treatment modalities traditionally employed in the management of neuroblastoma are surgery, chemotherapy, and radiotherapy. Intensive multimodal treatment in patients with neuroblastoma has resulted in improved survival rates. However, there is a considerable percentage of patients with refractory and relapsed disease. Targeted therapy for neuroblastoma involves treatment aimed at molecular targets that have a unique expression in this childhood cancer. A large number of molecular targets have been identified for the treatment of high-risk and relapsed neuroblastoma. Treatment in this way aims at providing a more selective way to treat the disease and decreasing toxicities associated with the conventional treatment regimen.

**Keywords:** neuroblastoma, target, therapy, refractory, relapse

## 1. Introduction

Neuroblastoma, originating in neural crest cells, can occur anywhere along the sympathetic nerve chain. By far the most common location for a primary tumor is the adrenal gland. In its most primitive form, the histology of neuroblastoma is marked by poorly differentiated small, round blue cells. At an intermediate stage of maturation, there is differentiation

toward ganglion cells. Tumors composed of a mixture of neuroblasts and mature ganglion cells are classified as ganglioneuroblastomas. At the most differentiated end of the spectrum is ganglioneuroma, a benign tumor composed entirely of mature ganglion cells, neuritis, and Schwann cell [1].

At diagnosis, 65% of patients have disseminated disease, most commonly spreading to the bone. Disseminated disease is typically manifested as fever and bone pain. Additionally, neuroblastoma is associated with a paraneoplastic syndrome of opsoclonus-myoclonus ("dancing eyes and feet syndrome") [2].

Treatment modalities traditionally employed in the management of neuroblastoma are surgery, chemotherapy, and radiotherapy. Recently immunotherapy has been established as an important component of advanced neuroblastoma treatment. Intensive multimodal treatment strategies improved the survival in children with neuroblastoma. However, issues related to treatment refractoriness and late effects as well as disease recurrence remain significant challenges [3].

Multiple therapeutic targets have been developed to offer an advantage of treating neuroblastoma in a more selective way to maximize the treatment efficacy and minimize its toxicity [3].

## 1.1. Neuroblastoma staging systems

Several staging systems have been used to classify disease extent. The International Neuroblastoma Staging System (INSS), which is based on clinical and surgical evaluations, was developed by consensus of major pediatric oncology groups (POGs) in the United States, Europe, and Japan. The INSS differentiates between INSS stages 1, 2A, 2B, 3, 4, and 4S, based on surgical excision, lymph node involvement, and metastatic sites [4].

Because the INSS is a surgically based staging system, the stage for patients with locoregional disease can vary based on degree of surgical resection. Noteworthy, patients with localized disease who will not undergo surgery cannot be adequately staged. For these reasons, the International Neuroblastoma Risk Group (INRG) Task Force developed a new pretreatment staging system in 2005 based on clinical criteria and specific image-defined risk factors. Required imaging modalities include CT or MRI, as well as MIBG scintigraphy. The International Neuroblastoma Risk Group Staging System (INRGSS) differentiates between L1, L2, M, and MS stages. The INRGSS differs from the INSS in that the INRGSS is based on preoperative imaging characteristics and not on surgical resection. The age for the INRGSS MS stage is set at 547 days (18 months) compared with 12 months in the INSS 4S stage (**Table 1**) [5].

## 1.2. Risk factors and risk stratification

Children's Oncology Group Neuroblastoma Risk Stratification is the most commonly used tool to stratify patients based mainly on tumor stage, patient's age, MYCN amplification, DNA index, and tumor histology (**Table 2**) [6].

| INSS | INRGSS |
| --- | --- |
| Stage 1: localized tumor with complete gross excision; ± microscopic residual disease; representative ipsilateral lymph node negative for tumor microscopically | Stage L1: localized tumor not involving vital structures as defined by IDRFs and confined to one body compartment |
| Stage 2A: localized tumor with incomplete gross excision; ipsilateral lymph node should be negative for tumor microscopically | Stage L2: locoregional tumor with the presence of one or more IDRFs |
| Stage 2B: localized tumor with or without complete gross excision; ipsilateral lymph node should be positive for tumor microscopically, while contralateral lymph nodes should be negative microscopically | Equals stage L2 |
| Stage 3: unresectable unilateral tumor crossing the midline, ± involvement of regional lymph node, or localized unilateral tumor with involvement of contralateral regional lymph node or midline tumor with bilateral extension by infiltration (unresectable) or by lymph node involvement | Equals stage L2 |
| Stage 4: primary tumor with dissemination to distant lymph nodes, bone, bone marrow, liver, skin, or other organs | Stage M: distant metastatic disease (except stage MS). Distant lymph node involvement is metastatic disease. Ascites and pleural effusion, even if malignant cells are present, do not constitute metastatic disease unless they are remote from the primary tumor |
| Stage 4S: localized primary tumor in infants younger than age 1 year (localized as in stage 1, 2A, or 2B) with dissemination limited to the skin, liver, or bone marrow (<10% malignant cells) | Stage MS: metastatic disease in children younger than 547 days (18 months) of age with metastases confined to the skin, liver, and/or bone marrow (<10% malignant cells); MIBG scan must be negative in the bone and bone marrow. Primary tumor can be L1 or L2 with no limitations in terms of crossing or infiltration of the midline |

**Table 1.** Comparison between INSS and INRGSS.

## 1.3. Multimodal treatment approach for neuroblastoma

### 1.3.1. Low-risk disease

Low-risk NB patients require minimal therapy; previous pediatric oncology group (POG) and children cancer group (CCG) studies have shown that treatment with surgery alone results in survival rate >95% for patients with stage 1 disease. Even if there is microscopic residual disease, adjuvant treatment with radiotherapy or chemotherapy is not indicated [7, 8]. Diploidy and unfavorable histology predict for local recurrence and, therefore, careful follow-up is necessary, as local recurrence or distant metastasis may rarely occur and require salvage treatment [9].

The primary goals of surgery are to determine an accurate diagnosis, provide accurate surgical staging, completely remove the tumor, offer adjuvant therapy for delayed primary surgery, and remove residual disease with second-look surgery [10].

Management of infrequent patient with stage 1 or 2 with NMYC amplification remains controversial. Although patient with MYCN-amplified stage 1 tumor has significantly event-free

survival rate [11], a subset may achieve long-term remission following surgery alone. These rare cases may require continued prospective evaluation to clarify optimal management [12].

### 1.3.1.1. Stage 4S disease

Infants with stage 4S disease have, in general, a good prognosis; high survival rate has been reported in those infants whose tumor lacks MYCN amplifications [13]. Interestingly Tonini and colleagues reported that, in Italian experience, favorable outcomes were also seen in infants with MYCN-amplified stage 4S neuroblastoma [14]. The tumor may regress spontaneously due to programmed cell death and, therefore treatment is not always necessary. If there are no distressing or life-threatening symptoms, it is possible to follow an observation policy hoping

| Stage | Age | MYCN | Ploidy | Histology | Other | Risk group |
|---|---|---|---|---|---|---|
| 1 | | | | | | Low |
| 2A/2B | | Not amplified | | | >50% resection | Low |
| | | Not amplified | | | <50% resection | Intermediate |
| | | Not amplified | | | Biopsy only | Intermediate |
| | | Amplified | | | | High |
| 3 | <547 days | Not amplified | | | | Intermediate |
| | ≥547 days | Not amplified | | Favorable | | Intermediate |
| | | Amplified | | | | High |
| | ≥547 days | Not amplified | | Unfavorable | | High |
| 4 | <365 days | Amplified | | | | High |
| | <365 days | Not amplified | | | | Intermediate |
| | 365–547 days | Amplified | | | | High |
| | 365–547 days | | DI = 1 | | | High |
| | 365–547 days | | | Unfavorable | | High |
| | 365–547 days | Not amplified | DI > 1 | Favorable | | Intermediate |
| | ≥547 days | | | | | High |
| 4S | <365 days | Not amplified | DI > 1 | Favorable | Asymptomatic | Low |
| | <365 days | Not amplified | DI = 1 | | | Intermediate |
| | <365 days | Missing | Missing | Missing | | Intermediate |
| | <365 days | Not amplified | | | Symptomatic | Intermediate |
| | <365 days | Not amplified | | Unfavorable | | Intermediate |
| | <365 days | Amplified | | | | High |

**Table 2.** Children's Oncology Group neuroblastoma risk stratification schema.

for spontaneous regression. Nonintensive chemotherapy is called for if there are major symptoms, e.g., massive hepatomegaly causing respiratory distress. Alternatively, low-dose irradiation may precipitate regression [15].

### 1.3.1.2. More advanced operable disease

Patients with lymph node involvement, that is, stage 2B and some stage 3 cases, are treated principally by surgery. The need for adjuvant treatment depends on age. In infants <6 months, the role of chemotherapy is controversial. However, in older children, chemotherapy is definitely warranted, using a schedule such as "OPEC," which comprises vincristine, cisplatin, etoposide, and cyclophosphamide [16]. It is often preferable to use chemotherapy as the initial treatment in these patients aiming at reducing the tumor bulk, making complete removal more likely and the surgery safer. Data from POG in patients with stage 2B and stage 3 disease have shown that complete resection is not associated with a significantly better event-free survival than incomplete resection, while patients with favorable Shimada histology have a significantly better event-free survival rate at 2 years than those with unfavorable Shimada (92% versus 58%, respectively) [17].

Irradiation of the tumor bed to eradicate residual disease is controversial.

In a retrospective review of patients with Children's Cancer Study Group (CCSG) stage II disease, no significant benefit was observed in irradiated children [18]. POG designed a randomized trial to evaluate the role of placing radiotherapy in addition to chemotherapy in patients >1 year with nodal disease detected at resection of the primary tumor. Significantly improved local control and survival rates were seen in irradiated children [19]. As the chemotherapy protocol used in this study was less intensive than that now considered standard, it remains possible that more intensive chemotherapy might be as good as combined chemotherapy and radiotherapy in the POG trial. Based on the fact that patients with biologically favorable tumors have a good prognosis [20], even in the presence of residual disease, it is reasonable not to irradiate patients with biologically favorable stage 2B and stage 3 tumors, with or without residual disease after chemotherapy and surgery. Despite the adverse effects of radiotherapy in young children, it should be considered in the treatment plan of patients with biologically unfavorable stage 2B and stage 3 tumors with residual disease [21].

### 1.3.2. Intermediate-risk disease

Intermediate-risk patients with favorable biology tumors are treated with a short course of chemotherapy (four cycles), while intermediate-risk patients with unfavorable biology receive a longer course of chemotherapy (eight cycles).

In previous POG studies, treatment for infants with regional and metastatic disease was stratified by tumor cell ploidy and MYCN amplification status. Infants with hyperdiploid tumors were treated with cyclophosphamide and doxorubicin, whereas infants with diploid tumors were treated with cisplatin and teniposide after an initial course of cyclophosphamide and doxorubicin [22].

### 1.3.3. High-risk disease

Survival for high-risk neuroblastoma patients has improved modestly during the past 20 years, although cure rates remain low [23]. This improvement was attributed to intensification of induction chemotherapy, megatherapy consolidation, as well as improved supportive care. Chemotherapy dose intensity has been reported to correlate strongly with both response and progression-free survival where response rates 70–80% have been observed with intensive multiagent induction treatment protocols [24]. Intensification of consolidation therapy with autologous hematopoietic stem cell transplantation following myeloablative conditioning chemotherapy with or without total body irradiation also contributes to improved overall survival in several single-armed studies [25].

Patients with advanced neuroblastoma such as those with stage 4 or inoperable stage 3 disease should receive initial chemotherapy with "OPEC" or a similar protocol. In stage 3 patients, if chemotherapy rendered the tumor operable, it should be removed. In stage 4 patients, surgery to remove residual primary tumor should also be considered complete remission at all metastatic sites has been achieved. Dose intensification treatment strategies, designed to achieve a greater degree of cytoreduction and to circumvent emergence of resistant clones by using a larger number of non-cross-resistant chemotherapeutics in higher doses over a shorter time, are feasible but have not yet proved significantly superior to OPEC [26].

### 1.3.3.1. Megatherapy

Megatherapy combining high-dose myeloablative chemotherapy and/or total-body irradiation with either autologous bone marrow transplantation or peripheral blood stem cell reinfusion is often used in advanced neuroblastoma. The rationale of megatherapy is to eradicate the undetectable minimal residual disease and prevent disease relapse. Chemotherapeutics at higher-dose levels can bypass inadequate membrane transport and saturate detoxification pathways and DNA repair mechanisms and therefore attack residual tumor cells which have survived initial chemotherapy and became resistant to chemotherapy at conventional doses. Single-agent high-dose melphalan, which was evaluated in European Neuroblastoma Study Group (ENSG) trial 1, is one of the more commonly used conditioning regimens [27].

Radiation, usually in the form of TBI, has been evaluated with high-dose chemotherapy and autologous bone marrow transplantation in treatment of advanced neuroblastoma, and the results were not superior to those achieved by chemotherapy alone, yet the early and late side effects were greater. Allogeneic BMT is not associated with improved results. The European Blood and Marrow Transplant Group has reviewed the data of more than a thousand of neuroblastoma patients who have received myeloablative therapy. Overall, survival was 33% at 5 years, but relapses may still be seen later. When patients relapsed after initial autologous BMT, salvage was not possible, but autologous BMT did salvage 15% of patients in the second or subsequent relapse who had not previously undergone transplantation. The poor outcome for transplantation in stage 4 patients > 1 year is mainly due to persistent skeletal or bone marrow involvement [28].

## 2. Promising therapeutic targets of neuroblastoma

The 5-year event-free survival for high-risk neuroblastoma is <50%, including children with metastatic neuroblastoma >18 months of age and patients with locoregional or metastatic neuroblastoma with MYCN amplification [5]. Improved outcome was achieved with intensive combination induction chemotherapy and surgery, followed by myeloablative therapy with hematopoietic stem cell transplantation and then differentiation therapy with isotretinoin. Isotretinoin is the first tumor-targeted therapy with established activity in neuroblastoma [29]. After that, a large number of molecular targets have been developed for treatment of high-risk, refractory, and relapsed neuroblastoma. These molecular targets can supplement or replace some of the intensive chemotherapy used for treatment of neuroblastoma.

### 2.1. Potential advantages of target therapy over conventional chemotherapy

1. Effective cancer treatment with less side effects (increased therapeutic index) due to the targeting of a unique characteristic within the tumor cells, which is usually absent in normal body cells

2. Decreased likelihood of the development of resistance to the targeted therapy due to the molecular target being essential for the viability of the cancer

### 2.2. Targeting human norepinephrine transporter (hNET) with $^{131}$I-metaiodobenzylguanidine (MIBG)

The rationale for using 131 I-MIBG as a targeted radiopharmaceutical for high-risk neuroblastoma was based on the observation that 90% of these tumors are MIBG avid. Studies reported impressive response rates in relapsed disease, and in the largest phase II trial, 37% of patients had a partial response (PR) or complete response (CR) [30].

Many efforts were made to maximize the benefits of MIBG:

a. Increasing tumor radiation dose: the response rate of 42 mCi/kg dose did not appear to be different from that obtained with the standard 18 mCi/kg dose [31].

b. Repeated MIBG infusions: investigators reported a benefit from repeated MIBG infusions given 6–12 weeks apart, with continued improved response in some of the patients with each successive infusion [32].

c. Different isotope (125 I-MIBG): the results were disappointing [33].

d. No carrier added: this approach had the advantage of infusion over 30 min instead of 90–120 min. However, NANT trial showed toxicity, and efficacy profiles equal to those observed with the standard preparation [34].

e. MIBG combined with radiosensitizers or chemotherapy:

- Italian investigators treated 16 children with refractory or relapsed neuroblastoma using ^{131}I-MIBG combined with cisplatin and cyclophosphamide with or without etoposide and vincristine and obtained 12 PRs [35].

- Two studies investigated MIBG combined with a camptothecin, with tolerable toxicity and measurable response [36, 37].

- Combination of a histone deacetylase inhibitor (Vorinostat) with 131I-MIBG showed that Vorinostat at 180 mg/m2/dose is tolerable with 18 mCi/kg MIBG, and A phase II trial comparing this regimen to single-agent MIBG is ongoing [38].

- 131I-MIBG before standard induction therapy with 66% response rate [39].

- 131I-MIBG at the end of induction and before myeloablative therapy for patients with residual MIBG-positive disease reported a response rate of 46% but no improvement in overall survival [40].

### 2.3. GD2-targeted immunotherapy of high-risk neuroblastoma

Promising results have been emerged with immunotherapy targeting the surface glycolipid molecule disialoganglioside (GD2) that is uniformly expressed by neuroblastomas and gliomas, sarcomas, and some melanomas [41].

GD2 expression is weak in normal human tissues and restricted to neurons, melanocytes, and peripheral pain fibers. Based on this, GD2 seems to be an ideal antigen target for immunotherapy of neuroblastoma [41].

#### 2.3.1. Mechanism of action

**a.** CDC: complement-dependent cytotoxicity

**b.** ADCC: antibody-dependent cell cytotoxicity

##### 2.3.1.1. First-generation anti-GD2 mAbs

Many phase I and phase II clinical trials led to the pivotal randomized COG phase III study. This study was conducted on 226 eligible patients to determine whether immunotherapy with ch14.18 combined with GM-CSF, IL-2, and isotretinoin would improve survival compared to isotretinoin alone for children with high-risk neuroblastoma in the first response after myeloablative therapy and stem cell rescue. EFS was significantly higher for patients randomized to immunotherapy, with a 2-year estimated EFS from randomization of 66% versus 46% for patients randomized to isotretinoin alone (P = 0.01). The immunotherapy group also showed significantly higher OS (86% versus 75% at 2 years; P = 0.02). This represents the first successful immunotherapy to target a nonprotein antigen [42].

*2.3.1.2. Second-generation anti-GD2 mAbs*

Phase II study of Hu14.18-IL-2 immunocytokine showed 5 CRs in 23 neuroblastoma patients evaluable only by bone marrow histology and/or MIBG, but no responses for patients with measurable disease. Preliminary findings of a phase I clinical trial of hu14.18K322 showed reduced neuropathic pain compared to Hu14.18-IL-2 [43].

Yu and colleagues in 2001 carried out a clinical trial of mAb 1A7 as a GD2 vaccine in 31 children with high-risk neuroblastoma who achieved the first or subsequent remissions. No systemic toxicities were observed with subcutaneous injections given periodically over 2 years, and only local reactions were seen. Sixteen of 21 children who enrolled during the first remission had no evidence of disease progression at a median of 6 years, whereas only one of ten children in the second remission remains progression-free. Yu and colleagues concluded that mAb1A7 vaccine is effective at inducing biologically active anti-GD2, has little toxicity, and may be useful for controlling minimal residual disease [44].

## 2.4. ALK as a therapeutic target in neuroblastoma

Anaplastic lymphoma kinase (ALK) is a tyrosine kinase that is mutated or amplified in about 10% of neuroblastomas and expressed on the surface of most neuroblastoma cells [45].

There are several reasons why inhibition of ALK would be a feasible therapeutic option in neuroblastoma:

1. ALK is expressed on the surface of most neuroblastoma tumor cells and is restricted to the brain following development [45].

2. A proportion of neuroblastoma cells that overexpress phosphorylated ALK that is neither mutated nor amplified respond to ALK depletion by undergoing apoptosis [46].

3. ALK inhibition may provide an effective targeting strategy against MYCN-amplified tumors [47].

Crizotinib was the first drug to be approved by FDA for treatment of ALK-rearranged cancers. Preclinical testing showed the sensitivity of neuroblastoma cell lines with ALK amplification. Crizotinib is being tested in a phase I/II trial for children with neuroblastoma and other solid tumors bearing ALK mutations [48].

## 2.5. The topoisomerase 1 inhibitors

The topoisomerase 1 inhibitors such as topotecan and irinotecan are often used early for treatment of children with relapsed neuroblastoma because of their acceptable efficacy and toxicity profiles. Topotecan efficacy is enhanced when it is combined with low-dose cyclophosphamide. Irinotecan and temozolomide are a well-tolerated combination, and efficacy is being studied [49].

## 2.6. Retinoids

A randomized clinical trial of 13-cis-retinoic acid following myeloablative chemotherapy regimen established the importance of retinoids in therapy for high-risk neuroblastoma. Fenretinide produced multi-log cell kill in multiple neuroblastoma cell lines, even those resistant to other retinoids [50]. Phase I studies have shown that fenretinide is generally well tolerated, and COG phase II trial has been completed. Newer formulations of this drug are currently in the pipelines to facilitate oral administration to young children [51].

## 2.7. Targeting CD133 biomarker

The tumor-initiating properties of CD133 have been discovered through studies such as the one performed by Cournoyer et al. Through genotype analysis CD133 expression is found to be associated with the expression of the Ephrin-A2 (EFNA2) protein. This protein can play a role in cancer development. EFNA2 is expressed in stem cells and can promote the formation of tumors. CD133 and the associated EFNA2 protein are in the pipelines as potential therapeutic targets for neuroblastoma [52].

## 2.8. Angiogenesis inhibitors

Tumor vascularity has been correlated with aggressiveness in neuroblastoma. Based on this, angiogenesis inhibitors seem to be an attractive therapeutic option. Furthermore, pro-angiogenic molecules appear to be differentially expressed in high-risk tumors, whereas lower-risk tumors are characterized by a stroma that provides anti-angiogenic molecules in the microenvironment. Preclinical studies of anti-angiogenic drugs in neuroblastoma showed promising results [53].

## 2.9. Targeting insulin-like growth factor I receptor (IGF-IR)

IGF-1R is involved in the regulation of cell proliferation, survival, differentiation, and transformation. IGF-1R is highly expressed in neuroblastoma, and activation of IGF-1R induces MYCN expression. The expression level of IGF-1R has been correlated with tumor metastasis. Blocking IGF-1R with anti-IGF-1R antibodies resulted in the inhibition of neuroblastoma cell growth and tumor regression in neuroblastoma xenograft mouse models. The anti-IGF-1R monoclonal antibody (IMC-A12) is under investigation in phase II trial [54].

## 2.10. Targeting tropomyosin receptor kinase (TRK)

TRK is now known as the high-affinity receptor for nerve growth factor and as such is crucially involved in the growth, differentiation, and apoptosis of neuronal cells in both the central and the peripheral nervous systems. High expression levels of TRK have been correlated with poor outcome in neuroblastoma and chemotherapy resistance. Several TRK-blocking small compounds, such as CEP-701, have been developed. Blocking TRK using CEP-701 results in induction of apoptosis and growth inhibition of human neuroblastoma xenografts in nude mice. CEP-701 is under investigation in phase I trial for refractory and relapsed neuroblastoma. Other tyrosine kinase inhibitors, including inhibitors of the epidermal growth

factor receptor, might have activity against neuroblastoma [55]. Imantanib mesylate has also been investigated in neuroblastoma because some tumors appear to express c-kit, PDGFR, or both; however, activating mutations in these receptors have not been reported [56].

### 2.11. Targeting Aurora A kinase (AURKA)

AURKA is a serine/threonine kinase, which stabilizes the microtubule at the spindle pole during segregation of chromosomes. Based on this, AURKA is essential for G2-M progression, and its inhibition results in cell cycle arrest and apoptosis. AURKA is overexpressed in neuroblastoma, and amplification of its gene has also been observed in neuroblastoma cells. In phase I trials, promising results have also been obtained with AURKA inhibitor MLN8237 [57].

### 2.12. Targeting PI3K/AKT/mTOR pathway

The PI3K/AKT/mTOR pathway is an intracellular signaling pathway important in regulating the cell cycle. Therefore, it is directly related to cellular quiescence, proliferation, cancer, and longevity. PI3K activation phosphorylates and activates AKT that sequentially activates mTOR. In many cancers, this pathway is overactive, thus reducing apoptosis and allowing proliferation. Rapamycin (Sirolimus), an antifungal agent with immunosuppressive properties, was first isolated in 1975 from the soil of the island of Rapa Nui or Easter Island. In the 1980s, rapamycin showed a broad anticancer activity. However, clinical development of rapamycin as an anticancer agent was hampered by unfavorable pharmacokinetics. Recent development of rapamycin analogs with favorable pharmacokinetics such as temsirolimus, everolimus, and ridaforolimus opened up the present era of mTOR inhibitors as anticancer agents [58].

### 2.13. Other strategies

Epigenetic silencing of genes that are crucial for induction of apoptosis, such as caspase-8, seems to occur frequently in neuroblastoma. Therefore, demethylating agents such as decitabine are currently being investigated. Histone deacetylase inhibitors showed preclinical activity against neuroblastoma. More than three histone deacetylase inhibitors are now in clinical trials for patients with refractory solid tumors. Heat shock protein 90 inhibitors are also of interest because they alter the function of molecules associated with neuroblastoma cell growth and proliferation, including AKT, IGF-1, and TrkB [59].

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