Institute of Hematology and Blood Transfusion: Department of Proteomics

Chronic myeloid leukemia derived models of resistance to therapy – development and characterization

Many anticancer drugs are currently available for cancer therapy, nevertheless treatment failure can occur due to the development of drug resistance or intolerance. Thus, it is necessary to identify the mechanisms playing a role in the resistance, to find appropriate ways to prevent its development and to find early markers for resistance development detection and ways to solve it.

One of the methods suitable for the study of resistance and of its mechanisms is the use of cell line models. Drug-resistant cell lines are not only good models for understanding of treatment resistance processes, but they are also useful for development and screening new drugs, and for testing new methodologies, disease indicators or treatment responses.

We are studying the resistance to treatment in chronic myeloid leukemia (CML), and therefore we have developed several sub-lines resistant to imatinib and dasatinib. All sub-lines were prepared by long-term cultivation with tyrosine kinase inhibitors (TKIs), namely imatinib (a first-generation drug) and dasatinib (a dual inhibitor, a second-generation drug).

Currently we have resistant clones developed from various types of CML-derived cell lines; both major BCR-ABL transcript variants (e13a2 and e14a2) are available. All the established sub-lines were characterized in terms of origin, the resistance to TKI was then confirmed by growth characteristics (example in Figure 1); EC50 values were defined as the effect of TKI on cells activity (Table 1).

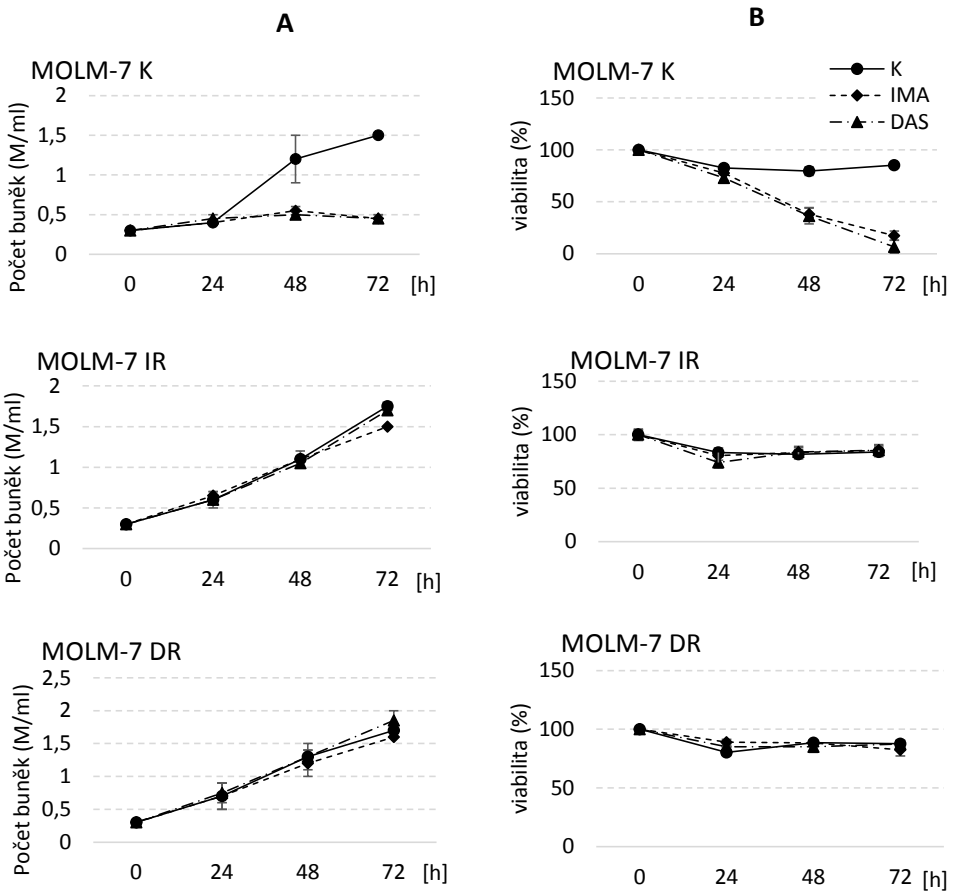

Figure 1 Growth characteristics of the MOLM-7 cell line and of the resistant sub-lines.

The graphs show the growth of cells (A) and their viability (B) alone (K) and in the presence of imatinib (10 µM IMA) or dasatinib (100 nM DAS). MOLM-7 K - control cell line, MOLM-7 IR - imatinib-resistant sub-line, MOLM-7 DR - dasatinib-resistant sub-line.

	imatinib (µM)			dasatinib (nM)		
	EC50	**range**		EC50	**range**	
JURL-MK1	**0,25**	0,1	0,34	**0,19**	0,17	0,22
JURL-MK1 IR	**39**	27	56	**> 100 nM**	-	-
JURL-MK1 DR	**1,1**	0,99	1,3	**1,3**	1,1	1,5
MOLM-7	**0,24**	0,18	0,32	**0,15**	0,06	0,37
MOLM-7 IR	**> 100 µM**	-	-	**1,1**	0,89	1,3
MOLM-7DR	**> 100 µM**	9,00E-08	2,60E+12	**> 100 nM**	-	-
K562	**0,25**	0,14	1,2	**0,35**	0,32	0,38
K562 IR	**5,1**	3,5	9,5	**3,8**	2,3	7,2
K562 DR	**25**	2	-	**4,7**	2,9	8,9

Table 1 EC50 values of imatinib and dasatinib effect on sensitive and resistant cell proliferation.

Cell lines were incubated with imatinib (range 0 - 100 µM) or dasatinib (range 0 - 100 nM). EC50 was defined as the TKI concentration that caused 50% reduction in proliferation/viability (as assessed by Alamar blue method). IR — imatinib-resistant cells, DR — dasatinib-resistant cells.