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Review

Lipoic acid a multi-level molecular inhibitor of tumorigenesis

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ABSTRACT

We discuss how lipoic acid (LA), a natural antioxidant, induces apoptosis and inhibits proliferation, EMT, metastasis and stemness of cancer cells. Furthermore, owing to its ability to reduce chemotherapy-induced side effects and chemoresistance, LA appears to be a promising compound for cancer treatment.

1. Introduction

Lipoic acid (LA) is a natural antioxidant compound which is present in all prokaryotic and eukaryotic cells. It is synthesized in small amounts by plants and animals, including humans. In human beings, LA acts as an essential co-factor for several mitochondrial multi-enzyme complexes involved in energy metabolism, such as the pyruvate dehydrogenase (PDG) and α-ketoglutarate dehydrogenase (KGDG) complexes. LA is able to scavenge oxygen species and to regenerate other antioxidants, and is thus frequently used as a treatment for oxidative stress-associated pathologies, like diabetes, atherosclerosis, liver and neurodegenerative diseases. Beyond its efficacy in treating these chronic diseases, researchers have demonstrated its positive effects in various types of cancers. In the current review we will summarize the anti-cancer properties of LA and highlight its effective role in reducing chemotherapy-induced side effects and in preventing chemoresistance.

2. The effects of lipoic acid on proliferation and apoptosis

Various studies have shown that the exogenous administration of LA inhibits proliferation in several type of cancers. It is well known that tumor proliferation is due to the overexpression of different tyrosine kinase receptors (TKRs), including the epidermal growth factor receptor (EGFR), leading to the activation of oncogenic signaling pathways such as PI3K/Akt, ERK and mTOR. In this latter case, or Interestingly, the downstream effectors of these proliferative pathways are inhibited upon LA treatment in several types of cancers contributing to its anti-tumor activity by the restriction of cancer cell proliferation [1,2]. Taken together, LA plays a crucial anti-proliferative role by targeting EGFR signaling pathways. Further studies are necessary to unveil other TKRs affected by LA, such as the insulin-like growth factor receptor 1 (IGF1R) (Fig. 1). Moreover, it has been demonstrated that LA targets some protein tyrosine phosphatases which are implicated in tumorigenesis. Indeed, in breast cancer cells, LA was shown to decrease their viability by reducing the activity PTP1B and SHP2 which are often over-expressed in breast cancer cells and present potential targets for anticancer therapy [3]. Moreover, LA fosters the upregulation of the cyclin-dependent kinase inhibitors $p27^{kip1}$ and $p21^{Cip1}$ in many types of cancers leading to cell cycle arrest [4] (Fig. 1). AMPK is a major sensor of energy level that maintains the energy homeostasis. Its activation by its phosphorylation has been described to restrict tumor progression by inhibiting the mTOR protein complex which is a downstream Akt effector. In addition to its inhibitory role, LA leads to strengthen the AMPK activation and then reinforces the inhibition of the Akt pathway thereby reducing cancer cell proliferation [5–8].

In addition to its anti- proliferative role, LA induces apoptosis of different types of cancer cells in a dose-dependent manner. Previous data have shown that LA is able to generate ROS, which promote apoptosis in a variety of cancer cell lines by deregulating the ratio between anti-apoptotic and pro-apoptotic proteins. Indeed, the

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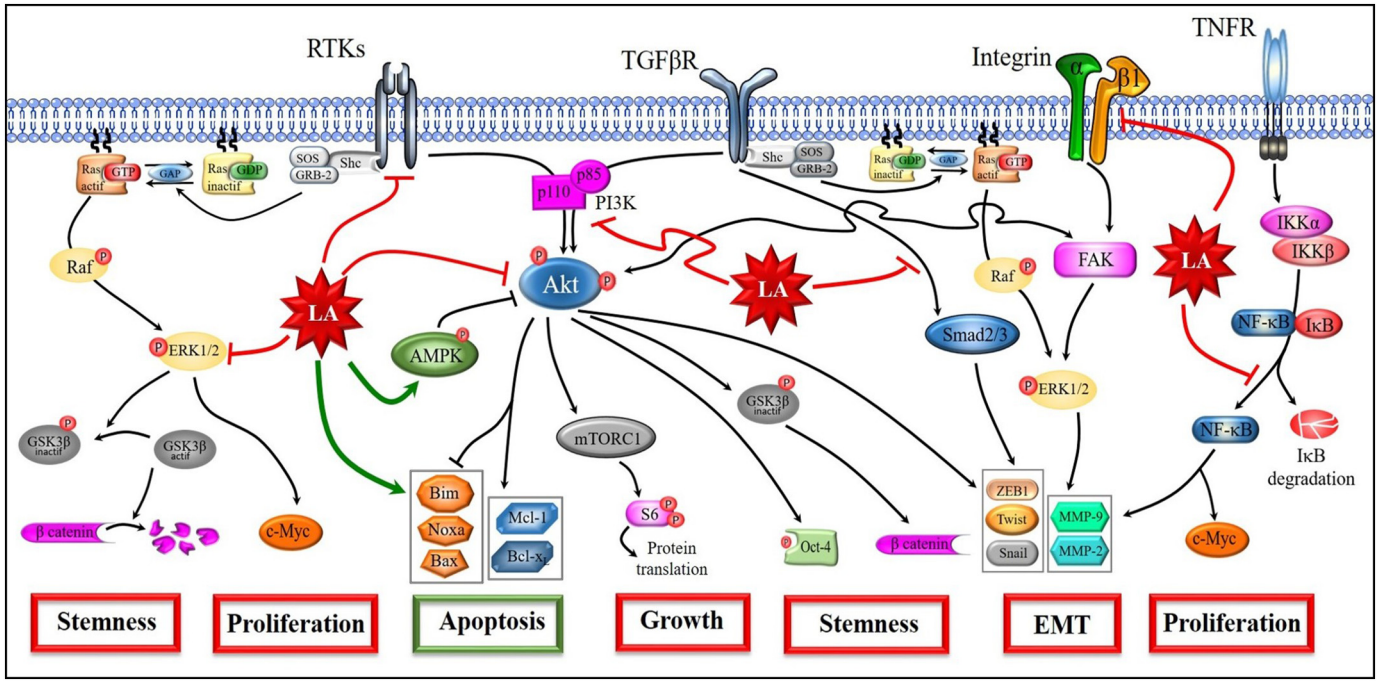

Fig. 1. Effects of lipoic acid (LA) on several signaling pathways implicated in tumorigenesis. Stemness is notably induced by activation of the Akt pathway, which in turn leads to the phosphorylation and inactivation of GSK3, resulting in the stabilization of β-catenin and activation of Oct-4 by phosphorylation. These events play a crucial role in stemness. LA inhibits the activation of Akt by reducing its phosphorylation, thus preventing the stemness process. **Cellular proliferation and growth** are controlled by the activation of ERK and PI3K/Akt pathways. LA prevents the phosphorylation of ERK, in particular by blocking the activation of c-Myc which is a centerpiece of the several processes implicated in cancer development. Moreover, LA inhibits the activation of Akt leading to the inhibition of mTORC1 which induces the translational process, thus LA reduces cellular proliferation and growth. **Apoptosis** is controlled by two subtypes of proteins, pro- or anti-apoptotic proteins. Phosphorylation of Akt provokes the induction of anti-apoptotic proteins (e.g. Mcl-1, bcl-x_L) and the reduction of pro-apoptotic proteins (e.g. Bim, Nova, Bax). LA acts at three different levels: (i) It inhibits the Akt pathway causing a repression of anti-apoptotic proteins and an increase in pro-apoptotic proteins, (ii) it induces the transcription of pro-apoptotic proteins by the generation of ROS, and (iii) it actives the AMPK protein which negatively regulates the Akt pathway. Moreover, LA impedes the activation of PI3K as well as some TKRs, such as EGFR, which are upstream of the Akt or ERK pathways, reinforcing the inhibition of these pathways. In addition, LA suppresses the downstream oncogenic pathways activated by TNFα by inhibiting **NF-κB** signaling which is a critical promoter of tumorigenesis. All of these events favor the inhibition of proliferation, growth and the induction of apoptosis. The **EMT process** leading to cell migration and invasion is controlled by several proteins (e.g. ZEB1) and enzymes (e.g. MMPs). LA inhibits FAK activation by downregulating β1/β3-integrin expression. This action prevents ERK activation, and reduces the mRNA levels of MMP-9 and -2. In addition, LA inhibits the Smad signaling which is a main signal transducer for receptors of the transforming growth factor beta (TGFβ) superfamily, which are essential for regulating growth and EMT. (Green arrows and frame reveal activation and red arrows and frames reveal inhibition). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)
AMPK: AMP-activated Protein Kinase; Akt: Protein Kinase B, Bax: Bcl-2-associated X protein; Bcl-2: B-cell lymphoma 2; c-Myc: Cellular Myelocytomatosis; ERK: Extracellular signal - Regulated Kinase; FAK: Focal Adhesion Kinase; GSK3β: Glycogen synthase kinase 3 beta; LA: Lipoic Acid; Mcl-1: myeloid cell leukemia 1; MMP: Matrix Metalloproteinase Protein; mTORC1: mammalian Target Of Rapamycin Complex 1; Oct-4: Octamer-binding transcription factor 4; PI3K: Phosphoinositide 3-kinase, S6: ribosomal protein S6; TGFβR: Transforming Growth Factor Beta Receptor; TKRs: Tyrosine Kinase Receptors.

mitochondrial anti apoptotic proteins Mcl-1, bcl-2 and bcl-X_L, are downregulated in a concentration-dependent manner following LA treatment in several ovarian lung and breast cancer cell lines [9]. Contrariwise, the expression of the pro-apoptotic proteins Bim and Noxa is upregulated in response to LA. All these events lead to cell death [10] (Fig. 1). In leukemia and breast cancer, the Bax/bcl2 ratio (apoptotic index) increases significantly after LA treatment with increased caspase-3 activity [11]. Another study performed in hepatoma cancer cells also showed that LA triggered the intrinsic apoptotic pathway by the activation of caspase-9 and caspase-3 [9]. In line with these findings, previous study has been done in colon cancer revealed that LA stabilizes the WT p53 protein by inhibiting the NF-κB signaling (Fig. 1). It is known that WTp53 is an activator of apoptosis by inducing the transcription of pro-apoptotic genes thereby triggering intrinsic apoptosis. However, Dörsam et al. demonstrated that LA triggered cell death in colorectal cancer independently of p53 stabilization [12]. Furthermore, a previous work on lung cancer cells suggested that LA treatment provoked other cell death pathways by caspase-independent mechanisms mediated by the Apoptosis-Inducing Factor (AIF) leading to necrosis and autophagy-associated cell death [13].

3. The impact of lipoic acid on metastasis, invasion, migration and EMT

The metastasis cascade is a complex multi-step process, the first critical step being cell invasion through extracellular barriers and penetration of the basement membrane, followed by infiltration of the vasculature, extravasation and colonization of distal organs. These events are regulated by two crucial pathways, namely the transforming growth factor β (TGFβ) and the focal adhesion kinase (FAK). Once these pathways are activated, epithelial-mesenchymal transition (EMT)-related transcription factors (TFs) such as vimentin, Slug, Twist and Snail are induced. These TFs promote EMT by downregulating E-cadherin and by triggering the overexpression of several EMT markers.

In two breast cancer cell lines, namely MDA-MB-231 and 4T1, a recent study demonstrated that LA inhibits cell migration by repressing TGFβ-induced EMT markers such as Snail, vimentin and Zeb1 [2]. The expression of these markers is also reduced upon LA treatment in other types of cancers [14] (Fig. 1). Moreover, the Smad pathway, known to be implicated in the EMT process after its activation by TGFβ signaling, is inhibited by LA [1]. Collectively, these results indicate that LA suppresses invasion and migration by inhibiting the EMT (Fig. 1). In

addition, during cancer cell invasion, small projections called invadopodia facilitate cell adhesion to the extracellular matrix (ECM) in an integrin-dependent manner. These cell surface proteins are known to activate the FAK pathway. LA treatment impairs the formation of these projections due to the downregulating of β1-integrin expression, leading to the inhibition of cancer cell invasion and migration [15] (Fig. 1). Invasion also relies on metalloproteinases (MMPs), since these key enzymes, mainly MMP-2 and MMP-9, degrade type IV collagen and play crucial roles in tumor cell invasion and malignancy [16], and once again LA was shown to inhibit invasion by reducing MMP-2 and MMP-9 mRNA expression in a dose-dependent manner in breast cancer cell lines [16] (Fig. 1). Consistent with these findings, LA also inhibits the EMT process by increasing E-cadherin and decreasing of N-cadherin, vimentin and activated β-catenin.

4. Lipoic acid reduces stemness properties

Cancer stem cells (CSCs) are rare immortal cells able to give rise to multiple cell types constituting the tumor, due to their self-renewal and differentiation capacities. This population is able to induce cell proliferation and dissemination. Despite advances in therapies targeting the tumor bulk, CSCs could remain unaffected, promoting cancer relapse and therapeutic drug resistance. The CSC phenotype is maintained through the activation of the Akt pathway which is important for the stabilizing CSC regulatory proteins, such as Oct-4, which promotes the transcription of Nanog, a key protein sustaining stemness, aggressiveness and chemoresistance of various cancer cell types. A recent study revealed that LA negatively regulates CSC-like phenotypes of human non-small cell lung cancer-derived cells by suppressing the Akt signaling pathway, leading to the proteosomal degradation of Oct-4 [14]. This study demonstrated the role of LA in suppressing CSC phenotypes and reinforces the crucial implication of LA in overcoming CSC-mediated chemoresistance, progression and metastasis (Fig. 1).

5. Lipoic acid enhances the cytotoxicity of chemotherapy and prevents their side effects

Despite advances in conventional therapies, mechanisms of resistance including the generation of DNA damage and free radicals, leading to the growth of secondary tumors, remain the underlying causes of morbidity and mortality. Several studies have shown that LA sensitizes cancer cells to chemotherapeutic agents. Notably, LA increases the paclitaxel efficiency in breast and lung cancer cells by inhibiting NF-κB signaling and integrin β1/β3, respectively [17,18] (Fig. 1). Similarly, a recent study has shown that co-loading lipoic acid and Docetaxel using solid lipid nanoparticles (DTX-ALA co-loaded SLN) increases apoptosis as compared to single drug-loaded SLNs (DTX -loaded SLN or ALA loaded SLN) in breast cancer cells [19]. Another study showed that LA potentiates the cytotoxicity of two anti-cancer agents used in colorectal cancer (5-fluorouracil, Temozolomide) which have different mechanisms of action. Likewise, LA overcomes gefitinib resistance by reducing the activation of growth factor receptors in non-small cell lung cancer cells [20]. Owing to these studies, it would be interesting to combine LA with anticancer drugs in order to improve their efficiency. In addition to its potential role in enhancing the cytotoxicity of chemotherapy, LA has a preventing role against the chemotherapy side effects by protecting patients against neuropathy, intestinal damage and diarrhea. In this context, pretreatment with LA before cisplatin administration can overcome cisplatin-induced damages such as nephrotoxicity, neurotoxicity and ototoxicity by restoring the redox system [21].

6. Conclusions

Hence, LA exhibits anti-tumor activities in several cancer models, by impacting several hallmarks on most of the signaling pathways implicated in proliferation, invasion, migration, EMT, stemness and apoptosis. Its mechanisms of action are so far not completely elucidated, and further investigations are needed to improve our understanding of these underlying anti-cancer effects. Another ongoing challenge is to prove its effectiveness in vivo in combination with chemotherapeutic agents. These additional studies may provide crucial data for the design of combinatorial LA therapies in order to increase its efficacy and to restore cancer cell chemo-sensitivity. LA could thus be a promising molecule to improve anti-cancer therapies.

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Declaration of Competing Interest

None

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References

[1] M.J. Jeon, W.G. Kim, S. Lim, H.-J. Choi, S. Sim, T.Y. Kim, et al., Alpha lipoic acid inhibits proliferation and epithelial mesenchymal transition of thyroid cancer cells, Mol. Cell. Endocrinol. 419 (2016) 113–123.
[2] J. Tripathy, A. Tripathy, M. Thangaraju, M. Suar, S. Elangovan, α-Lipoic acid inhibits the migration and invasion of breast cancer cells through inhibition of TGFβ signaling, Life Sci. 207 (2018) 15–22.
[3] A. Kuban-Jankowska, M. Gorska-Ponikowska, M. Wozniak, Lipoic acid decreases the viability of breast cancer cells and activity of PTP1B and SHP2, Anticancer Res. 37 (2017) 2893–2898.
[4] E. Dozio, M. Ruscica, L. Passafaro, G. Dogliotti, L. Steffani, P. Marthyn, et al., The natural antioxidant alpha-lipoic acid induces p27(Kip1)-dependent cell cycle arrest and apoptosis in MCF-7 human breast cancer cells, Eur. J. Pharmacol. 641 (2010) 29–34.
[5] S. Park, S.K. Choi, Y. Choi, H.-S. Moon, AMPK/p53 Axis is essential for α-Lipoic acid-regulated metastasis in human and mouse colon cancer cells, J. Investig. Med. 63 (2015) 882–885.
[6] W. Li, S.M. Saud, M.R. Young, G. Chen, B. Hua, Targeting AMPK for cancer prevention and treatment, Oncotarget. 6 (2015) 7365–7378.
[7] K.-G. Park, A.-K. Min, E.H. Koh, H.S. Kim, M.-O. Kim, H.-S. Park, et al., Alpha-lipoic acid decreases hepatic lipogenesis through adenosine monophosphate-activated protein kinase (AMPK)-dependent and AMPK-independent pathways, Hepatol. Baltim Md. 48 (2008) 1477–1486.
[8] W.J. Lee, K.-H. Song, E.H. Koh, J.C. Won, H.S. Kim, H.-S. Park, et al., Alpha-lipoic acid increases insulin sensitivity by activating AMPK in skeletal muscle, Biochem. Biophys. Res. Commun. 332 (2005) 885–891.
[9] B. Dörsam, J. Fahrer, The disulfide compound α-lipoic acid and its derivatives: a novel class of anticancer agents targeting mitochondria, Cancer Lett. 371 (2016) 12–19.
[10] P. Kafara, P. Icard, M. Guillamin, L. Schwartz, H. Lincet, Lipoic acid decreases Mcl-1, Bcl-xL and up regulates Bim on ovarian carcinoma cells leading to cell death, J. Ovarian Res. 8 (2015) 36.
[11] M.H. Na, E.Y. Seo, W.K. Kim, Effects of alpha-lipoic acid on cell proliferation and apoptosis in MDA-MB-231 human breast cells, Nutr. Res. Pract. 3 (2009) 265–271.
[12] B. Dörsam, A. Göder, N. Seiwert, B. Kaina, J. Fahrer, Lipoic acid induces p53-independent cell death in colorectal cancer cells and potentiates the cytotoxicity of 5-fluorouracil, Arch. Toxicol. 89 (2015) 1829–1846.
[13] S.Y. Choi, J.H. Yu, H. Kim, Mechanism of alpha-lipoic acid-induced apoptosis of lung cancer cells, Ann. N. Y. Acad. Sci. 1171 (2009) 149–155.
[14] P.P. Phiboonchaiyanan, P. Chanvorachote, Suppression of a cancer stem-like phenotype mediated by alpha-lipoic acid in human lung cancer cells through down-regulation of β-catenin and Oct-4, Cell Oncol. Dordr. 40 (2017) 497–510.
[15] M. Yamasaki, M. Iwase, K. Kawano, Y. Sakakibara, M. Suiko, M. Ikeda, et al., α-Lipoic acid suppresses migration and invasion via downregulation of cell surface β1-integrin expression in bladder cancer cells, J. Clin. Biochem. Nutr. 54 (2014) 18–25.
[16] H.S. Lee, M.H. Na, W.K. Kim, Alpha-Lipoic acid reduces matrix metalloproteinase activity in MDA-MB-231 human breast cancer cells, Nutr. Res. N Y N 30 (2010) 403–409.
[17] B.J. Li, X.Y. Hao, G.H. Ren, Y. Gong, Effect of lipoic acid combined with paclitaxel on breast cancer cells, Genet. Mol. Res. GMR 14 (2015) 17934–17940.

[18] P. Puchsaka, C. Chaotham, P. Chanvorachote, α-Lipoic acid sensitizes lung cancer cells to chemotherapeutic agents and anoikis via integrin β1/β3 downregulation, Int. J. Oncol. 49 (2016) 1445–1456 Internet. cited 2019 Feb 15. Available from http://www.spandidos-publications.com/ijo/49/4/1445/abstract.

[19] I.R. Kothari, S. Mazumdar, S. Sharma, K. Italiya, A. Mittal, D. Chitkara, Docetaxel and alpha-lipoic acid co-loaded nanoparticles for cancer therapy, Ther. Deliv. 10 (2019) 227–240.

[20] H. Michikoshi, T. Nakamura, K. Sakai, Y. Suzuki, E. Adachi, S. Matsugo, et al., α-Lipoic acid-induced inhibition of proliferation and met phosphorylation in human non-small cell lung cancer cells, Cancer Lett. 335 (2013) 472–478.

[21] K.-H. Kim, B. Lee, Y.-R. Kim, M.-A. Kim, N. Ryu, D.J. Jung, et al., Evaluating protective and therapeutic effects of alpha-lipoic acid on cisplatin-induced ototoxicity, Cell Death Dis. 9 (2018) 827.