5. Summary

The antiapoptotic Bcl-x_L protein plays a key role in the control of apoptosis and in the pathogenesis of a variety of malignant diseases. The influence of the 5'-noncoding region of the *bcl-x* gene and the influence of the 5'–nontranslated region of the *bcl-x* mRNA on the expression of the *bcl–x* gene has been investigated in this work.

Previously unknown parts of the human *bcl-x* gene were sequenced and the sequence of the hitherto unidentified complete human *bcl-x_L* mRNA-sequence was determined. Identification and detection of mono- and bicistronic spliceforms of the *bcl-x* mRNA in several human cell lines and mapping of different transcription start sites in the 5'–NCR of the human *bcl-x* gene helped identify different mechanisms for the control of *bcl-x* gene expression.

The position of several transcription start-sites and the location of the major promoter activity in the 5'-noncoding region of the human *bcl-x* gene support the hypothesis that the selection of certain transcription start sites and transcriptional activity are controlled by downstream promoter elements.

Functional analysis revealed that the 5'-nontranslated region of the human *bcl-x* mRNA contains an internal ribosome entry site (IRES) which facilitates the translation of this particular mRNA by a CAP-independent mechanism. These results lead to the conclusion that the expression of the human *bcl–x* gene is significantly controlled at the level of translation. This constitutes the first example of an apoptosis control gene regulated by IRES-mediated initiation of translation.

Transient expression of antisense and anti-antisense RNA which correspond to the 5'–nontranslated region of different *bcl-x* mRNA forms enabled the identification of the preferentially translated *bcl-x* mRNA form. In addition, these experiments show for the first time that CAP-independent translation of the *bcl-x* mRNA can be competitively inhibited by the expression of truncated RNAs containing only the translational control sequences.

A cellular *bcl-x* antisense RNA that shows no influence on the *bcl-x* gene expression was identified and detected in several human cell-lines. The low concentration of this antisense RNA and the existence of two bicistronic *bcl-x_L* mRNAs support the hy-

pothesis that the cellular *bcl-x* antisense mRNA may have a catalytic function in trans-splicing.

Finally, determining the equilibrium concentration of a protein in a certain tissue or cell type from the total concentration of its mRNA variants is not acceptable if the mRNA variants are translated differentially.