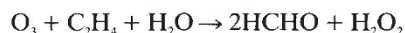


Ozone and ethylene stress

SIR—In a recent News and Views article¹ “Adding ethylene to injury”, M. Unsworth discussed the results of Mehlhorn and Wellburn², who reported that both endogenous ethylene (a stress-hormone) and exogenous ozone are co-operating prerequisites for injury to pea plants. This important experimental result is indeed a milestone in plant toxicology. But Unsworth’s statement that a co-operation of ozone and ethylene in producing plant damage... “does not seem to have been suggested previously...” needs some comments.

In 1985, we reported³ changes in anti-oxidative activities in damaged spruce needles correlated with the ethylene precursor ACC or malonyl-ACC as a stress marker. We concluded that reaction products of the interaction between ozone and ethylene, namely peroxides and reactive aldehydes, might be the damaging species. We summarized these conclusions in the simplified equation:



The basis of this theory was published in 1984 in German⁴. In later publications^{5,6} we state that “the primary damaging reactions in spruce needles may operate as follows: (1) Trees under ‘stress’ produce the plant hormone ethylene; (2) ethylene and ozone react extremely fast, forming hydrogen peroxide and formaldehyde, compounds which may damage the wax layer; (3) ozone as a very aggressive...” (from ref.6). The findings of Mehlhorn and Wellburn with peas justify our earlier assumptions on forest decline and are therefore of utmost importance.

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1. Unsworth, M. *Nature* **327**, 364–365 (1987).
2. Mehlhorn, H. & Wellburn, A.R. *Nature* **327**, 417–418 (1987).
3. Elstner, E.F., Osswald, W. & Youngman, R. J. *Experientia* **41**, 591–597 (1985).
4. Elstner, E.F. & Osswald, W. *Naturw. Rundschau* **37**(2), 52–61 (1984).
5. Osswald, W. & Elstner, E.F. *Ber. Deutsch. Bot. Ges.* **99**, 313–339 (1986).
6. Osswald, W. & Elstner, E.F. *Free Radical Res. Commun.* **3**, 185–192 (1987).

Sequence specificity of retroviral proteases

SIR—The protease encoded by retroviruses cleaves the *gag*-polyprotein to produce the amino terminus of the major core protein (p24, p27 or p30, depending on the virus)^{1–3}. On examining the amino-acid sequence that spans this cleavage site in available retroviral sequences we have found the strongly conserved pattern X-Y-Pro-Z, where X is generally small with some hydrophobic preference, Y is aromatic or large and hydrophobic, and Z

Fig.1 Alignment of sequences in retroviral *gag*-polyproteins, which are cleaved to yield the N-terminal proline of the major core protein. Sequences are from mouse mammary tumour virus (MMTV), hamster intracisternal A-particle (HIAP-18), simian AIDS retrovirus (SRV-1), human T-cell leukaemia virus type II (HTLV-II), AKV murine leukaemia virus (AKV-MuLV), Moloney murine leukaemia virus (Mo-MuLV), feline leukaemia virus (FeLV), H7-baboon endogenous virus (H7BEV), human immunosuppressive virus type 1 (HIV-1), human immunosuppressive virus type 2 (HIV-2), visna lentivirus (VLV), Rous sarcoma virus (RSV), Fujinami sarcoma virus (FSV), human T-cell leukaemia virus type I (HTLV-I), feline sarcoma virus (FeSV), simian sarcoma virus (SSV) and bovine leukaemia virus (BLV).

AKV-MuLV	Ser Ala Leu Tyr	Pro	Ala Leu	} p15/p12 <i>gag</i>
Fr-SFFV	Ser Ala Leu Tyr	Pro	Ala Leu	
FeLV	Ser Ser Leu Tyr	Pro	Ala Leu	
Mo-MuLV	Ser Ser Leu Tyr	Pro	Ala Leu	
FeSV	Ser Ser Leu Tyr	Pro	Val Leu	
EqIAV	Ser Glu Glu Tyr	Pro	Ile Met	
SSV	Pro Pro Ile Tyr	Pro	Ala Thr	
RSV	Phe Gln Ala Tyr	Pro	Leu Arg	
HIV-1	Thr Leu Asn Phe	Pro	Ile Ser	} Prt/RT <i>pol</i>
HIV-2	Ser Leu Asn Leu	Pro	Val Ala	

is small and hydrophobic (Fig. 1). The totally conserved proline forms the amino terminus of the major core protein. Some weaker clusters of conservation are also seen in the flanking residues.

An additional closely related pattern of sequences is found in some *gag*-polyprotein sequences (Fig. 2), corresponding to the junctions of the p15 and p12 proteins, which may also be a site for cleavage by the viral protease. In the *pol*-polyprotein sequence of Rous sarcoma virus, a sequence matching this pattern occurs at the junction of the reverse transcriptase and the endonuclease, while in the AIDS viruses HIV-1 and HIV-2, a matching sequence occurs at the junction of the (presumed) carboxy terminus of the protease sequence itself with the amino-terminus of the reverse transcriptase, implying that the production of active reverse transcriptase in these viruses may well be dependant upon the action of the virally encoded protease. No matches with this sequence pattern are found at known cleavage sites in the various *env*-polyproteins. It is likely that this overall pattern represents the preferred amino-acid sequence for cleavage by retroviral proteases and may be of use in the design of specific inhibitors of retroviral protease activity for the chemotherapy of AIDS.

Observations that retroviral proteases may be inhibited by thiol-specific reagents^{4,5} has led to suggestions that these

MMTV	Thr Phe Thr Phe	Pro	Val Va
HIAP-18	Gln Met Ala Phe	Pro	Val Pi
SRV-1	Lys Asp Ile Phe	Pro	Val Ti
HTLV-II	Thr Gln Cys Phe	Pro	Ile Li
AKV-MuLV	Ser Arg Ala Phe	Pro	Leu Ai
Mo-MuLV	Ser Gln Ala Phe	Pro	Leu Ai
FeLV	Ser Gln Ala Phe	Pro	Leu Ai
H7BEV	Ser Ser Leu Phe	Pro	Leu Ar
HIV-1	Ser Gln Asn Tyr	Pro	Ile Va
HIV-2	Gly Gly Asn Tyr	Pro	Val Gl
VLV	Arg Glu Val Tyr	Pro	Ile Va
RSV	Val Val Ala Met	Pro	Val Va
FSV	Met Val Ala Met	Pro	Val Va
HTLV-I	Pro Ala Ile Leu	Pro	Ile I.
FeSV	Ser Gln Ala Leu	Pro	Leu Ar
SSV	Thr Val Ile Leu	Pro	Leu Ar
BLV	Pro Ala Ile Leu	Pro	Ile I.

Fig. 2 Alignment of other sequences in viral polyprotein which are cleaved to yield N termini and which may be substrates for the virally encoded proteases. Sequences are from AKV-MuLV, Friend splee focus-forming virus (Fr-SFFV), FeLV, Mo-MuLV, FeSV equine infectious anaemia virus (EqIAV), SSV, RSV, HIV-1 and HIV-2.

enzymes might be thiol-proteases⁶. A more related sequences have become available this hypothesis has appeared more unlikely since the position a number of cysteine residues are found to be poorly conserved. In the recently published⁷ sequence of HIV-2, we have identified the likely position of the amino terminus of the protease as close to residue 85, with the carboxy terminus immediately preceding the probable amino terminus of the reverse transcriptase close to residue 185 (residue numbers are relative to the start of the *pol* open reading frame). No cysteine residues occur within this sequence.

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1. Yoshinaka, Y., Shames, R.B., Luftig, R.B., Smythe G.W. & Oroszlan, S. *J. gen. Virol.* **66**, 379–383 (1985).
2. Yoshinaka, Y., Katoh, I., Copeland, T.D., Smythers, G. & Oroszlan, S. *J. Virol.* **57**, 826–832 (1986).
3. Kramer, R.A. *et al. Science* **231**, 1580–1584 (1986).
4. Yoshinaka, Y. & Luftig, R.B. *Cell* **12**, 709–719 (1977).
5. Dittmar, K. & Moelling, K. *J. Virol.* **28**, 106–118 (1987).
6. Yoshinaka, Y., Katoh, I., Copeland, T.D. & Oroszlan, S. *Proc. natn. Acad. Sci. U.S.A.* **82**, 1618–1622 (1985).
7. Guyader, M. *et al. Nature* **326**, 662–669 (1987).