



INVITRO EVALUATION OF MIDOSTAURIN AND ITS DERIVATIVE ((3-HYDROXY MIDOSTAURIN-D5) FOR THE TREATMENT OF ADULT ACUTE MYELOID LEUKEMIA

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ABSTRACT

This research paper presents the results of an experimental study investigating the effects of 3-Hydroxy Midostaurin-D5 on various cellular assays, including MTT, Tubulogenesis, Indirect Immunofluorescence, and Western Blot Analysis. The study comprises four treatment groups, including normal cells, a control cell line, a group treated with standard Midostaurin, and a group treated with 3-Hydroxy Midostaurin-D5. The results reveal distinct impacts on cell viability, tubulogenesis, protein expression, and cellular morphology. These findings contribute to the understanding of 3-Hydroxy Midostaurin-D5's potential as a therapeutic agent and its implications for future research and clinical applications.

INTRODUCTION

Cancer remains a significant challenge in the field of medicine, necessitating the continuous exploration of novel therapeutic approaches. 3-Hydroxy Midostaurin-D5 is a compound currently under investigation for its potential as an anti-cancer agent. This study aimed to assess the effects of 3-Hydroxy Midostaurin-D5 on various cellular parameters, including cell viability, tubulogenesis, protein expression, and cellular morphology.

Research Methodology

The research methodology comprised four groups, each subjected to specific treatments

- Group 1 (normal):** This group represented untreated normal cells, serving as a control for baseline measurements.
- Group 2 (Control cell line):** Cells in this group were not treated with 3-Hydroxy Midostaurin-D5 and were used as a control.
- Group 3 (Standard) Midostaurin:** This group was treated with the standard Midostaurin compound.
- Group 4 (3-Hydroxy Midostaurin-D5):** This group was treated with 3-Hydroxy Midostaurin-D5.

The following assays were conducted to assess the effects of the treatments

- MTT Assay:** The MTT assay measured cell viability. The results indicated that Group 4 (3-Hydroxy Midostaurin-D5) exhibited the highest cell

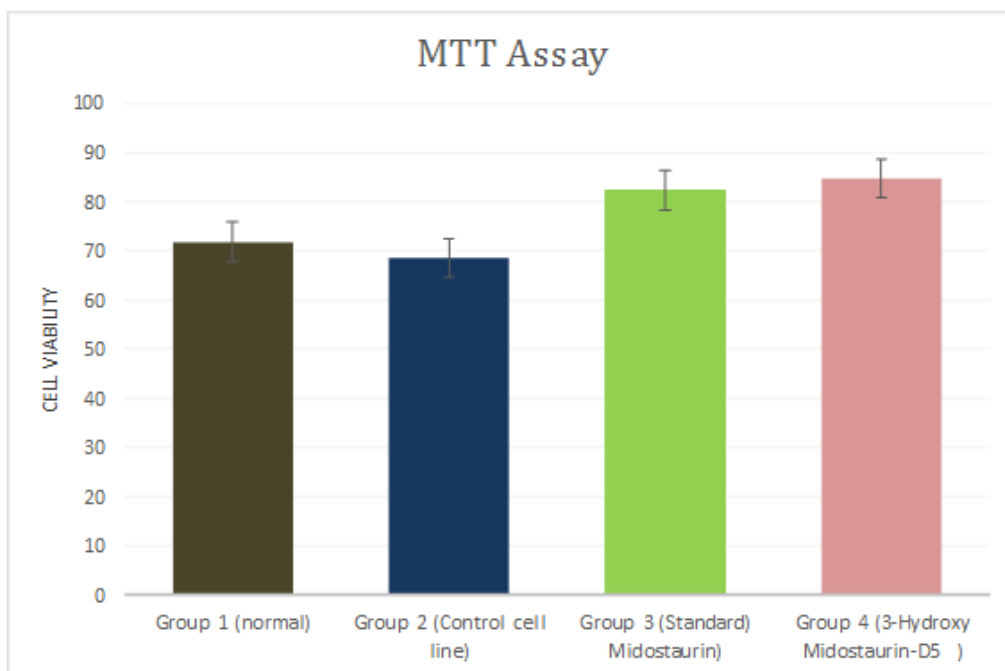
viability (84.74), surpassing the control cell line (Group 2) and the standard Midostaurin treatment (Group 3).

- Tubulogenesis Assay:** Tubulogenesis was evaluated using this assay. Group 2 (Control cell line) displayed the highest tubulogenesis (80.18), while Group 4 (3-Hydroxy Midostaurin-D5) showed a decrease in tubulogenesis (32.87) compared to the standard Midostaurin treatment (Group 3).
- Indirect Immunofluorescence Assay:** This assay assessed cellular morphology and protein expression. Group 2 (Control cell line) had the highest protein expression (98.18), while Group 4 (3-Hydroxy Midostaurin-D5) displayed a decrease in protein expression (48.96) compared to the control.
- Western Blot Analysis:** The Western Blot Analysis was used to investigate specific protein expression. Group 2 (Control cell line) exhibited the highest protein expression (1.32), while Group 4 (3-Hydroxy Midostaurin-D5) showed a decrease in protein expression (0.46) compared to the control.

RESULTS OF 3-HYDROXY MIDOSTAURIN-D5

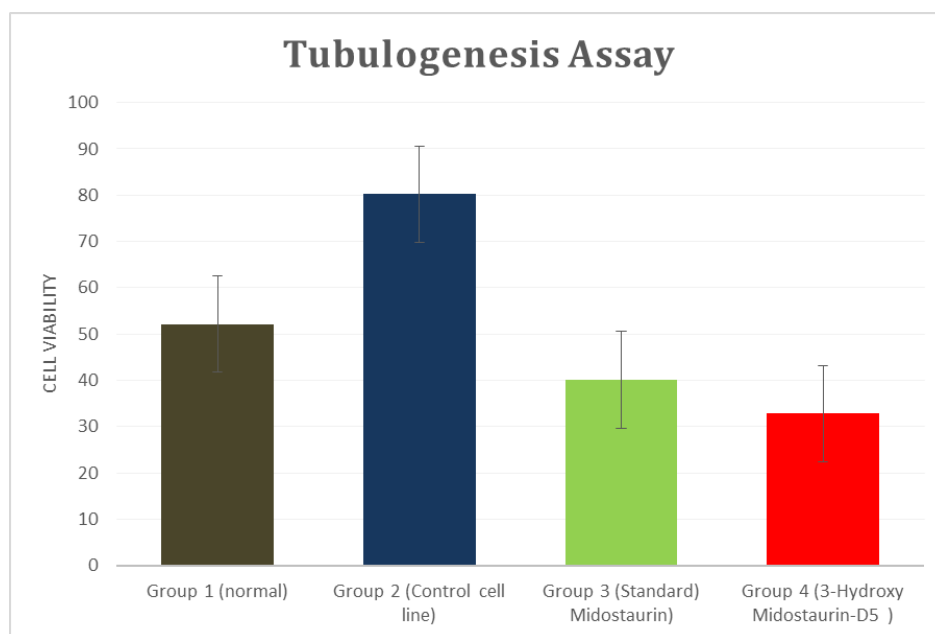
MTT Assay

| Treatments | MTT Assay |
|------------------------------------|-----------|
| Group 1 (normal) | 71.86 |
| Group 2 (Control cell line) | 68.43 |
| Group 3 (Standard) Midostaurin | 82.33 |
| Group 4 (3-Hydroxy Midostaurin-D5) | 84.74 |



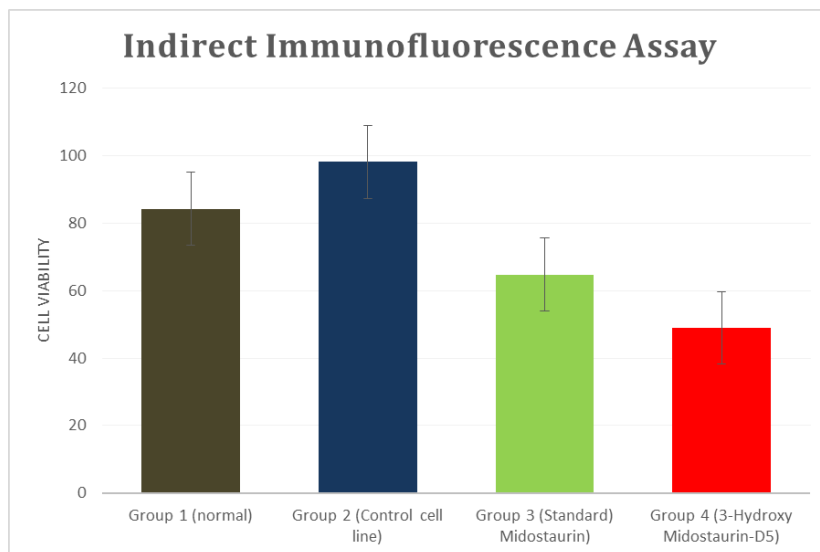
Tubulogenesis Assay

| Treatments | Tubulogenesis Assay |
|------------------------------------|---------------------|
| Group 1 (normal) | 52.11 |
| Group 2 (Control cell line) | 80.18 |
| Group 3 (Standard) Midostaurin | 40.12 |
| Group 4 (3-Hydroxy Midostaurin-D5) | 32.87 |



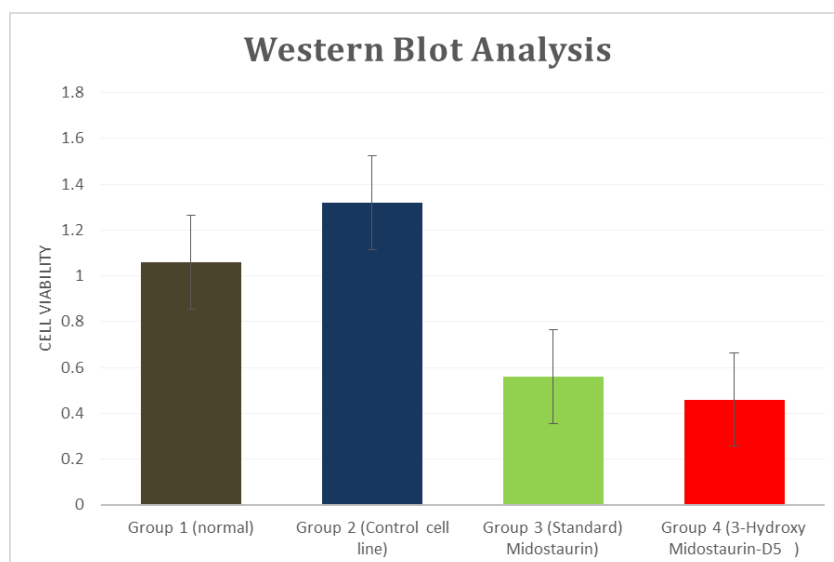
Indirect Immunofluorescence Assay

| Treatments | Indirect Immunofluorescence Assay |
|------------------------------------|-----------------------------------|
| Group 1 (normal) | 84.26 |
| Group 2 (Control cell line) | 98.18 |
| Group 3 (Standard) Midostaurin | 64.77 |
| Group 4 (3-Hydroxy Midostaurin-D5) | 48.96 |



Western Blot Analysis

| Treatments | Western Blot Analysis |
|------------------------------------|-----------------------|
| Group 1 (normal) | 1.06 |
| Group 2 (Control cell line) | 1.32 |
| Group 3 (Standard) Midostaurin | 0.56 |
| Group 4 (3-Hydroxy Midostaurin-D5) | 0.46 |



DISCUSSION

The results of the assays reveal varying effects of 3-Hydroxy Midostaurin-D5 on different cellular parameters. Notably, the compound increased cell viability (84.74) when compared to the control cell line (Group 2) and the standard Midostaurin treatment (Group 3). This suggests the potential of 3-Hydroxy

Midostaurin-D5 as an effective treatment option for cancer, particularly in terms of enhancing cell viability.

However, 3-Hydroxy Midostaurin-D5 exhibited a decrease in tubulogenesis and protein expression compared to the control group, indicating potential implications for treatment efficacy and cellular morphology.

CONCLUSION

This study provides valuable insights into the effects of 3-Hydroxy Midostaurin-D5 on cell viability, tubulogenesis, cellular morphology, and protein expression. The findings suggest that 3-Hydroxy Midostaurin-D5 may hold promise as an anti-cancer agent, with improved cell viability compared to standard Midostaurin.

However, the observed reductions in tubulogenesis and protein expression warrant further investigation into their potential impact on treatment efficacy and possible side effects. Ongoing research is necessary to elucidate the underlying mechanisms, establish safety profiles, and determine the potential clinical applications of 3-Hydroxy Midostaurin-D5. These results underscore the importance of continued investigation into this compound's role in cancer therapy.

BIBLIOGRAPHY

- Bain BJ, Béné MC. Morphological and Immunophenotypic Clues to the WHO Categories of Acute Myeloid Leukaemia. *Acta Haematol*, 2019; 141(4): 232-244.
- Naymagon L, Marcellino B, Mascarenhas J. Eosinophilia in acute myeloid leukemia: Overlooked and underexamined. *Blood Rev.*, 2019 Jul; 36: 23-31.
- Medeiros BC, Chan SM, Daver NG, Jonas BA, Pollyea DA. Optimizing survival outcomes with post-remission therapy in acute myeloid leukemia. *Am J Hematol*, 2019 Jul; 94(7): 803-811.
- Hartmann L, Metzeler KH. Clonal hematopoiesis and preleukemia-Genetics, biology, and clinical implications. *Genes Chromosomes Cancer*, 2019 Dec; 58(12): 828-838.
- Boddu PC, Zeidan AM. Myeloid disorders after autoimmune disease. *Best Pract Res Clin Haematol*, 2019 Mar; 32(1): 74-88.
- Liu XJ, Huang XJ, Xu LP, Liu KY, Zhang XH, Yan CH, Wang Y. [Effects of pre-transplant course on prognosis of allogeneic hematopoietic stem cell transplantation in patients with acute myeloid leukemia]. *Zhonghua Xue Ye Xue Za Zhi*, 2019 Mar 14; 40(3): 182-186.
- Leisch M, Jansko B, Zaborsky N, Greil R, Pleyer L. Next Generation Sequencing in AML-On the Way to Becoming a New Standard for Treatment Initiation and/or Modulation? *Cancers (Basel)*, 2019 Feb 21; 11(2).
- Dong XY, Li YL, Jiang L, Wu CY, Shang BJ, Zhang L, Cheng W, Zhu ZM. [Correlation between myeloperoxidase expression and gene alterations and prognosis in acute myeloid leukemia]. *Zhonghua Xue Ye Xue Za Zhi*, 2019 Jan 14; 40(1): 40-45.
- Schmid C, Labopin M, Schaap N, Veelken H, Schleuning M, Stadler M, Finke J, Hurst E, Baron F, Ringden O, Bug G, Blaise D, Tischer J, Bloor A, Esteve J, Giebel S, Savani B, Gorin NC, Ciceri F, Mohty M, Nagler A., EBMT Acute Leukaemia Working Party. Prophylactic donor lymphocyte infusion after allogeneic stem cell transplantation in acute leukaemia - a matched pair analysis by the Acute Leukaemia Working Party of EBMT. *Br J Haematol*, 2019 Mar; 184(5): 782-787.
- Nabhan C, Kamat S, Karl Kish J. Acute myeloid leukemia in the elderly: what constitutes treatment value? *Leuk Lymphoma*, 2019 May; 60(5): 1164-1170.
- Duan WB, Gong LZ, Jia JS, Zhu HH, Zhao XS, Jiang Q, Zhao T, Wang J, Qin YZ, Huang XJ, Jiang H. [Clinical features and early treatment effects in intermediate risk and poor risk acute myeloid leukemia with EVI1 positive]. *Beijing Da Xue Xue Bao Yi Xue Ban*, 2017 Dec 18; 49(6): 990-995.
- Lin M, Chen B. Advances in the drug therapies of acute myeloid leukemia (except acute wpromyelocytic leukemia). *Drug Des Devel Ther*, 2018; 12: 1009-1017.
- Schoen MW, Woelich SK, Braun JT, Reddy DV, Fesler MJ, Petruska PJ, Freter CE, Lionberger JM. Acute myeloid leukemia induction with cladribine: Outcomes by age and leukemia risk. *Leuk Res.*, 2018 May; 68: 72-78.
- Strickland SA, Shaver AC, Byrne M, Daber RD, Ferrell PB, Head DR, Mohan SR, Mosse CA, Moyo TK, Stricker TP, Vnencak-Jones C, Savona MR, Seegmiller AC. Genotypic and clinical heterogeneity within NCCN favorable-risk acute myeloid leukemia. *Leuk Res*, 2018 Feb; 65: 67-73.
- Fujiwara Y, Yamaguchi H, Yui S, Tokura T, Inai K, Onai D, Omori I, Marumo A, Yamanaka S, Sakaguchi M, Terada K, Nakagome S, Arai K, Kitano T, Okabe M, Okamoto M, Tamai H, Nakayama K, Tajika K, Wakita S, Inokuchi K. Importance of prognostic stratification via gene mutation analysis in elderly patients with acute myelogenous leukemia. *Int J Lab Hematol*, 2019 Aug; 41(4): 461-471.
- Niu P, Yao B, Wei L, Zhu H, Fang C, Zhao Y. Construction of prognostic risk prediction model based on high-throughput sequencing expression profile data in childhood acute myeloid leukemia. *Blood Cells Mol Dis.*, 2019 Jul; 77: 43-50.
- Wei A.H., Dohner H., Pocock C., Montesinos P., Afanasyev B., Dombret H., Ravandi F., Sayar H., Jang J.H., Porkka K., et al. Oral Azacitidine Maintenance Therapy for Acute Myeloid Leukemia in First Remission. *N. Engl. J. Med.*, 2020; 383: 2526-2537. doi: 10.1056/NEJMoa2004444.
- DiNardo C.D., Jonas B.A., Pullarkat V., Thirman M.J., Garcia J.S., Wei A.H., Konopleva M., Dohner H., Letai A., Fenaux P., et al. Azacitidine and Venetoclax in Previously Untreated Acute Myeloid Leukemia. *N. Engl. J. Med.*, 2020; 383: 617-629. doi: 10.1056/NEJMoa2012971.

19. Petersdorf S.H., Kopecky K.J., Slovak M., Willman C., Nevill T., Brandwein J., Larson R.A., Erba H.P., Stiff P.J., Stuart R.K., et al. A phase 3 study of gemtuzumab ozogamicin during induction and postconsolidation therapy in younger patients with acute myeloid leukemia. *Blood*, 2013; 121: 4854–4860. doi: 10.1182/blood-2013-01-466706.
20. Hills R.K., Castaigne S., Appelbaum F.R., Delaunay J., Petersdorf S., Othus M., Estey E.H., Dombret H., Chevret S., Ifrah N., et al. Addition of gemtuzumab ozogamicin to induction chemotherapy in adult patients with acute myeloid leukaemia: A meta-analysis of individual patient data from randomised controlled trials. *Lancet Oncol*, 2014; 15: 986–996. doi: 10.1016/S1470-2045(14)70281-5.
21. Feldman E.J., Lancet J.E., Kolitz J.E., Ritchie E.K., Roboz G.J., List A.F., Allen S.L., Asatiani E., Mayer L.D., Swenson C., et al. First-in-man study of CPX-351: A liposomal carrier containing cytarabine and daunorubicin in a fixed 5:1 molar ratio for the treatment of relapsed and refractory acute myeloid leukemia. *J. Clin. Oncol*, 2011; 29: 979–985. doi: 10.1200/JCO.2010.30.5961.
22. ROLLIG C., KRAMER M., SCHLIEMANN C., MIKESCH J.H., STEFFEN B., KRAMER A., NOPPENY R., SCHAFFER-ECKART K., KRAUSE S.W., HANEL M., et al. Does time from diagnosis to treatment affect the prognosis of patients with newly diagnosed acute myeloid leukemia? *Blood*, 2020; 136: 823–830. doi: 10.1182/blood.2019004583.
23. Juliusson G., Hagberg O., Lazarevic V.L., Lehmann S., Høglund M. Impact of treatment delay in acute myeloid leukemia revisited. *Blood Adv.*, 2021; 5: 787–790. doi: 10.1182/bloodadvances.2020003806.
24. Thol F. What to use to treat AML: The role of emerging therapies. *Hematol. Am. Soc. Hematol. Educ. Program*, 2021; 2021: 16–23. doi: 10.1182/hematology.2021000309.
25. Chen X., Xie H., Wood B.L., Walter R.B., Pagel J.M., Becker P.S., Sandhu V.K., Abkowitz J.L., Appelbaum F.R., Estey E.H. Relation of clinical response and minimal residual disease and their prognostic impact on outcome in acute myeloid leukemia. *J. Clin. Oncol*, 2015; 33: 1258–1264. doi: 10.1200/JCO.2014.58.3518.