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Research Article

Salinomycin Suppresses PDGFRβ, MYC, and Notch Signaling in Human Medulloblastoma

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Abstract

The most frequent brain tumour in children is medulloblastoma (MB).Despite better treatment and care, over 30% of people pass away from the illness. The effects of salinomycin on cell proliferation, cell death, and cell cycle progression in human MB cell lines were examined in an effort to find a more potent therapy approach. The findings showed that salinomycin interrupts cell cycle progression, promotes cell death, and reduces cell proliferation in MB cells. Salinomycin's effects on the expression of crucial genes involved in proliferation and survival signalling were also examined, and it was discovered that salinomycin up-regulates the expression of cyclin A while down-regulating the expression of PDGFR, MYC, p21, and Bcl-2. The outcomes also show that salinomycin inhibits Hes1 and Hes5 expression in MB cells. Our findings give light on salinomycin's potential as a novel therapeutic treatment for MB patients.

Abbreviations

PDGFR stands for beta-type platelet-derived growth factor receptor; Bcl-2 stands for B-cell lymphoma 2; DLL1 stands for delta-like 1 in drosophila; DLL3 stands for delta-like 3 in drosophila; Hes1 stands for hairy and enhancer of split 1 in drosophila; Hey1 stands for hairy and enhancer of split related with yrpw motif 1; Hey2 stands for hairy and enhancer MAML1 stands for Mastermind-Like 1 (Drosophila), MAML2 for Mastermind-Like 2, and MAML3 for Mastermind-Like 3 (Drosophila). RBPJ stands for Recombination Signal Binding Protein for Immunoglobulin Kappa J Region (4–Sulfophenyl) Dimethyl sulfoxide; -2H- Tetrazolium, Inner Salt MAML1 stands for Mastermind-Like 1, MAML2 for Mastermind-Like 2, and MAML3 for Mastermind-Like 3. Immunoglobulin Kappa J Region Recombination Signal Binding Protein; MTS: 3-(4, 5-Dimethylthiazol-2-Yl)-5-(3-Carboxymethoxyphenyl)-2- (4–Sulfophenyl) Dimethyl sulfoxide; -2H- Tetrazolium, Inner Salt

Introduction

The most typical malignant brain tumour in children is called a medulloblastoma (MB), an embryonal neuroepithelial tumour of the cerebellum [1]. Early on in its progression, this highly invasive tumour has a propensity to spread throughout the central nervous system. Around one-third of patients with MB tumours are still incurable, despite improvements in medical treatment outcomes for children with MB over the past few decades. Moreover, existing medical procedures have side effects that are hazardous and can leave long-term survivors with serious problems [2]. As a result, more potent medications are required to treat MB sufferers.Salinomycin, a mono carboxylic polyether antibiotic with a molecular weight of 751, is a common anti-coccidial medication. In comparison to paclitaxel, a commonly prescribed medication for breast cancer, salinomycin has recently been demonstrated to significantly lower the fraction of breast cancer stem cells (CSC) [3]. Salinomycin is a selective killer of human CSC and an effective murderer of multi-drug resistant human CSC-like cells, according to cumulative data [4–11]. Salinomycin has been demonstrated to regulate a number of signalling pathways, including the Wnt, NF-B, and p38 MAPK pathways, in cancer and CSCs [12–14].MB cells are thought to have stem cell origins because of their capacity to develop into neuronal and/or glial cells [15,16]. A CSC-like population may exist and

contribute to MB treatment resistance, according to mounting data [17–19]. Through controlling downstream effectors such MYC, Notch signalling is essential for cell differentiation and proliferation and is vital for the onset and progression of MB [20–22]. The Notch pathway inhibitors, such as the -secretase inhibitor MK-0752, suppress the cleavage of Notch, eradicate the stem cell-like population [23–25], reduce cell proliferation, and increase apoptosis [23–25], which implicates Notch signalling as a target and may represent a further promising treatment approach for MB patients. In the current investigation, we for the first time identified salinomycin's anticancer properties.in cell lines of 3 MB. Also, we examined how salinomycin affected the expression of a few genes essential for MB cell proliferation, survival, and differentiation.

Discussion

In this investigation, we discovered that salinomycin strongly suppresses cell proliferation at concentrations between 0.25 and 4 M and causes cell death and cell cycle arrest. Salinomycin suppresses the expression of PDGFR, MYC, Bcl-2, p21 and some important effectors in the Notch signalling pathway, according to our analysis of changes in gene and/or protein expression that are involved in cell proliferation, cell death, and the Notch signalling pathway in response to salinomycin treatment (e.g., Hes1).Salinomycin has been demonstrated to have potent anti-cancer and anti-CSC actions in additional cancer types in vitro, in vivo xenografted mouse models, as well as pilot clinical investigations in people [33-35] after the revelation that salinomycin has anti-CSC activity in breast cancer [3]. Despite this, the Salinomycin's effects on MB cells haven't been investigated before. In this study, we show that salinomycin exerts severe cytotoxicity towards human MB cells. A dose-dependent increase in cell mortality (the sub-G0 population) and a notable decrease in cell proliferation after salinomycin therapy provided evidence for this finding. Cyclin A is necessary for DNA replication in both the S and G2 phases [36]. As salinomycin therapy prolongs the S and G2 phases, higher amounts of cyclin A may be the result. Also, there was a strong correlation between the data on cell proliferation and the cell cycle arrest during S-G2 phases and up-regulation of cyclin A expression.Hes1 and Hes5 are essential Notch signalling pathway effectors that are necessary for the development of MB illness and patient survival. Hes1 expression that is activated by Notch signalling is linked to considerably worse survival in MB patients, according to a study by Fan et al. [24]. The blocking of the Notch pathway has also been linked to cell death, cell cycle exit, and differentiation in MB cells, according to research by the same team [23]. According to this study, Notch signalling may be important for maintaining MB CSCs.

Our findings demonstrate that salinomycin reduced the transcription of Hes1 and DLL1. This showed its significance in the preservation of MB CSCs and partially explained the effects of salinomycin on MB cell survival. Moreover, MAML1, which was likewise reduced by salinomycin in MB cells, which had previously been demonstrated to serve as a coactivator to increase the Notch-induced transcription of Hes1 [37]. The downregulated protein levels of p21, another target gene of Notch signalling, may also be partially explained by the inhibition of the Notch signalling gene expression [38].The downstream target of canonical Wnt signalling has been identified as MYC [39]. In fact, in Wnt-transfected HEK293 cells, salinomycin inhibits the phosphorylation of the Wnt co-receptor lipoprotein receptor related protein 6 (LRP6) and causes its destruction [13]. It's likely that salinomycin's effect on Wnt signalling contributed to the downregulation of MYC. Salinomycin has been shown in this study to decrease Notch signalling in MB cells, and MYC is another target molecule for Notch signalling [40,41]. There's a chance that salinomycin uses Notch signalling to at least partially downregulate MYC. Furthermore, PDGFR signalling is downstream of MYC [42,43]. As a result, simultaneously addressing these routes for MB should offer an efficient treatment plan.MYC is It is frequently dysregulated in MB [44–46] and affects a variety of cellular processes by changing the expression of several functionally significant target genes [47]. According to recent research, of the four subtypes of MB, Group 3 MB, which is characterised by MYC overexpression, is associated with an aggressive illness and a poor prognosis [48]. Moreover, inhibiting MYC dramatically slows the development of MB cells [49]. High levels of PDGFR have also been linked to an aggressive phenotype of MB, in addition to MYC [50]. Our findings in this study reveal a novel molecular element of salinomycin's significant anti-cancer activities and emphasise the usefulness of salinomycin as a very promising therapy for treating MB by demonstrating that salinomycin may reduce the expression of MYC and PDGFR simultaneously [51].

Conclusions

Our research shows that salinomycin causes cytotoxicity in human MB cells. Our findings show that salinomycin therapy is effective in preventing MB cell proliferation, delaying the cell cycle, and triggering cell death. We further demonstrate that salinomycin therapy has changed several signalling pathways

in MB cells. The cytotoxic effects of salinomycin are most likely a result of the down-regulation of PDGFR and MYC as well as the inhibition of the Notch signalling pathway. When taken as a whole, this study implies that salinomycin may be a useful therapeutical drug for MB and calls for additional research.

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