Destabilizing actin filaments

During phagocytosis, actin filaments depolymerize[8] and during *Salmonella* entry this depolymerization would require downregulation of the filament-stabilizing activity of SipA. This could be achieved by other *Salmonella* effectors acting in concert to regulate SipA function, or could SipA regulate itself? Zhou *et al.*[4] used a recombinant derivative containing the carboxy-terminal 226 residues of SipA. As full-length SipA is nearly three times this size, it is legitimate to wonder if the SipA moiety that was used reflects the full range of functions of SipA, because cytoskeletal proteins often have more than one effect on actin. For example, another characteristic of *sipA* mutants is their scattered localization after internalization by the host cell; this could be interpreted as the bacteria becoming 'entangled' in the cortical actin meshwork because they cannot destabilize actin filaments in the absence of SipA. An in-depth characterization of SipA function requires the purification of the full-length protein and the study of its effects on actin *in vitro*, as well as after introduction into host cells.

Salmonella vs *Shigella*

To what extent can we compare *Salmonella* and *Shigella* cell invasion? The SipA–D proteins have similarities with the *Shigella* IpaA–D proteins, which are implicated in a bacterial invasion process resembling that of *Salmonella*. Like SipA, IpaA is not involved in the polymerization of actin at the site of bacterial entry, but is involved in the transformation of the cell extensions extruded by the host into a structure that is suitable for invasion[9]. IpaA binds to vinculin via the carboxy-terminus, though this shares no significant homology with the carboxy-terminus of SipA (G. Tran Van Nhieu, unpublished). Interestingly, IpaA appears to perform the opposite function to SipA, as the IpaA–vinculin complex depolymerizes actin filaments (R. Bourdet-Sicard *et al.*, submitted). One can infer from these contrasting results that *Salmonella* and *Shigella* have evolved effectors that perform opposing functions but which, when acting together with other effectors, ultimately aim at bacterial entry into host cells.

Acknowledgements

R. B-S. is a recipient of a grant from the Ministère de l'Education Nationale de la Recherche et de la Technologie. This work was supported by grant 94092 from the Direction des Recherches et Techniques to P.J. Sansonetti.

References

1 Galan, J.E. and Bliska, J.B. (1996) *Annu. Rev. Cell Dev. Biol.* 12, 221–255
2 Finlay, B.B. and Cossart, P. (1997) *Science* 276, 718–725
3 Hardt, W-D. *et al.* (1998) *Cell* 93, 815–826
4 Zhou, D. *et al.* (1999) *Science* 283, 2092–2095
5 Carlier, M-F. *et al.* (1997) *J. Cell Biol.* 136, 1307–1323
6 Lee, E. *et al.* (1997) *Exp. Cell Res.* 235, 295–299
7 Steinmetz, M.O. *et al.* (1997) *J. Cell Biol.* 183, 559–574
8 Massol, P. *et al.* (1998) *EMBO J.* 17, 6219–6229
9 Tran Van Nhieu, G. *et al.* (1997) *EMBO J.* 16, 2717–2729

Letters

HHV-8 and multistep tumorigenesis

Human herpesvirus 8 [HHV-8 or Kaposi's sarcoma-associated herpesvirus (KSHV)] is associated with pleural effusion lymphoma (PEL), multicentric Castleman's disease (MCD) and Kaposi's sarcoma (KS)[1]. These diseases are rare, but HHV-8 can infect more than 50% of the general population, depending on the geographical area. This indicates that additional factors are required for HHV-8 pathogenicity[1]. This is not surprising, as the concept of multistep tumorigenesis is now generally accepted and it does not rule out an etiologic role for HHV-8 in tumor development.

Recent data have challenged the idea that HHV-8 triggers a single and defined step of tumorigenesis in the same manner as a proto-oncogene. Firstly, HHV-8 can infect different cell types – endothelial cells, B cells, T cells and monocytes[2–4]. Secondly, HHV-8 appears to cause neoplastic transformation in PEL [in certain cases in cooperation with Epstein–Barr virus (EBV)] but not of endothelial cells or KS tumor cells[1]. Thirdly, HHV-8 is recruited secondarily into KS lesions by inflammatory cells[3], exerts paracrine effects in early lesions and only has autocrine tumorigenic activity in late lesions[5]. This is in agreement with the hyperplastic development of KS (Ref. 1). Finally, at the molecular level, several HHV-8-encoded genes with potential pathogenic activity are differentially expressed in different diseases[1]. For example, viral-encoded interleukin 6, the most likely candidate for an HHV-8-encoded B-cell mitogen, is expressed in MCD but not in KS (Ref. 6). By contrast, viral-encoded cyclin D (mitogenic) and viral-encoded FLICE-inhibitory protein (v-FLIP) (anti-apoptotic) are highly expressed in latently infected KS spindle cells and could drive tumor progression[6,7].

HHV-8 has a broad cellular tropism and this could differ between patients with different diseases. However, the outcome of HHV-8-induced diseases cannot yet be predicted. This supports the idea that HHV-8 pathogenicity could be triggered by viral (e.g. HIV-1 or EBV) and/or cellular (e.g. cytokines or growth factors) cofactors.

Two key questions have not yet been answered in relation to HHV-8 pathogenicity – what are the mechanisms of viral reactivation and what is the role of the immune system in the control of viral infection? These issues are critical to our understanding of the role of this virus in disease, particularly in KS. In fact, virus load and tissue spread are higher in individuals at risk of KS, or with KS, compared with HHV-8-infected, non risk individuals[2]. $CD8^+$ T cells (and possibly cytotoxic

T cells) are abundant in KS lesions; however, they are unable to clear the virus[8,9]. Why is HHV-8 so well controlled in the general population? Is it because of the lack of reactivating factors, the immune control of the virus or both? Recent studies indicate an excess risk of KS in HIV-1–HHV-8 coinfected homosexual men that is not explained by HHV-8 (Ref. 10). If we look at the risk groups for KS, it is clear that the dysregulation of the immune system has a role in disease initiation[1]. This is also evident from the histopathology of the lesions: very early lesions are characterized by inflammatory cells producing cytokines, even prior to the appearance of the spindle cells[1,8]. Recent evidence indicates that the same cytokines (Th-1 type) produced in KS or in at-risk individuals can reactivate virus infection and increase viral load in a fashion similar to cytomegalovirus after allogenic stimulation[2].

Taken together, these data suggest that HHV-8 could be a passive mediator rather than an active inducer of tumorigenesis. HHV-8 might be unique because it could catalyze different steps in the pathways of the multistep tumorigenesis process in different types of tumors (Fig. 1). This could explain why HHV-8 is involved in such contrasting and apparently unrelated diseases.

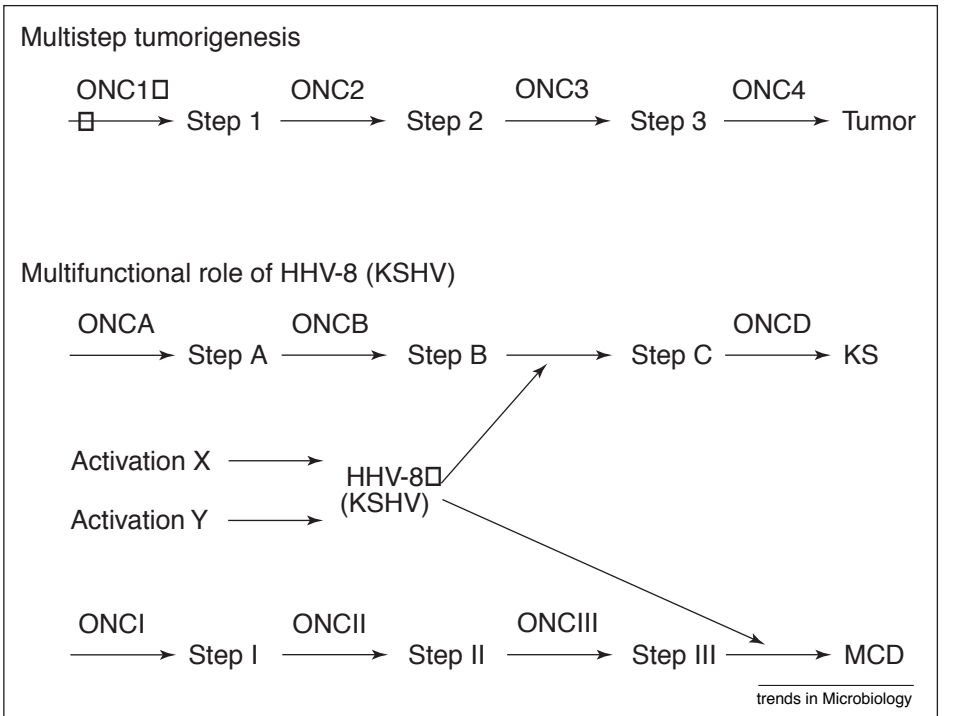

Fig. 1. Multifunctional role of HHV-8 in multistep tumorigenesis. Tumor development occurs in several different steps regulated by differentially activated oncogenes. Depending on its activation status (X or Y), HHV-8 could regulate tumor progression at different steps in unrelated pathways leading to different diseases. Abbreviations: KS, Kaposi's sarcoma; KSHV, Kaposi's sarcoma-associated herpesvirus; MCD, multicentric Castleman's disease.

Barbara Ensoli
Laboratory of Virology,
Istituto Superiore di Sanità,
Viale Regina Elena 299,
00161 Rome, Italy

Michael Stürzl
GSF-National Research Center
for Environment and Health,
Institute of Molecular Virology,
85764 Neuherberg and
Bavarian Nordic Research
Institute AS, 82152 Martinsried,
Germany

References

1 Ensoli, B. and Stürzl, M. (1998) *Cytokine Growth Factor Rev.* 9, 63–83
2 Monini, P. *et al.* (1999) *Blood* 93, 4044–4058
3 Blasig, C. *et al.* (1997) *J. Virol.* 10, 7963–7968
4 Stürzl, M. *et al.* (1997) *Int. J. Cancer* 72, 68–71
5 Dupin, N. *et al.* (1999) *Proc. Natl. Acad. Sci. U. S. A.* 96, 4546–4551
6 Stürzl, M. and Ensoli, B. (1999) *Int. J. Oncol.* 14, 287–289
7 Davis, M.A. *et al.* (1997) *J. Natl. Cancer Inst.* 89, 1868–1874
8 Fiorelli, V. *et al.* (1998) *Blood* 91, 956–967
9 Sirianni, M.C. *et al.* (1998) *Blood* 91, 968–976
10 Rezza, G. *et al. J. Natl. Cancer Inst.* (in press)

Response from Schulz and Moore

We agree with Drs Stürzl and Ensoli that, in addition to Kaposi's sarcoma-associated herpesvirus (KSHV) infection, many factors can determine whether or not clinical disease occurs. This is true for all infectious processes, with the potential exceptions of rabies and AIDS. The most critical factor is clearly cell-mediated immune surveillance, which is damaged by AIDS and post-transplantation immunosuppression. This is not surprising for a virally induced tumor in which surveillance for foreign antigens plays a critical role. It is unlikely that complementing mutations are required for Kaposi's sarcoma (KS) initiation as it is an extremely common tumor among immunosuppressed and KSHV-infected people (up to 50% of KSHV-infected gay men with AIDS develop KS)[1]. By contrast, despite nearly 100% Epstein–Barr virus (EBV) infection rates in cohorts of AIDS patients, EBV-induced lymphoproliferative disorders are uncommon (<2%).

We have difficulty, however, with their proposal that KSHV infection is a late event in a multistep model of KS pathogenesis. The majority of evidence contradicts their assertion that pre-existing KS lesions recruit KSHV-infected inflammatory cells to the tumors. Numerous studies (reviewed in Ref. 2) have shown that virtually all KS tumors are infected with KSHV. The few non-infected lesions are probably a result of misdiagnosis (particularly at early stages) or technical problems such as DNA degradation. Furthermore, KSHV infection is highly predictive of disease[1,3] and it is extremely improbable that the virus can determine who will develop KS in ten years time and then specifically infect them. Although KSHV infection rates can exceed 50% among gay men and some African populations, these groups also have the highest rates of clinical KS disease. Rates of infection