The Current State and Future Prospects of Multidrug-Resistance in Cancer Cells

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Abstract

Drug resistance in cancer cells is a serious complication that is always continuously evolving. Rather than just one or two factors, drug resistance is a combination of a handful of elusive mechanisms. Many of these mechanisms and factors have been studied in the past, however new methods of analysis and treatment are being developed and tested rigorously. Along with new progress and breakthroughs, the pharmaceutical industry must also recognize the increasing expensive cost factor and its burden on cancer patients of the future. Ultimately, new treatment methods accompanied by cost-efficient analysis will provide patients with the best cancer treatment possible.

Introduction

Cancer arises in individuals through genetic mutations that eventually accumulate and result in uncontrollable growth. Over the years, drug resistance in cancer cells has become a serious threat to patients seeking treatment for their illness. According to the American Cancer Society, it is determined that roughly 562,340 Americans will die as a result of cancer in 2009 (Jemal, 2009). There are various ways that cancer has become so highly resistant to many of the anticancer drugs being used against them. Because it is so complex and aggressive in nature, cancer cells now have the ability to elude drugs targeting a specific mechanism, giving rise to multi-drug resistance (MDR) cancer. MDR cancer can be
intrinsic, which is present at diagnosis or extrinsic, when it is acquired from drug therapy treatment (Mijatovic, 2009).

The resistance to chemotherapeutic agents is one of the main factors that cause the failure of cancer treatment for 90% of patients (Wilson, 2009). There is no doubt now that researchers and scientists must work even harder to strive and find new ways to exploit any point of weakness in cancer cells. We have learnt the hard way that drug resistant cancer never stops evolving even under the pressure of a drug treatment (Hu, 2008).

There are currently new methods and techniques being looked into such as focusing on alternate drug targets, inducing necroptosis and using proteomics to better understand the resistant mechanisms of cancer cells. The “magic bullet”, or the “one drug to treat all”, conceptualized by Paul Ehrlich in 1908 does not seem too lively nowadays given the rise of cytotoxin resistant cancer. Instead, the idea of attacking alternate death pathways and promoting multiple death pathways simultaneously is on the rise and may finally be a hopeful remedy to MDR cancer. The anticancer drug of the future should ideally be one which increases survival rates with the least amount of side effects as possible and it should also be cost-efficient.

Mechanisms of Drug Resistance in Cancer Cells

MDR cancer cells have become increasingly more difficult to combat as they have several mechanisms that allow them to withstand almost all man-made efforts to destroy them. There are three main ways that cancer cells elude anticancer drugs that include altered influx and efflux pumps, drug inactivation and evasion of apoptosis (Dean, 2009).

Increased efflux and decreased influx is one of the main mechanisms that cancer cells use to overcome drugs. ABC transporters are part of a family of integral membrane proteins which are responsible for the transfer of many substrates across cell membranes by ATP usage (Rees, 2009). The protection of the human body’s stem cells from toxins, damage and death is enormously important as they
need to remain healthy and functional throughout the body’s life and expression of ABC transporters is the primary mechanism responsible for this role. However, these same ABC transporters have also been proven to play a significant role in MDR cancer cells (Dean, 2009). Cancer cells rely on these intercellular mechanisms that reduce the effectiveness of many cytotoxic anticancer drugs (Türk, 2009). ABC transporters translocate many compounds such as phospholipids, peptides, ions and amino acids just to name a few. There are currently 49 ABC transporter genes described in humans (Gimenez-Bonafe, 2009). An alteration of drug efflux by an ABC transporter protein like the three most noted proteins, P-glycoprotein (P-gp), multidrug resistance associated protein (MRP) and breast cancer resistant protein (BCRP) allow cancer cells to actively “pump out” drugs and show resistance to chemotherapy (Wilson 2009). The main physiological task of these pumps is to provide a mechanism for resistance to xenobiotic molecules (Türk, 2009).

P-gp was discovered about 30 years ago and is notorious as being one of the major factors that causes MDR cancer (Hu, 2008). P-gp is the most studied and well described ABC transporter to date. It plays a vital role in the human body as it maintains the cholesterol levels and distribution. However, it exports many other lipophilic molecules, which include anticancer drugs (Rees, 2009). It contains 12 transmembrane domains (TMDs) that bind to hydrophobic anticancer drug substrates and results in the activation of two ATP hydrolysis actions that pump the drug out of the cell then allows the cell to return to its normal state (Gimenez-Bonafe, 2009). MRP are found in cancers that rarely express P-gp such as in lung cancers and are structurally different from P-gp (Hu, 2008). Every member of the MRP family gain resistance to drugs because of their lipophilic anion pump. BCRP contains six TMDs that allow it to bind with drugs and pump them out of the cell. This protein has broad drug specificity and it is capable of transporting a high capacity of anticancer drug molecules (Gimenez-Bonafe, 2009). All three of these proteins mentioned above function to block the entrance of chemotherapeutical drugs into the cancer cell from reaching their targets (Hu, 2008).
Drug inactivation decreases the bioavailability of anticancer drugs to bind to its purposed intracellular target. Fluorouracil-5 (5-FU) is a pyrimidine analog chemotherapeutic drug that has been widely used for the last forty years since the 1950s (Han, 2008). 5-FU, and drugs alike, needs to become “active” in order to exert therapeutic effects on cancer cells. However, almost all of 5-FU undergo catabolism in the liver by dihydropyrimidine dehydrogenase (DPD). In cancer, over expression of DPD has been observed and the increased levels of DPD degrades effective 5-FU molecules and correlate to resistance to 5-FU (Wilson, 2009). Daunorubicin (DRC), considered one of the most effective anticancer drugs, had a significant decrease in effectiveness when the metabolic inactivation by carbonyl reduction took place. Plebuch (2007) found that over expression of DRC carbonyl reductase enzyme allowed cancer cells to survive the therapeutic effects of DRC.

Another promising drug oracin, was found to be victim of metabolic inactivation due to carbonyl reduction as well. Novotna (2008) revealed and confirmed that an aldo-keto reductase family related gene called AKR1C3 actually promotes the drug inactivation of oracin and believe that it plays a large role in one of the main mechanisms of drug resistance in cancer cells, drug inactivation.

Evasion of apoptosis is another mechanism cancer cells use in order to survive and evade our attempts to destroy them. Apoptosis is commonly known as programmed cell death and is described as the body’s way of regulating cell proliferation. Many of the genetic mutations that give rise to cancer cells also affect genes that are responsible for regulating cell cycle and apoptosis. Therefore, to induce apoptosis in cancer cells is the main goal of cytotoxic chemotherapy methods (MacKenzie, 2008). It has been found that cancer cells may lack expression of pro-apoptotic molecules or may over express anti-apoptotic molecules (La Porta, 2007). Tumour protein 53 (p53) has an important role of being a DNA-damage monitor, in that it has the ability to induce apoptosis when mutations are found. It has been studied that internal stressors such as DNA-damage, hypoxia, and oxidative stress activate p53 normally. When activated, p53 is able to act as a tumour suppressor by triggering apoptosis of the now defective cell. This process induces cysteine proteases family member caspases which are responsible for
destroying and clearing of cancer cells. However, p53 is inactive in all cancers allowing them to successfully evade apoptosis (Meulmeester & Jochemsen, 2008).

Drugs that induce apoptosis can also be inhibited by the expression of decoy receptors such as decoy receptor 3 (DcR3). It was found that DcR3 was over-expressed in 73% of colorectal cancer patients (Wilson, 2009). In rat models, expression of DcR3 demonstrated decreased infiltration of immunocompetent cells, proving that DcR3 can lead to interferences in the apoptotic process.

Future Prospects in Treating Cancer Patients:

Cancer cells have evolved to be drug and apoptotic resistant over the years and this has put a great deal of limitations on scientists and researchers trying to overcome it. Instead of trying to induce apoptosis, new pathways to target cell death are emerging. Yang (2009) proposes that a drug which induces non-apoptosis like death can possibly overcome drug resistance in cancer cells. Necrosis, in contrast with apoptosis, is viewed by many as unregulated or accidental cell death caused by stress which is morphologically distinct from apoptosis. Necroptosis includes inflammation, loss of membrane integrity and affects groups of cells as opposed to apoptosis’ more orderly and non-inflammatory symptoms. Necrosis is gaining much attention as a possible way to combat MDR cancer, especially apoptotic resistant cancer. This concept is now moving forward as scientists are characterizing molecular mechanisms that can induce necrosis.

Shikonin, a natural organic naphthoquinone (Yang, 2009), is a necroptotic inducer that has been shown to be affective against cancer cells that express P-gp, MRP, and BCRP (Hu, 2007). Shikonin induces cell death via pathway different from apoptosis. And since necroptosis is a pathway that is unique from apoptosis, all of the resistant cancers cells that relied on circumventing the apoptotic pathway now reveal this weak spot. Shikonin has been found to inhibit the growth of lung cancer with an effective rate of 63% (Yang, 2009). Shikonin’s efficacy to induce necroptosis was found to be neither affected by P-gp, MRP and BCRP drug transporter over expression (Hu, 2008). Proteasomes play a key role in cells
as they regulate homeostasis and in cancer patients, they start to lose the ability to maintain cellular homeostasis. A proteasome inhibitor is ideal in this situation as they can block the progression of cancers degradation of regulatory proteins, as they have anti-proliferative and pro-apoptotic effects (Fuchs, 2009). Researchers have a strong belief that shikonin is a proteasome inhibitor because of its chemical structure when it was analyzed and that this inhibition contributes to cell death (Yang, 2009). Along with Shikonin, three other drugs derived from natural compounds have been gaining attention in the development of future drug treatments. Ixabepilone, trabectedin, and temsirolimus, each with their own unique effects, see figure 1, were approved for use in 2007. As we can see, more natural products are starting to be either re-studied or new ones are being investigated in the field of oncology (Bailly, 2009).

Na+/K+-ATPase alpha subunits have recently been claimed to be alternate and important targets for cancer treatments (Mijatovic, 2009). Na+/K+-ATPase include ligands with cardiotonic steroids which have anti-proliferative abilities that may allow them to overcome MDR effectively (Lefranc, 2008). When treated with a 19-hydroxy-2”oxovoruscharin, a modified cardenolide steroid and currently in phase I trial, cancer cells have demonstrated that they are unable to become resistant to this compound (Mijatovic, 2009). Na+/K+-ATPase is primarily an ion transporter. However, it is also involved in the migration mechanism of cancer (Lefranc, 2008). 19-hydroxy-2”oxovoruscharin has been found to be effective in human cancer cells from different locations of the body like breast, lung and colon cancer with resistance to different chemotherapeutic drugs (Mijatovic, 2009). If 19-hydroxy-2”oxovoruscharin can enhance apoptotic activity and decrease cancer cell migration, then this compound will definitely be an ideal anticancer candidate in the future.

Activating multiple death pathways may ultimately be the method of choice for combating MDR cancer cells. As discussed prior in this review, current approaches against MDR cancer include inhibiting drug transporters or reactivating the apoptosis process. However, Hu (2008) describes cancer as “complex, dynamic and elusive” implying that in order to destroy cancer cells, targeting more than one death pathway is necessary to be completely effective instead of only killing a percentage and allowing
the rest to gain more resistance and re-grow, see figure2. In either the laboratory or clinical setting, it is often very difficult to determine exactly what mechanism is active in causing MDR cancer growth (Zhang, 2007). Hu (2008) proposes that bypassing MDR in cancer is possible by activating multiple death pathways such as apoptosis, drug targeting, and necroptosis at the same time.

Proteomics, which incorporates gel electrophoresis, mass spectrometry and vast databases, are used to study the function and structure of proteins. It is now being used as a diagnostic tool to confirm known mechanisms and identify new mechanisms of resistance in cancer cells. Because proteins are released by tumour cells, clinicians are then able to measure and compare the serum protein profile from the normal range in an individual without cancer (Jain, 2008). In a recent study using proteomics, it was demonstrated and confirmed that multiple mechanisms were responsible for drug resistance in any single cancer cell line (Zhang, 2007).

Lung cancer makes up around one third of cancer deaths in the world according to the American Cancer Society followed closely by breast and prostate cancer (Jemal, 2009). As with many cancers, lung cancer has a poor prognosis in its late stages, which is why it is so important to develop early and relatively simple screening procedures. One way of early detection is to look at cancer biomarkers through proteomics. It was recently found that poorly differentiated lung carcinoma cells presented indications of changes in protein expression at the DNA level which happen to protect the cancer cells from apoptotic signals (Keenan, 2009). Further investigation with proteomics and the understanding of the correlation between drug resistant cancer and protein biomarkers may one day provide valuable methods of quick and early detection procedures for cancer patients and determine which patients are suitable candidates for certain treatments and ones who are not.

**The Price to Consider**

Generally speaking, if a single cause of resistance is not known, multiple anticancer drugs can be administered at the same time in order to “hit all the targets” leading to a much higher chance of death for
the cancer cells. This method may be more effective, but it is not the most cost-efficient method to treat cancer, as it wastes resources and ends up being very expensive on the patient’s behalf. Sharkey and Goldenberg (2006) looked at the cost of certain anticancer drugs and found that bevacizumab, which blocks the angiogenic ability of cancer cells and cetuximab, which impairs proliferation of cancer cells, comes to $5,000 and $12,000 respectively per month of treatment, a price that many cannot afford. For many cancer patients, they are not fully cured as drugs can only provide a moderate amount of survival. In a recent study, it was found that metastatic colorectal cancer patients do demonstrate increased life expectancies when they are administered a combination of therapeutic agents. However, these good results came at a high cost of $100,000 for every “discounted life-year” or extra year recovered via prescribed treatment (Wong, 2009).

Prevention

Even though many breakthroughs in detection and treatment are currently being developed and as the majority of this article has been focused on those, we must not lose sight of the main arsenal of combating cancer, which is to prevent cancer in the first place. In a recent study, it was found that about half of the major cancers are avoidable if awareness of unhealthy lifestyle risk factors such as smoking, obesity, exercise, alcohol abuse and infection were significantly increased in the general population (Redeker, 2008). Therefore promoting healthier lifestyles targeted to different demographics is a heightened public health priority in order to address individualized community issues (Sanderson, 2008). It is also noted that those whom are less fortunate are not as aware of the many risk factors as they have poorer education and hygiene health practices (Redeker, 2008). Therefore, more effort from the government and organizations should be focused into educating the less fortunate and general public about cancer risk factors. Obviously, preventing cancer from beginning to proliferate in an individual is the best choice health-wise, emotionally and economically.
Conclusion

Many of the mechanisms of cancer drug resistance have now been studied, identified and characterized. However, resistance is a forever evolving process and therefore, we must still try to anticipate cancer’s next move. Scientists and researchers do this by developing new methods of detection and identification such as proteomics, finding new target sites like Na+/K+-ATPase alpha subunits and studying new drugs like that induce necroptosis rather than apoptosis. The exploration of alternate pathways to overcome cancer cells is a must in these times of increasing cases and deaths. With the current rigorous research being performed, it seems that there is a somewhat universal agreement that combining treatments may ultimately be the best way to go in order to maximize effectiveness in helping cancer patients.

The future for those who do not have cancer yet seems very promising as the rapid paced research in detection methods such as proteomics may be at the forefront of preventing cancer cells from proliferating and ideally destroying them before new resistant lines have a chance to evolve and metastasise to other locations of the body. It seems more prevalent that we will stand a better chance in the future of preventing cancer rather than fully curing complex late or end stage cancer cells in individuals. This new emerging field of screening will play a vital and critical role in the future of cancer treatment.

But as new treatment options become available and longer survival rates increase, the cost that patients have to pay will also rise significantly. Therefore, assessments on cost effectiveness should also be incorporated in future treatment studies. A holistic approach that includes the patient’s cancer history, current stage of cancer, and cost analyses of treatments will present a cancer patient with the “big picture” of what he or she is currently going through as well as presenting viable options for the future.
Acknowledgements

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### Figure 1. Characteristics of Currently Investigated Natural Anti-Cancer Products

<table>
<thead>
<tr>
<th>Compound Name</th>
<th>Derived From</th>
<th>Effects</th>
<th>Effects (Cont.)</th>
<th>Approval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ixabepilone</td>
<td><em>Sorangium cellulosum</em></td>
<td>Arrests tumour cell division</td>
<td>Decreased susceptibility to resistance because of P-gp overexpression</td>
<td>FDA approved in October 2007 for infusion for breast cancer</td>
</tr>
<tr>
<td>Trabectedin</td>
<td><em>Ecteinascidia turbinata</em></td>
<td>Functions as a DNA alkylating agent, thus being a challenge to the DNA repair mechanism of cancer cells</td>
<td>Alkylation results in the bending of DNA. This affects multiple transcription factors that take part in cell proliferation.</td>
<td>FDA and EMEA approved in 2008 for ovarian cancer</td>
</tr>
<tr>
<td>Temsirolimus</td>
<td><em>Streptomyces hygroscopicus</em></td>
<td>Inhibition of the protein kinase mTOR, thus arresting cancer proliferation and angiogenesis</td>
<td>Includes anti-inflammatory and anti-tumor properties</td>
<td>FDA and EMEA approved May 2007 and November 2007 respectively for renal cancer</td>
</tr>
</tbody>
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### Figure 2. Targeting Multiple Death Pathways to Completely Eliminate Cancer Cells

![Diagram showing apoptosis and necrosis pathways](image)
References:


