Abstract

Taxanes are cytostatic routinely used for the treatment of solid breast, ovarian, prostate, head and neck tumors and other types of tumors. Resistance of tumor cells to the effect of taxanes represents serious obstacle for the employment of taxanes in the treatment of tumors. This resistance can be associated, among other things, with lower rate of apoptosis induction in cancer cells or also with increased level of transporters transporting taxanes out of the cell.

In this PhD thesis we tried: (1) to contribute to elucidation of the role of molecular mechanisms of apoptosis induction by taxanes in cells of human breast cancer. Specifically, it meant to contribute to elucidation of the role of initiator caspase -8 a -9 and mainly of initiator caspase-2. Next, to contribute to elucidation of the role of executioner caspase -3 - 6, and -7 and selected proteins of the Bcl-2 family. (2) To contribute to elucidation of molecular mechanisms of resistance of human breast cancer cells to taxanes. Specifically, it meant to describe the role of selected functional groups in taxane structure in bringing about and overcoming resistance to taxane and next to contribute to elucidation of the role of P-glycoprotein (ABCB1 transporter) in the resistance to individual taxanes.

1) We found that caspase-2 represents apical caspase in apoptosis induction by taxanes in breast cancer cells originated from mammary gland. Protein p53 as well as PIDDosome complex do not play crucial role in the activation of caspase-2 in these cells. It is possible that caspase-2 is activated due to its transport into the cytosol after nuclear envelope disintegration. Activated caspase-2 subsequently activates executioner caspase -3 and -7. Mutual activation of caspase -3 a -7 is important for the amplification of pro-apoptotic signal. Caspase-9 is also activated by caspase-2. Thus, it does not seem probable that the mitochondrial pathway is crucial for apoptotic induction. In cells with ductal origin, only inititiator caspase-8 and executioner caspase-7 and -6 are activated. Therefore, apoptosis is probably induced by an alternative mechanism in these cells. Concerning Bcl-2 family proteins, apoptosis induction by taxanes in breast cancer cells is associated with increased level of pro-apoptotic protein Bad. Next, we observed some increase in levels of pro-apoptotic proteins Bim a Bok. On the contrary, the level of anti-apoptotic protein Bcl-2 decreases.

2) We showed that the resistance to taxanes could be brought about in breast cancer cells only to such taxanes that have phenyl groups at C3´ a C3´N positions (e.g. clinically used paclitaxel). If at least one of phenyl groups at C3´ a C3´N positions is replaced by a non-aromatic group, it is impossible to bring about resistance to such taxane. These derivates overcome resistance of cancer cells to taxanes with phenyl groups at both C3´ a C3´N positions. Next we found that taxanes with phenyl groups at both mentioned positions have high affinity to binding site of ABCB1 transporter and thus they are effectively transported by ABCB1 transporter out of the cell. When there is at least one non-aromatic group at mentioned positions, the binding affinity of taxane to ABCB1 transporter is lower and the taxane is not effectively transported out of the cells. Then it is impossible to bring about resistance to such taxane.

We can summarize that the induction of apoptosis by taxanes can differ between individual breast cancer cell types depending on their origin. In some types, caspase-2 plays crucial role as an apical caspase, and some proteins of the Bcl-2 family play an important role here. Resistance can be brought about only to those taxanes having phenyl groups at both C3´ and C3´N positions. Only these taxanes are effectively transported out of cells by ABCB1 transporter, which has key role in bringing about resistance to these taxanes.