silicate grains seem to be concentrated near β Pic (as expected, due to Poynting–Robertson drag).

Crystalline silicate grains are also concentrated near β Pic. This could be attributed to the higher temperatures there, because amorphous silicate grains are annealed (converted to crystalline form) above about 1,100 K (ref. 9). Alternatively, these grains could have been shed by comets entering β Pic's inner solar system[10]. Carbon monoxide molecules, which are easily destroyed by ultraviolet radiation from β Pic, have been detected in the β Pic disk[3] — strongly implying that they are being replaced through the continuing evaporation of comets. In our own Solar System, a high proportion (about 30%) of the silicate grains shed from comets such as Hale–Bopp are crystalline[11].

The presence of small amorphous grains at the particular radii of 16 and 30 AU is intriguing; these radii match the locations of the dust bands previously inferred for the β Pic disk[7]. Okamoto *et al.*[1] have found that the dust grains at these radii are amorphous silicates and are small enough to require constant replenishment. These data strongly suggest the presence of belts of comets or planetesimals at these radii in the β Pic disk (Fig. 1). The improved resolution of Okamoto and colleagues' data, however, has led them further, to the detection of another dust belt at a radius of 6.4 AU. As the authors discuss, this arrangement of a belt of planetesimals or comets at 6.4 AU as well as 16 AU suggests the gravitational influence of a shepherding planet at a radius of 12 AU.

As for the dominance of amorphous silicates in these belts, it could be that there are no crystalline silicates at these radii (unlike the situation inferred from comets from our own Solar System), or that silicates are transformed within the planetesimals from crystalline to amorphous form. Future work can confirm or deny these hypotheses. But it is clear that astromineralogical observations will provide further, exciting insight into the formation of rocky planets in β Pic and other solar systems. ■

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Molecular biology

No exception to reversibility

Yi Zhang

Histone proteins, which serve as scaffolds for packaging DNA, can be modified in numerous ways. It's been thought that one modification, methylation, is irreversible — but that view must now change.

Each of our cells contains about two metres or so of DNA, which must be packed down very tightly to fit into the cell nucleus. The compact form of DNA is known as chromatin, the basic unit of which consists of DNA wrapped around an octamer of histone proteins[1]. The histones are not merely architectural proteins, however: they also influence chromatin dynamics. One way in which they do so is through their covalent modification with certain chemical groups or small proteins — acetyl groups, phosphate groups, ubiquitin proteins or methyl groups[2]. Enzymes that catalyse the addition or removal of the first three modifications have been identified. But enzymes that remove methyl groups have been more elusive, raising the question of whether methylation is an exception to the rule: the only histone modification that is irreversible[3]. Two new papers, however — published in *Science* by Wang *et al.*[4] and in *Cell* by Cuthbert *et al.*[5] — reverse that view.

The histone octamer consists of two copies each of four 'core' histones: H2A, H2B, H3 and H4. Methylation occurs on certain lysine and arginine amino acids within H3 and H4, and is catalysed by distinct families of enzymes[6]; PRMT1 and CARM1, for instance, are enzymes involved in arginine methylation. Lysine residues can be mono-, di- or tri-methylated, whereas arginine

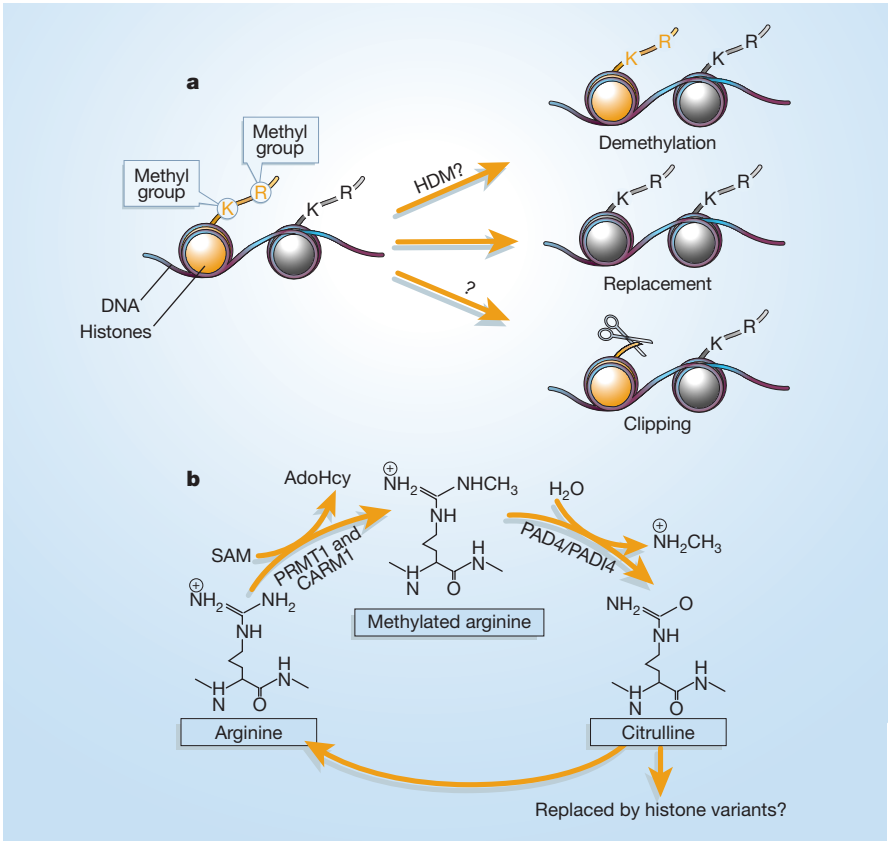

Figure 1 **Reversing methylation in histone proteins.** a, **Three theoretical mechanisms: top, enzymatic demethylation, perhaps regulated by the HDM enzyme; centre, replacement of methylated histones by unmethylated variants; bottom, clipping of histone 'tails', by enzymes unknown. K, lysine; R, arginine.** b, **Wang *et al.*[4] and Cuthbert *et al.*[5] have discovered another mechanism: they show that methylated and unmethylated arginine amino acids in two histone proteins can be converted to citrulline. A possible cycle of events is shown. Methylation of arginines is accomplished by the PRMT1 or CARM1 enzymes, with SAM (*S*-adenosyl-L-methionine) as cofactor; AdoHcy (*S*-adenosyl-L-homocysteine) is released. Conversion to citrulline is achieved by PAD4/PADI4. It remains unknown what then happens to citrulline — whether it is converted back to arginine (by an aminotransferase enzyme, perhaps), or whether histones containing citrulline are replaced by unmodified versions.**

100 YEARS AGO

The recent tube operations in London have brought to the surface specimens of the London Clay from different districts. Samples of this clay taken from such different points as Hyde Park Corner, Brompton Road, and Haverstock Hill have been tested in the physical laboratory of the South-western Polytechnic for the presence of a radio-active gas by Mr. H. Cottam, and he has been unable to detect with his apparatus any marked quantity of active gas from the clays. With the same apparatus he has detected quite easily the radio-active gas from the water of a deep well… which goes below the clay to the greensand. We have come to the conclusion that the London Clay forms a floor through which the radio-active gas does not penetrate; or it may be said that the radio-active substance only travels when the water with which it is associated can travel. This is an argument in support of Prof. J. J. Thomson's view, that the radio-active gas, which he found in deep well waters, arises from the splitting up of a trace of soluble radium salt which comes up with the water.

From *Nature* 6 October 1904.

50 YEARS AGO

Little more than a hundred years have passed since hunters such as Gordon Cumming were at their work among the vast herds of big game in the southern quarter of Africa, and little more than fifty since Selous was making a living by ivory hunting somewhat farther to the north. All those countless thousands of animals have now disappeared for ever… The opening-up and white settlement of East Africa did not come until later, but although the same process of decimation began, and many parts of the country have been cleared of game, it is still possible to see great herds of wild animals that recall the accounts of conditions in South Africa given by the early travellers. Nevertheless, the presence of enormous herds of game animals is quite incompatible with the economic exploitation of the country and the rapid expansion of the native population; the game must go, and there can be no hope of its survival outside the National Parks and game reserves… As yet, however, there is practically no information available on the biology of these mammals, information that is essential for successfully managing such parks and reserves.

From *Nature* 9 October 1954.

residues can only be mono- or di-methylated.

Histone methylation has been linked to biological processes ranging from the regulation of gene transcription, to the inactivation of one copy of the X chromosome in females, to RNA-mediated gene silencing[2,6,7]. One way in which it works is by serving as a docking site for other proteins[2]. The nature of a specific methylation determines the protein that it recruits, which in turn dictates the biological outcome (see, for example, ref. 6).

Unlike other histone modifications, methylation has generally been regarded as stable[3] — a notion that comes from early studies showing that histone proteins and methylated lysine or arginine residues within them have similar turnover rates[8]. But although a non-reversible methyl mark would fit with a role for histone methylation in long-term gene silencing, it is not compatible with situations in which rapid reversal of gene expression takes place. To solve this paradox, mechanisms including enzyme-catalysed demethylation, replacement of methylated histones by unmodified histones, and clipping of methylated histone 'tails' have been proposed[3,9] (Fig. 1a) — but none has yet been demonstrated experimentally. Now, however, Wang *et al.*[4] and Cuthbert *et al.*[5] have found that the human enzyme peptidylarginine deiminase 4 (PAD4/PADI4) can catalyse the conversion of methylated arginines to citrulline, providing yet another mechanism by which histone methylation levels could be controlled (Fig. 1b).

How is it that two groups independently identified the same enzyme and came to similar conclusions? Previous studies[10] established that arginine residues in other proteins can be converted to citrulline by enzymes of the peptidylarginine deiminase family. Of this family, only PAD4/PADI4 is found in the nucleus; moreover, its expression correlates with the appearance of citrulline in histones[11]. These facts made it a good candidate for a histone arginine demethylase.

So Wang *et al.* and Cuthbert *et al.* carried out direct tests of PAD4/PADI4 *in vitro*. They found that it 'deiminated' numerous unmethylated arginines — converted them to citrulline — in histones H3 and H4. It also decreased the amount of arginine methylation that is catalysed by PRMT1 and CARM1 in both histones. Notably, at the same time that the amount of this methylation decreased, the quantities of citrulline in both histones increased. The detection of methylamine as a released product[4] supports the idea that methylated arginine is a genuine substrate of this enzyme. So, PAD4/PADI4 can catalyse both the deimination of unmethylated arginine and the 'demethylimination' of methylated arginine *in vitro*. As Wang *et al.* find, it can also do so in granulocyte cells.

What are the consequences of these reactions? First, as Cuthbert *et al.* show, the conversion of histone arginines to citrullines actually prevents histone methylation by CARM1. Second, Wang *et al.* find that demethylimination of methylated histone arginines reverses the effects associated with methylation. So, PAD4/PADI4 presumably antagonizes the functions of the methylating enzymes CARM1 and PRMT1. For instance, previous studies have linked histone arginine methylation by these enzymes to transcriptional activation by nuclear hormone receptors[12–15] — so PAD4/PADI4 is likely to repress such transcription. Indeed, both groups link the recruitment of PAD4/PADI4 to an oestrogen-responsive gene, pS2, with the appearance of citrullinated histones and the downregulation of this gene.

These papers[4,5] provide convincing evidence that the methylation of arginine amino acids in histone proteins can be reversed enzymatically. But, as ever, the findings raise questions. For example, it seems that PAD4/PADI4 has a very loose substrate specificity *in vitro*. It works on both methylated and unmethylated arginines. And the arginine residues it deiminates are not limited to the sites that are targeted by CARM1 and PRMT1; indeed, arginine 8 in histone H3, a site not known to be methylated by either enzyme, is the preferred target[4]. There might be an interplay between the citrullination of H3 arginine 8 and the methylation of H3 lysine 9, but for now the significance of this citrullination remains unknown.

The second issue worth noting is that, *in vitro*, PAD4/PADI4 cannot demethyliminate H4 or H3 peptides containing di-methylated arginines[4,5]. This presents a puzzle. Although mass-spectrometry analysis[14,15] identified mono-methyl groups as the major methyl form on arginine 3 in histone H4, most arginines 17 and 26 in histone H3 become dimethylated after incubation with CARM1 *in vitro*[16]. How, then, can this methylation be removed? Although an unidentified enzyme might be required, the available evidence suggests that PAD4/PADI4 is responsible: for instance, in granulocytes, activation of this enzyme led to less methylation on H4 arginine 3 and H3 arginine 17, as analysed using site-specific antibodies that recognize dimethylated arginine[4]. The most likely solution is that PAD4/PADI4 can work only on intact chromatin substrates. Alternatively, it might need to work with a partner.

As well as revealing that arginine methylation can be reversed, the new papers[4,5] show that a new form of histone proteins — containing citrulline residues — exists in cells. This finding, too, raises questions. How stable are citrullinated histones? Do they have any effects on chromatin structure? Can other proteins recognize and bind to them? And what is the fate of these histones? With regard to the last question, the transient presence of citrullines in the pS2 gene[5] suggests

that their rapid removal must be possible. The availability of site-specific antibodies against citrullinated histones will allow at least some of these issues to be addressed. And one final question: is the methylation of lysine residues in histone proteins also reversible? ■

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Palaeontology

Ecology of ice-age extinctions

John Pastor and Ron A. Moen

The last ice age saw the extinction of numerous large mammals — but perhaps not as many as was thought. The woolly mammoth survived to much more recent times, and so, it now seems, did the Irish elk.

Sabre-toothed tigers, mastodons, woolly mammoths — these and many other spectacular large mammals are generally thought to have become extinct about 10,000 years ago, at the end of the Pleistocene epoch, otherwise known as the last ice age. But it's becoming clear that some of these species clung on tantalizingly close to the present day. Thomas Jefferson's instruction to Meriwether Lewis and William Clark to search for live woolly mammoths in the American West in 1804 was perhaps a little optimistic. But the species survived on Wrangel Island in the northeastern Siberian Arctic until some 4,000 years ago[1], making it contemporaneous with the Bronze Age Xia Dynasty in China. On page 684 of this issue, Stuart *et al.*[2] report that another charismatic ice-age mammal that was thought to have become extinct 10,000 years ago — the giant deer or Irish elk (*Megaloceros giganteus*) — survived in western Siberia to the dawn of historic times. The finding lends weight to the idea that there is no one explanation for the so-called Pleistocene extinctions.

The Irish elk (Fig. 1) must have cut an impressive figure, standing more than two metres high at the shoulder — about the same as a bull moose, the largest living member of the deer family. But when and why did it become extinct? In their investigation, Stuart *et al.*[2] began by carrying out radiocarbon dating of five skeletal specimens, including a complete skeleton of an antler-bearing male. By combining this information with maps of the specimens' locations, they show that Irish elk were widespread in Europe — from Ireland to Russia, and from Scandinavia to the Mediterranean — before 20,000 years ago. But by the last glacial maximum 15,000 years ago, they may have been restricted to refuges in the shrub steppes of central Asia. From there, Irish elk apparently recolonized northwestern Europe following the retreat of the Alpine and Scandinavian ice sheets during a period of climatic warming. The European population made a last stand in the British Isles before dying out 10,500 years ago, but the Siberian population persisted for another 3,000 years.

What caused the extinction of so many large mammals 10,000 or so years ago? Human hunting[3], changes in climate or vegetation, or both[4], are often proposed to be causal factors. But the "'ragged' nature" (to use Stuart and colleagues' phrase) of these Late Pleistocene extinctions, with isolated pockets of populations surviving for longer, suggests that the extinctions have a complex ecology, with no single mechanism responsible for the demise of every species in every location.

Theories for both the expansion and the extinction of Irish elk populations, for instance, often focus on the animals' huge antlers, which weighed 40 kilograms and spanned 3.5 metres, making them 30% larger than those of modern moose. It has been suggested[5] that female Irish elk selected males with large antlers, as this might have signified an ability to find sufficient food to support building and shedding a rack each year. This ability would then be passed on to their male progeny.

But the large antlers, which contained as much as 8 kilograms of calcium and 4 kilograms of phosphate, would have posed a large annual nutritional burden on bulls[6]. The antlers would also be physically unwieldy in dense forests[7]. So both physical and nutritional constraints probably restricted the

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Figure 1 **Prehistoric giant — artist's impression of the Irish elk.**

intrinsic mortality (and a longer intrinsic lifespan) when all else is equal, many organisms face a trade-off between higher levels of reproduction or lower levels of intrinsic mortality. One of the main reasons that senescence occurs is because repair is costly: resources that are devoted to maintaining an organism are not available for reproduction. In the 1950s, Peter Medawar[2] and George Williams[3] pointed out that high extrinsic mortality could favour shorter intrinsic lifespan. Why, they reasoned, should an organism invest in costly repair that will probably only ensure that it is in prime physical condition when its life ends? Higher extrinsic mortality should favour low investment in repair, and thus a high intrinsic mortality and a short intrinsic lifespan.

But this reasoning didn't take account of two further factors. One is that higher extrinsic mortality also slows the rate of population growth, and more slowly growing populations are expected to evolve to have lower rates of intrinsic mortality and a longer lifespan[4,5]. The other is the interaction between extrinsic mortality factors and physiological repair or maintenance[5,6]. If predators can be evaded by fast, but not by slow prey, greater predation risk should select for greater maintenance of the body systems essential for fast movement. This higher level of repair would then prolong intrinsic lifespan.

Higher extrinsic mortality (more predators) could also have indirect effects that Medawar and Williams did not consider. For example, it reduces population size, which in turn increases the abundance of food or other resources. These changes may have their own effects on both population growth and the level of intrinsic mortality favoured by selection. Other complications arise if the mortality factor has a greater effect on some ages than on others[5,6] — if, for example, predators prefer to capture larger, older prey. As a result of these complicating features, many types of mortality are expected to reduce intrinsic death rates at some ages while increasing them at others[6]. In any event, theory suggests that higher extrinsic mortality will produce evolutionary conditions that can either extend or shorten the intrinsic lifespan.

Given these complexities, the curious feature of previous observational[7] and experimental work[8] has been its support for the Medawar–Williams prediction. There have been exceptions[9], if only suggestively so. But the almost unanimous evidence that high extrinsic mortality is associated with shorter lifespan is puzzling because there is no reason to believe that the conditions that produce the opposite outcome are rare in nature.

So it is reassuring that Reznick *et al.*[1] found longer intrinsic lifespans in guppies from populations characterized by higher predation rates. The authors also looked at whether these evolutionary changes might be an indirect consequence of predator-caused deaths, such as the availability, in natural settings, of more food for the remaining guppies. Reznick and colleagues' study is unique in examining this effect. They found that food alone could not account for the difference in intrinsic mortalities seen in their experiments, but that having more food enhanced the lifespan-lengthening effect of a high-predation background.

There is no doubt that the guppies from high-predation sites have both longer intrinsic lifespans and longer reproductive spans. But is it valid to conclude that they have slower senescence? This is a more difficult question. A hypothetical population with no senescence (that is, no age-related decline in survival or reproduction) could still have a short lifespan if it had a high mortality rate that was independent of age. If guppies from high-predation sites begin their adult life with a lower rate of intrinsic mortality than those from low-predation environments, they could have the same rate of increase with age in their mortality rate, but would still have a longer lifespan. One could then argue that the two populations had identical rates of senescence. Some measures of the rate of change of intrinsic mortality with age suggest that senescence is delayed in guppies from high-predation sites.

However, senescence encompasses relationships between many different components of fitness and age, none of which can be adequately summarized by a single number: there are many potential measures of the rate of senescence, and conclusions about this rate depend on the measure chosen. It might be possible to make a case that guppies from high-predation environments are more robust, but age at a rate equal to or higher than that of low-predation guppies. Regardless of the mathematical measure used to quantify the rate of senescence, the work of Reznick *et al.* clearly shows that rates of senescence differ among the different components of fitness examined: survival, reproduction or swimming performance. The reasons for these differences are not yet understood.

It would be surprising if guppies were the only species for which an added risk of mortality lengthens intrinsic lifespan. Similar studies on other species will help us understand the underlying reasons why Medawar and Williams' predictions hold for some species and not for others. Such studies should follow Reznick and colleagues' lead in quantifying declines in several fitness components and studying the indirect ecological consequences of higher extrinsic mortality. ■

D. GHALAMBOR

Figure 1 **Star survivor — the longest lived of the guppies studied by Reznick *et al.*[1]. The photo was taken shortly before her death at the age of 1,464 days.**

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Correction

In Yi Zhang's News and Views article "Molecular biology: No exception to reversibility" (*Nature* **431,** 637–639; 2004), there were errors in Fig. 1b. In the side chain of citrulline, a double bond should have been shown between NH and O, rather than between NH and NH_2. Several of the connecting atoms are erroneously shown as H rather than N. And the leaving methylamine should have been represented as $+NH_3CH_3$ rather than $+NH_2CH_3$.