If we accept the arguments speaking in favour of v-*src* being a one-hit oncogene, we face a problem how to explain the absence of its oncogenic activity, apart from several cases when it was transduced by a retrovirus. In fact, no case is known of a non-viral tumour that arose by activation of the c-*src* protooncogene. It might be argued that an oncogenic mutation in c-*src* is a rare event. It does not agree with findings demonstrating a series of cell-transforming mutations spanning not only the regulatory domain, but also all the main functional c-*src* domains (Jove and Hanafusa, 1987; Parsons and Weber, 1989; Boeuf et al. 1995; Gonfloni et al., 1997). There remains, however, the unanswered question to what degree these mutants are oncogenic, because their cell-transforming activity has been tested mainly *in vitro* in some cases using mouse NIH 3T3 cells which, as a permanent cell line, are highly sensitive to transformation. Therefore, the oncogenicity of a panel of such mutants should be evaluated under identical conditions *in vivo*, especially because even *in vitro* tests indicated that some of the mutants were less efficient in cell transformation *in vivo* than v-*src*. However, only four cases of v-*src* transduction by a retrovirus were proven to be oncogenic (see Geryk et al., 1989), all of them having an altered carboxy end and lacking critical regulatory tyrosine at the position 527. It was proposed that a high oncogenic activity of v-*src* evolved as a result of repeated passaging and selection of v-*src* mutants (Hjelle et al., 1988). This might have occurred in the case of long-term *in vivo* passaged RSV strain. However, we have shown that v-*src* PR2257, which arose *de novo* by one frame-shift mutation from c-*src* during retroviral transduction, exerts, even in the form of cloned proviral DNA, at least a comparable tumorigenic efficiency in chickens as cloned v-*src* DNA, metastasizing included (Yatsula et al., 1996). Thus, v-*src* transduction and its insertion in a retroviral genome appear to be of utmost importance. For v-*src*, this provides strong promoter-enhancer sequences located in retroviral LTR, which should ensure its efficient expression. However, as discussed above, v-*src* stripped of the 5'end LTR and containing only part of the 3'end LTR enhancer region can efficiently produce chicken sarcomas. Hence, the v-*src* incorporation in a retrovirus might be required for several other reasons. We can imagine that retroviral transduction essentially allows v-*src* escape from the c-*src* locus (juxtaposition) and, therefore, its ectopical expression in another genome position. Conversely, when v-*src*-producing mutations remain part of the c-*src* locus, they become efficiently down-regulated. This could explain why possible oncogenic mutations in the non-juxtaposed c-*src* do not trigger oncogenesis. This hypothesis should be tested experimentally, using a cloned c-*src* locus where the coding region would be replaced by v-*src*. There are several variations on this theme. One of them implies a down-regulatory role of introns and 3' untranslated region (3'UTR) because, in contrast to c-*src*, v-*src* is lacking the intron sequences and 3'UTR, which were lost in the course of retroviral transduction involving the reverse-transcription step. The investigation of c-*src* 3'UTR influence on gene expression yielded evidence that this regulatory element significantly decreases the oncogenicity of v-*src* PR2257 and expression of reporter genes such as luciferase (Trejbalová, unpublished). These questions should be analyzed and answered in order to provide a concise explanation for v-*src* oncogenicity.

Does it have any sense to study a one-hit oncogene when in many cases of human tumours we are facing several-hit oncogenesis? A one-hit oncogene should exert multiple functions, which in case of multi-hit oncogenesis are exerted by different genes and their modifications. In contrast to one-hit oncogenes, multi-hit oncogenesis is therefore made possible by acquisition, change or loss of several independent functions mediated by different genes. In most cases genetic alterations are involved, but in relation to the loss of function, even epigenetic modification like methylation can play an important role. Therefore, a full knowledge of single-hit oncogenesis should provide insight into the functions required for this process. Furthermore, by depicting the oncogene domains responsible for each function it should be possible to proceed with their deletion or alteration and to learn if and how they are required for oncogenesis. Finally, we should take into account that some additional oncogenes, especially those that arose by translocation and therefore are expressed ectopically, might require only very limited changes in other genes, or that they might act essentially as single-hit oncogenes.

I share with George Klein his admiration for Daniel Dennett (1995) in *Darwin's Dangerous Idea*, who questions, in the light of the evolutionary theory, the significance of traditional categories like purpose (telos) and who installs this theory on a pedestal of science. In the subchapter "Darwin's Assault on the Cosmic Pyramid", Dennett interprets Darwin as suggesting: "Give me Order and Time and I will give you Design". Evolution certainly proceeded through different levels of Order, until reaching *Homo sapiens* with his most ordered brain.

The problem of cancer, however, lies in disorder. Dennett justly admits that evolution requires a certain level of disorder represented by a functionally acceptable mutation rate. As G. Klein (1996) pointed out, deficient DNA repair provides the ground that favours steps leading to tumorigenesis. However, not every gene alteration is significant for cancerous growth, and therefore knowledge of key functions and their coding should represent an efficient therapeutic target for so far immature gene therapy. In spite of the limited progress in cancer control, the discoveries of the 20th century laid a solid ground for new solutions and therapeutic strategies, which we should witness at the beginning of the new Millenium.

I would like to finish by referring to Peyton Rous (1965), whose virus brought us the first defined oncogene: "Perhaps to-morrow some cleaving discovery on

causation of tumours (by viruses) will demolish the inferences of to-day: yet what we now know would be worth little if none was made."

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