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Global change

Deciphering methane's fingerprint

Helmut Weissert

Two hundred years ago Georges Cuvier came up with the concept of what we now call 'proxy indicators' in geology. These are biological, chemical or physical signatures preserved in the rock record that serve as fingerprints of the biosphere's evolution through time. Much of the detective work on Earth's history has depended on these fingerprints.

These days, increasingly sophisticated tracer techniques are used in combination, and their robustness is tested by simulations of past global change. This is the approach that Hesselbo *et al.*¹ have chosen in the study described on page 392 of this issue — a carbon-isotope analysis of fossil wood deposited in near-shore sediments 180 million years ago during the Toarcian Stage of the Jurassic (200–140 million years ago). The authors have identified an atmospheric carbon-isotope signal in their samples that records an anomaly in the atmospheric carbon system; moreover, it is coupled to a known perturbation of the marine carbon reservoir. Hesselbo *et al.* propose that a massive release of methane, stored in gas hydrates beneath the sea floor, caused the perturbation in the carbon-isotope record, and that the methane pulse triggered major changes in climate and the oceans.

These conclusions are based mainly on carbon-isotope geochemistry. The relative concentrations of the two stable carbon isotopes (carbon-12 and carbon-13) in organic matter and biologically generated carbonate are used as fingerprints that reflect events in the carbon system. Marine sediments deposited over the past 600 million years retain the carbon-isotope signature from the time of their deposition, and oscillations in the geological record show that marine carbon-isotope composition was not stable². Differences in that composition can be related either to changes in the carbon isotopes entering the oceans, or to isotopic variations in total carbon sedimentation, which are caused mainly by varying rates of organic-carbon burial in sediments on the sea floor.

Organic carbon is enriched in the light isotope, carbon-12. So, at times of high rates of burial, carbon-13 became more abundant in the ocean water and is reflected as a positive

carbon-isotope anomaly in the rock record left by marine sediments. Burial of excess organic carbon was initiated by a warm, greenhouse climate, and coincided with crises in biocalcification in reefs and in calcareous plankton³, which were triggered by high levels of atmospheric CO₂ (ref. 4) and related changes in ocean chemistry and climate. Both processes contributed to a lowering of the amount of CO₂ in the atmosphere.

Negative carbon-isotope anomalies record episodes of oceanic carbon-12 enrichment in the past. Extraordinary carbon-12 enrichment of the marine carbon reservoir within only a few tens of thousands years coincided with known environmental catastrophes, as at the Cretaceous/Tertiary boundary⁵, some 65 million years ago. A collapse of the marine ecosystem and the con-

sequent interruption of the marine carbon flux into sediments may have caused the observed carbon-12 enrichment⁵. Other negative anomalies are evident at the base of positive carbon-isotope events⁶, and have been considered as the fingerprint of rapid influx of volcanic CO₂ enriched in carbon-12. This volcanic CO₂ would have triggered the biotic changes recorded in the subsequent positive carbon-isotope anomalies.

A few years ago, however, Dickens *et al.*⁷ proposed a new explanation for the negative carbon-isotope anomaly at the Palaeocene/Eocene boundary 55 million years ago. They calculated that only a sudden influx of methane derived from sources under the sea floor could account for the anomaly. Methane is enriched in the light isotope, carbon-12. It is stored in sedimentary gas hydrates beneath the oceans, and consists of solid crystals of water and methane that are stable under a wide range of temperatures and pressures.

Dickens *et al.* argued that deep-water warming or a rapid pressure decrease (or both) resulted in sudden release of methane (Fig. 1). The methane was oxidized to CO₂, which was enriched in methane-derived carbon-12. Higher CO₂ concentrations in oceans and the atmosphere were a consequence of this environmental catastrophe. Plants take up CO₂ in photosynthesis, so a pulse of methane into the atmosphere should be evident in the terrestrial carbon-isotope records of organic matter.

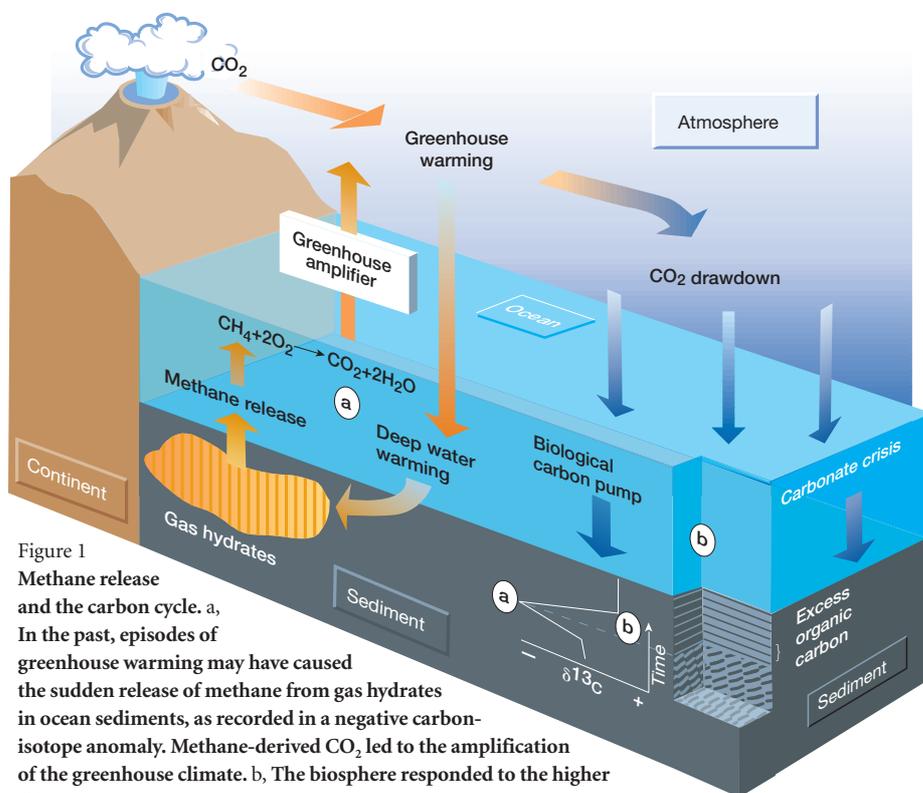


Figure 1 Methane release and the carbon cycle. a, In the past, episodes of greenhouse warming may have caused the sudden release of methane from gas hydrates in ocean sediments, as recorded in a negative carbon-isotope anomaly. Methane-derived CO₂ led to the amplification of the greenhouse climate. b, The biosphere responded to the higher CO₂ levels with accelerated burial of organic carbon on the ocean floor, and with crises in carbonate production, as recorded in positive carbon-isotope anomalies. Both processes contributed to a lowering of atmospheric CO₂ levels. Hesselbo *et al.*¹ have identified a terrestrial isotopic signal that provides evidence of a massive release of methane about 180 million years ago.

This is what Hesselbo and co-authors¹ have looked for, taking as their target one of the most prominent negative carbon-isotope anomalies known from the Jurassic. This anomaly occurs at the base of a large positive carbon-isotope 'excursion', which corresponds to the Toarcian oceanic anoxic event; this was a time when the deep-ocean waters were severely depleted in oxygen and there was excess burial of organic matter. The terrestrial carbon-isotope curve measured by Hesselbo *et al.* from their wood samples shows that the negative marine carbon-isotope anomaly is also reflected in the record of the global carbon cycle.

So what was the cause? The negative carbon-isotope event coincides with an episode of strong volcanic activity. But mass-balance calculations show that volcanic carbon cannot by itself account for the amount of carbon-12, which is calculated to have been generated within a few tens of thousands of years. Instead, Hesselbo *et al.* conclude that the Toarcian carbon-isotope event was a consequence of sudden methane release from beneath the ocean floor. Climate warming triggered by CO₂ from volcanoes could have led to a sudden warming of the deep ocean and the destabilization of gas hydrates. The authors think that a huge amount of methane was released, corre-

sponding to about 20% of the present-day gas-hydrate reservoir and twice that emitted at the end of the Palaeocene.

The work of Hesselbo *et al.*, and of others⁸, indicates that during the Mesozoic (250–65 million years ago) a warmer, greenhouse climate may have favoured episodic release of gas hydrates into the atmosphere. This raises the provocative question of whether the carbon-isotope anomalies at the Cretaceous/Tertiary boundary, and also at the 250-million-year-old Permian/Triassic boundary, were a result of sudden methane release — in these cases possibly triggered by an extraterrestrial object hitting the Earth. As the accuracy of our information from proxy indicators increases, Mesozoic environmental history is becoming an exciting arena for high-resolution detective work. ■

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Developmental biology

Fringe benefits to carbohydrates

Mark E. Fortini

The past decade has seen tremendous advances in our understanding of the molecular mechanisms underlying development. Papers on pages 369 and 411 of this issue^{1,2}, along with studies in *Nature Cell Biology*³ and *Current Biology*⁴, provide fascinating insights into the biochemical features of one such mechanism, which establishes spatial boundaries in developing tissues. The authors make a convincing case that carbohydrate chains attached to the extracellular portion of a particular receptor on the cell surface can modulate the interaction of the receptor with its binding partners. Remarkably, the carbohydrates thereby lead to the receptor being activated in only a particular tissue area.

The proteins producing these carbohydrate modifications are Fringe — found in the fruitfly *Drosophila melanogaster* — and its vertebrate counterparts, Lunatic Fringe, Radical Fringe and Manic Fringe. Notch is the receptor in question, and it is activated at the cell surface when it binds to a ligand from the Delta or the Serrate/Jagged protein families. *Notch* was one of the first genes to be identified in *Drosophila*, being

described by T. H. Morgan and colleagues as early as 1916. It has long been known to control tissue-patterning events in which certain cells are singled out from a group of equivalent cells⁵. More recently, Notch has captured the attention of biologists studying

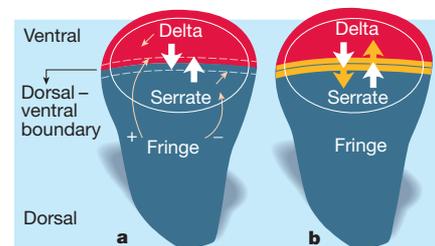


Figure 1 Creation of simple boundaries through complex carbohydrates. Early development of the *Drosophila* wing imaginal disc, an undifferentiated structure from which the wing will arise. a, The Notch protein and one of its ligands, Delta, are expressed widely in the disc. Serrate (another Notch ligand) and Fringe (a protein that modifies Notch's response to its ligands) are restricted to the dorsal compartment. Fringe inhibits the ability of Serrate to activate Notch (minus symbol),

vertebrate development, in part because malfunctioning of the Notch signalling pathway underlies an ever-growing list of human diseases, including T-cell leukaemia, adult-onset dementia, and the developmental disorders spondylocostal dysostosis and Alagille syndrome.

In certain situations during development, Notch signalling can be spatially modulated by Fringe, such that Notch is activated along a discrete line of cells. This specialized adaptation of the pathway is vital in establishing a sharp boundary between dorsal and ventral compartments during development of the insect wing, patterning of the chick limb bud, and other developmental processes. Genetic studies have led to an elegant model for how Fringe regulates boundary formation^{6–8} (Fig. 1). The key feature of this model is that Fringe somehow causes Notch to become more sensitive to Delta and less sensitive to Serrate/Jagged. As a result, Notch signalling is amplified in a narrow band of cells running along the dorsal–ventral boundary of the wing. But how does Fringe do this?

On the basis of slight similarities of the amino-acid sequence of Fringe to those of some bacterial enzymes⁹, Fringe has been proposed to be a glycosyltransferase — an enzyme that adds sugar residues to proteins. But, until now, direct evidence to support this suspicion was lacking.

Moloney *et al.*¹ and Brückner *et al.*² now show that Fringe is indeed a glycosyltransferase, with a new *N*-acetylglucosaminyltransferase activity. Within the extracellular portion of Notch are 36 consecutive motifs called epidermal-growth-factor (EGF)-like domains. A five-carbon sugar, fucose, is linked to the side chains of serine, threonine or hydroxylysine amino acids within a subset of these domains (sugars linked to any of these three amino acids are known as *O*-linked sugars). Fringe causes elongation of these *O*-linked sugars to more complex forms.

Moloney *et al.*¹ transfected mammalian

whereas it potentiates the ability of Delta to activate Notch (plus symbol). Notch is thus activated most strongly in the area between the dashed lines: it is activated by Serrate in the Fringe-negative ventral cells along the boundary, and by Delta in the Fringe-positive dorsal cells along the other side of the boundary. b, As wing development proceeds, amplification of Notch signalling (yellow band) in cells on either side of the boundary results in a transcriptional feedback loop (yellow arrows) that eventually restricts ligand expression to the boundary (not shown). This system has similar functions in development in other organisms. New results^{1–4} show that the *Drosophila* and mammalian Fringe proteins modify the response of Notch to its ligands by adding sugar groups to Notch's epidermal-growth-factor (EGF)-like extracellular domains (see Fig. 6 on page 373 and Fig. 4 on page 413).