

Our results and conclusion differ from those of Elsner and Tsonis because we determine the variance of the white-noise forcing for the physical and statistical models, together with the autoregressive coefficients for the latter, from the observed surface temperature record after removal of the temperature changes contributed by increasing greenhouse gases and anthropogenic sulphate aerosols, that is, after detrending. Elsner and Tsonis determine the white-noise variance and autoregressive coefficient for their statistical model from the nondetrended observed temperature record. Thus, they assume that all temperature changes observed since the middle of the nineteenth century are due to natural variability, whereas we assume that only the temperature changes other than those forced by greenhouse gases and anthropogenic sulphate aerosols are due to natural variability. Their assumption means that, unless the anthropogenic forcing of the climate system has been zero year after year, the sensitivity of the

climate system is zero. Although this would mean that there is no global-warming problem, it is not consistent with the varied palaeoclimatic history of the Earth.

Michael E. Schlesinger
 Department of Atmospheric Sciences,
 University of Illinois,
 at Urbana-Champaign,
 Illinois 61801, USA

Navin Ramankutty
 Climate, People and Environment Program,
 Institute for Environmental Studies,
 University of Wisconsin-Madison,
 Madison 53706, USA

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Glutamatergic signals in *Ephedra*

SIR — Plants frequently synthesize toxic non-protein amino acids that disrupt amino-acid transport, metabolism and protein synthesis in leaf- and seed-eating mammals and insects^{1,2}. *Ephedra foeminea* Forssk. (Ephedraceae, Gnetales), the joint-fir, widely distributed in the eastern Mediterranean and the near East³, contains in its fresh stems substantial amounts of the two *cis*-diastereoisomers of the L-glutamate analogue L-2-carboxycyclopropyl glycine, (2*S*, 3*S*, 4*R*)- and (2*S*, 3*R*, 4*S*)-2-(carboxycyclopropyl) glycine (L-CCGIII and L-CCGIV, respectively)⁴. These two compounds, which constitute about 1% of the stem dry weight in this and a related Mediterranean species, *E. altissima* (see table), are rare. L-CCGIII has been reported⁵ in a few North American species of *Aesculus* (the buckeyes). The presence of L-CCGIV in plants has not been reported before. The stem tissue of *E. foeminea* also contains *cis*-3,4-methano-L-proline, a cyclopropane amino acid previously reported only in *Aesculus parviflora*⁶.

Because L-glutamate is central to

amino-acid metabolism and is a neurotransmitter molecule in vertebrates and arthropods, ingestion of L-2-(carboxycyclopropyl)glycines might alter herbivore behaviour. We tested the membrane activity of three L-CCG isomers on two insect tissues *in vitro*. A Na⁺-dependent glutamate transporter in the beetle epidermis⁷ concentrated both *cis*-isomers, whereas L-CCGIII and L-CCGI (the *trans*-2*S*, 3*S*, 4*S*-isomer found in *Blighia sapida*⁸) caused a cockroach hindgut preparation⁹ to contract. In mammalian neurons, L-CCGIII potentiates responsiveness to L-glutamate⁹ and L-CCGIV activates the N-methyl D-aspartate (NMDA) subtype of the L-glutamate receptor¹⁰.

The stems of many *Ephedra* species also contain substantial amounts of a quinoline, 6-hydroxykynurenic acid (6-HKYNA), known to occur in trace amounts in many plants¹¹. In *Ginkgo biloba*, a primitive tree notably free of insect pests, however, it may constitute 0.24% of the leaf dry weight¹². Similar amounts are seen in *Ephedra* (see table). In the mammalian brain, substituted

kynurenic acids are selective antagonists competing with glycine for binding sites on NMDA and non-NMDA glutamate receptors¹³. 6-HKYNA, however, had no activity on the hindgut, whether applied alone or together with glutamate or quisqualate, suggesting that 6-substituted kynurenic acids are not glutamate receptor antagonists in insect visceral muscle. 7-Methoxykynurenic acid, reported to occur in *E. alata*¹⁴, is not a glutamate receptor antagonist in mammalian neurons¹³.

Most Eurasian species of *Ephedra*, such as *E. distachya* and *E. fragilis*, contain the sympathomimetic drugs ephedrine, pseudoephedrine and pharmacologically related amines¹⁵. Ephedrine in the diet is toxic to the pea weevil². The ephedrine alkaloids and the tannins present in the stems of most Eurasian species are absent from *E. foeminea*, *E. altissima* and *E. foliata* (see table). As methanoproline, too, is restricted to tannin- and ephedrine-free species, we propose that cyclopropyl amino acids act as an alternative form of feeding deterrent. A possible reason why some *Ephedra* species are tannin-free may be that tannins block Na⁺-dependent amino-acid transport, compromising the defensive role of ingested cyclopropyl amino acids. The ephedrine alkaloids found in many *Ephedra* species containing tannins are non-polar and do not need special transporter proteins for their absorption. It is remarkable that, despite the more than 100 papers published over the past 100 years or so on the alkaloid content of *Ephedra*, the presence of cyclopropyl amino acids and 6-hydroxykynurenic acid has previously gone undetected.

Stanley Caveney
 Department of Zoology,
 University of Western Ontario,
 London, Ontario, Canada N6A 5B7

Alvin Starratt
 Pest Management Research Centre,
 Agriculture and Agri-Food Canada,
 London, Ontario, Canada N5V 4T3

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CYCLOPROPYL AMINO ACIDS, 6-HYDROXYKYNURENIC ACID AND EPHEDRINE ALKALOIDS, AND PRESENCE OF TANNINS IN THE FRESH STEMS OF SELECTED *EPHEDRA* SPECIES

Species	L-CCGIII	L-CCGIV	Methano-proline	6-Hydroxy-kynurenic acid	Ephedrine	Pseudo-ephedrine	Tannins
<i>E. foeminea</i>	++++	+++	++	+++	—	—	no
<i>E. altissima</i>	++++	+++	—	++	—	+	no
<i>E. foliata</i>	—	—	++	+++	—	+	no
<i>E. fragilis</i>	++	—	—	++	++++	—	yes
<i>E. distachya</i>	—	—	—	++	—	++++	yes

Amounts of amino acids and alkaloids are expressed as per cent dry tissue weight: (—), not detected; (+), < 0.05 %; (++) , 0.05–0.19%; (+++) , 0.2–0.5%; (++++), > 0.5%.