Drug Resistance Updates 24 (2016) 23–33

Contents lists available at ScienceDirect

Drug Resistance Updates

journal homepage: www.elsevier.com/locate/drup

Lysosomes as mediators of drug resistance in cancer

Benny Zhitomirsky, Yehuda G. Assaraf*

The Fred Wyszkowski Cancer Research Laboratory, Department of Biology, Technion-Israel Institute of Technology, Haifa, Israel

ARTICLE INFO

Article history:
Received 3 September 2015
Received in revised form 4 November 2015
Accepted 19 November 2015

Keywords:
cancer
Chemotherapeutics
Multidrug resistance
Lysosomes
Drug sequestration
Drug transport

ABSTRACT

Drug resistance remains a leading cause of chemotherapeutic treatment failure and cancer-related mortality. While some mechanisms of anticancer drug resistance have been well characterized, multiple mechanisms remain elusive. In this respect, passive ion trapping-based lysosomal sequestration of multiple hydrophobic weak-base chemotherapeutic agents was found to reduce the accessibility of these drugs to their target sites, resulting in a markedly reduced cytotoxic effect and drug resistance. Recently we have demonstrated that lysosomal sequestration of hydrophobic weak base drugs triggers TFEB-mediated lysosomal biogenesis resulting in an enlarged lysosomal compartment, capable of enhanced drug sequestration. This study further showed that cancer cells with an increased number of drug-accumulating lysosomes are more resistant to lysosome-sequestered drugs, suggesting a model of drug-induced lysosome-mediated chemoresistance. In addition to passive drug sequestration of hydrophobic weak base chemotherapeutics, other mechanisms of lysosome-mediated drug resistance have also been reported; these include active lysosomal drug sequestration mediated by ATP-driven transporters from the ABC superfamily, and a role for lysosomal copper transporters in cancer resistance to platinum-based chemotherapeutics. Furthermore, lysosomal exocytosis was suggested as a mechanism to facilitate the clearance of chemotherapeutics which highly accumulated in lysosomes, thus providing an additional line of resistance, supplementing the organelle entrapment of chemotherapeutics away from their target sites. Along with these mechanisms of lysosome-mediated drug resistance, several approaches were recently developed for the overcoming of drug resistance or exploiting lysosomal drug sequestration, including lysosomal photodestruction and drug-induced lysosomal membrane permeabilization. In this review we explore the current literature addressing the role of lysosomes in mediating cancer drug resistance as well as novel modalities to overcome this chemoresistance.

1. Introduction

Lysosomes are highly acidic, membrane bound, intracellular organelles, carrying a battery of hydrolytic enzymes as well as several membrane-associated proteins (Saftig, 2005). While all eukaryotic cells, except red blood cells, contain lysosomes, their structure and number vary depending on the cell type and functional state (Appelqvist et al., 2013; Saftig, 2005). Lysosomes are characterized by a highly acidic pH (pH ≤ 5), achieved by the activity of the vacuolar-type H^+-ATPase (V-ATPase), which enables optimal activity of various hydrolases characterized by an acidic pH optimum (Saftig, 2005). Lysosomal biogenesis was recently shown to be regulated by the coordinated lysosomal expression and regulation (CLEAR) gene network, which is activated by the master regulator E basic helix-loop-helix protein 35 (transcription factor EB, TFEB), upon its translocation into the nucleus (Palmieri et al., 2011; Sardiello et al., 2009; Settembre et al., 2012).

Although when first described in the 1950s, lysosomes were considered to be mere "waste bags" used by the cell to dispose of unwanted biomolecules, they are now known to partake in numerous key physiological processes including the breakdown of macromolecules and worn out organelles, endocytosis, autophagy, exocytosis, plasma membrane repair, homeostatic maintenance of several key metabolites including cholesterol and amino acids, as well as apoptosis (Aits and Jaattela, 2013; Appelqvist et al., 2013; Feng et al., 2014; Liu et al., 2012; Saftig, 2005). Accordingly, lysosomes were shown to play a major role in various diseases ranging from lysosomal storage diseases caused directly by lysosomal dysfunction due to mutations in lysosomal enzymes (Aronovich and Hackett, 2015), through cardiovascular disorders, affected by the elevated activity of lysosomal cathepsins (Appelqvist et al., 2013; Lutgens et al., 2007), to several increasingly apparent roles in various malignancies. In this regard, lysosomes were found to undergo diverse alterations associated with malignant transformation

* Corresponding author. Tel.: +972 4 8294211; Fax: +972 4 8225153.
E-mail address: assaraf@technion.ac.il (Y.G. Assaraf).

http://dx.doi.org/10.1016/j.drup.2015.11.004
1368-7646/

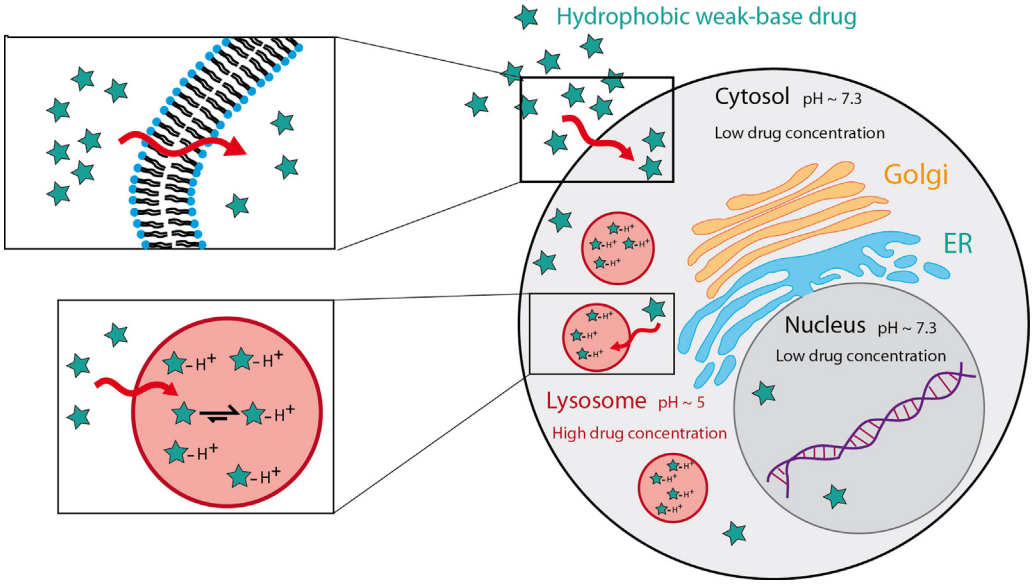

Fig. 1. *Passive lysosomal sequestration of hydrophobic weak-base drugs reduces their concentration at their target sites.* Hydrophobic weak-base compounds diffuse freely both across the plasma membrane and lysosomal membrane. Upon encountering the acidic lysosomal lumen, these drugs become protonated and are thus unable to traverse the lipid lysosomal membrane, leading to their accumulation in lysosomes. Lysosomal accumulation results in a reduced drug concentration in the cytoplasm and the nucleus.

including elevated expression of lysosomal proteins which drives tumor progression and metastasis, down-regulation of expression of lysosomal proteins, hence hindering lysosome-mediated apoptosis, as well as increased secretion of lysosomal enzymes which partake in tumor invasion and angiogenesis (Kallunki et al., 2013). In light of this correlation between lysosomes and cancer, lysosomes have been recently considered as a novel target for anticancer therapy. Consistently, several approaches for targeting cancer cell lysosomes have been suggested, including lysosomal photodestruction, targeting of lysosomal acid sphingomyelinase (ASM) and induction of lysosomal membrane permeabilization (LMP) (Adar et al., 2012; Appelqvist et al., 2013; Gyparaki and Papavassiliou, 2014; Saftig and Sandhoff, 2013).

Multidrug resistance (MDR) is a phenomenon in which cancer cells acquire resistance to multiple drugs which are structurally and functionally unrelated (Holohan et al., 2013; Szakacs et al., 2006). Various mechanisms of MDR, which can be either intrinsic or acquired following drug treatment, have been described, including: reduced cellular drug uptake, ATP-driven drug efflux from the cell, quantitative and qualitative alterations in target proteins, drug compartmentalization, drug metabolism as well as evasion of apoptosis (Assaraf, 2007, 2006; Gonen and Assaraf, 2012; Gottesman, 2002; Gottesman et al., 2002). Despite various approaches implemented in an attempt to overcome MDR, the latter continues to be the leading cause of chemotherapeutic treatment failure (Kunjachan et al., 2013; Livney and Assaraf, 2013).

Cumulative evidence indicate that lysosomes play an important role in MDR. It was demonstrated that lysosomes contribute to hydrophobic weak base chemotherapeutic drug resistance via a mechanism known as lysosomal sequestration (Adar et al., 2012; Gotink et al., 2011; Hrabeta et al., 2015; Jansen et al., 1999; Kaufmann and Krise, 2007; Zhitomirsky and Assaraf, 2015a). Lysosomal sequestration is a phenomenon based on cation trapping, which occurs when hydrophobic weak base drugs become entrapped in lysosomes due to their protonation in the acidic lumen of this organelle; thus, these sequestered drugs are unable to reach their target sites and therefore fail to exert their cytotoxic activity. Additional evidence point to other mechanisms of lysosome-mediated MDR, including transporter-mediated sequestration of various chemotherapeutics, as well as lysosomal exocytosis of sequestered drugs, suggesting that lysosomes and lysosomal proteins play a key role in the resistance to a wide spectrum of commonly used anticancer drugs. In the present paper we review and discuss the current literature in the field of lysosome-mediated cancer drug resistance, and the attempts to overcome this chemoresistance phenomenon.

2. Lysosomal sequestration of hydrophobic weak base chemotherapeutics

Lysosomal sequestration, also known as lysosomal trapping, is a process in which hydrophobic weak base compounds become highly accumulated in acidic lysosomes (MacIntyre and Cutler, 1988). Such substances travel freely across lipid membranes, including the plasma membrane and the lysosomal membrane, due to their hydrophobic nature. Upon encountering the acidic environment of the lysosomal lumen these compounds become protonated due to their weak base properties, and thus can no longer cross the lipid membrane of the lysosome, resulting in their marked lysosomal accumulation and compartmentalization (de Duve et al., 1974; Goldman et al., 2009; MacIntyre and Cutler, 1988) (Fig. 1). Several anticancer drugs have been shown to undergo lysosomal sequestration including sunitinib (SU11248, Sutent), doxorubicin (DOX, adriamycin), daunorubicin (DNR, cerubidine), vincristine (oncovin), mitoxantrone (novantrone), and various preclinical cytotoxic agents such as imidazoacridinones (Table 1) (Adar et al., 2012; Gong et al., 2006; Gotink et al., 2012; Groth-Pedersen et al., 2007; Herlevsen et al., 2007; Jansen et al., 1999; Kazmi et al., 2013; Smith et al., 1992; Zhitomirsky and Assaraf, 2015a). Lysosomal sequestration of hydrophobic weak base compounds in general, and of hydrophobic weak base anticancer drugs in particular, can have a significant impact on their subcellular distribution (Duvvuri and Krise, 2005; Kazmi et al., 2013). Since the target sites of anticancer drugs are seldom lysosomes or lysosomal proteins, drugs sequestered in the lysosome will not have access to their targets. Therefore, lysosomal compartmentalization of these cytotoxic agents was suggested to lower their concentration at the target sites, thereby hindering their ability to exert a cytotoxic effect (Duvvuri and Krise, 2005; Jansen et al., 1999). In support of this