Given the apparent conformational equivalence of the reactive phosphate in these two enzymes, does the hairpin ribozyme also use this catalytic strategy? On this point we can only speculate. RNA is inherently compromised in its ability to promote general acid–base chemistry. Unlike histidine, which has a near-neutral acidity constant (pK_a), all four of RNA's nucleotides have pK_a values that are either too high or too low to be useful in proton transfer at neutral pH. So perturbation of an active-site residue's pK_a would be required for it to function like either of the histidines in RNase S. Such perturbations are not common, but they have been observed in RNA and are proposed to play key roles in the hepatitis delta virus ribozyme and the ribosome reaction mechanisms[10–13]. The hairpin crystal structure alone cannot define which, if any, of the bases have an unusual pK_a, but it limits the catalytically relevant candidates to four: G8, A9, A10 and A38. Of these, G8 contacts the nucleophile, whereas the three adenines line a small pocket adjacent to the 5′-OH leaving group (Fig. 1a).

If it does turn out that this small ribozyme uses a nucleotide with a perturbed pK_a for catalysis, then it may be mechanistically comparable to its much larger catalytic cousin the ribosome, which performs protein synthesis and has an active-site adenine with a near-neutral pK_a (refs 12, 13). The molecular weight of the ribosome is more than a hundred times that of the hairpin, so this small ribozyme would define a minimal RNA element that can use this sophisticated mechanistic strategy. In this way the hairpin structure may provide clues about an ancient RNA world in which there were no protein enzymes with which to make any comparisons. ■

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Physiology

A hunger for cannabinoids

Raphael Mechoulam and Ester Fride

Cannabinoids — molecules found naturally in the body, as well as in cannabis — stimulate appetite. Leptin, a hormone produced by body fat, decreases appetite. The effects of these molecules have now been linked.

The hormone leptin and neurotransmitter molecules called cannabinoids both have effects on appetite, but do they work together or separately? Writing on page 822 of this issue[1], Di Marzo and colleagues provide an answer. They show that — at least in rodents — leptin modifies the levels of cannabinoids found naturally in the body (endocannabinoids), and that endocannabinoids and leptin belong to a common system that regulates body weight.

Cannabis users are well aware of the appetite-enhancing effects of the drug. Indeed, the major active component of cannabis, a cannabinoid called Δ^9-tetrahydrocannabinol, is used by doctors to prevent some of their patients — particularly people suffering from AIDS — from losing too much weight as a result of their illness[2]. Fatty-acid-derived endocannabinoids (such as anandamide, isolated from pig brains in 1992; ref. 2) have many of the same properties as Δ^9-tetrahydrocannabinol. For example, they all bind to cannabinoid receptor proteins, found on nerve or immune cells. Anandamide also increases food intake in rats and mice[3,4], an effect that is prevented by a drug that selectively blocks the CB1-type cannabinoid receptor[3,4]. The implication is that the endocannabinoids and their receptors are involved in feeding behaviour, and Di Marzo *et al.*[1] now confirm this. They find that genetically engineered 'knockout' mice that lack the CB1 cannabinoid receptor eat less than their normal littermates after being deprived of food (to enhance appetite).

The hormone leptin, meanwhile, is produced by fat tissue and affects the hypothalamus — a region in the brain that is important in weight regulation. A region of the hypothalamus called the arcuate nucleus contains neurons with receptors for two appetite-stimulating peptides (neuropeptide Y and agouti-related protein), as well as receptors for two peptides that reduce appetite (α-melanocyte-stimulating hormone and cocaine-and-amphetamine-regulated transcript). Leptin directly suppresses the activity of the two appetite-stimulating peptides, and stimulates the activity of the appetite-reducing ones, thereby decreasing appetite. Other molecules are indirectly affected by leptin. These include melanin-concentrating hormone and a family of neuropeptides called orexins, all of which enhance appetite, as well as corticotropin-releasing hormone and oxytocin, which cause mice to eat less and to lose weight.

So, leptin is considered to be a key signal through which the hypothalamus senses the nutritional state of the body. A decrease in the amount of body fat and other energy reserves, which occurs after fasting, reduces the level of leptin. There is therefore less of this hormone around to suppress appetite-stimulating peptides and to stimulate appetite-reducing molecules, so food intake is stimulated. Higher levels of body fat and other energy reserves increase the level of leptin, leading to a reduction in food intake. This intricate mechanism is important for maintaining weight within a narrow range[5,6].

Di Marzo *et al.*[1] find that this weight-regulating system is even more complicated than previously thought. They show that administration of leptin reduces the levels of the endocannabinoids anandamide and 2-arachidonoyl glycerol in the hypothalamus of normal rats. Further evidence strengthens the idea that leptin downregulates endocannabinoids. In a strain of obese rats in which leptin activity is impaired, the levels of endocannabinoids are higher than normal[1]. The same is true of obese *ob*/*ob* mice, which have an inherited lack of leptin, and of obese *db*/*db* mice, which have defective leptin receptors. Endocannabinoid levels are not affected in the cerebellum of these mice; this brain structure is commonly associated with motor coordination, but not with feeding. It is not yet clear how leptin might lower endocannabinoid levels specifically in the hypothalamus. There seems to be a link between leptin and the biological pathways by which the endocannabinoids are made, but further studies will be needed to pin down the details.

So far, only the food-related connection between leptin and endocannabinoids has been studied. But both types of molecule also affect other body functions, and it would be interesting to see if, and how, they interact. For example, obese *ob*/*ob* mice are sterile, and this is associated with low levels of leptin[7]. Indeed, these mice become fertile when treated with leptin[7]. Moreover, endocannabinoids are also involved in reproduction. Low levels of anandamide in mice are associated with the uterus being receptive to embryo implantation; higher levels coincide with the uterus being less receptive[8]. An enzyme called fatty acid amide hydrolase is needed to break down anandamide;

decreased activity of this enzyme, presumably leading to increased anandamide levels, correlates with spontaneous abortion in humans[9].

The results of Di Marzo *et al.* show that, at least with regard to appetite and feeding, the levels and effects of leptin and anandamide are inversely related. Is a similar balance important in controlling reproduction? No role for leptin after mating and fertilization (for example, in embryo implantation) has been found, and apparently it has no role in feeding during pregnancy[7]. But perhaps leptin regulates endocannabinoids in the reproductive system, as it does in nutrition.

Endocannabinoids are also involved in regulating several other processes, including stress[10]. Moreover, endocannabinoid-induced 'reward' effects (that is, the pleasure derived from eating tasty foods, for example) have come under scrutiny in attempts to unravel the addictive potential of cannabis[11]. These effects are mediated by the mesolimbic brain region, and involve the neurotransmitter dopamine. Leptin also plays a role in stress[12], and may likewise affect the mesolimbic dopamine pathway and the satisfaction derived from eating[13]. Here again, as in the control of feeding, leptin and the endocanabinoids seem to work in opposite directions. Endocannabinoids activate the 'stress' (hypothalamic–pituitary–adrenal) axis[10], and enhance the 'reward' experience. In contrast, leptin inhibits the hypothalamic–pituitary–adrenal axis[12] and attenuates brain reward circuitry[13]. It remains to be seen whether or not leptin affects the involvement of endocannabinoids in stress and in the reward system. But it is clear from Di Marzo *et al.*'s work[1] that much can be learned from studying the interactions between these important molecules. ■

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Global change

A piece in the CO_2 jigsaw

Dorothee Bakker and Andrew Watson

A study of the year-to-year variation in net CO_2 uptake by the oceans helps in assessing the mechanisms of global climate change.

The burning of fossil fuels releases carbon dioxide into the atmosphere, where its increasing concentration promotes global warming. Some of this CO_2 is absorbed by terrestrial systems and the oceans. But the location and year-to-year variation of these 'sinks' are badly quantified. Writing in *Geophysical Research Letters,* Loukos *et al.*[1] describe an innovative approach to estimating the exchange of CO_2 between the air and the sea for the equatorial Pacific Ocean over a ten-year period. One remarkable finding, narrowing the gap between other studies[2–6], is that there is little variation from year to year in such CO_2 air–sea transfer in the tropical oceans.

During the period 1981–92, about 5.5 gigatonnes of carbon (Gt C) were released annually through fossil-fuel burning; 1 Gt C is 10^{15} g C. Of that, a variable amount, ranging from 1.4 to 5.0 Gt C, has remained in the atmosphere[7]. Research using the carbon-13 (^{13}C) isotopic content of atmospheric CO_2 has ascribed this year-to-year variation to the oceanic CO_2 sink being highly variable[2,3]. By contrast, two other studies[4,5], which were further constrained by measurements of the atmospheric CO_2 concentration, indicate that oceanic and terrestrial carbon reservoirs have each contributed ± 0.6–1.0 Gt C to the interannual variability.

Using a different approach, Loukos *et al.*[1] have come up with a year-to-year variation of ± 0.4 Gt C for tropical CO_2 air–sea transfer. The tropical oceans are an important contributor to variation in global oceanic CO_2 uptake[4,6], so this finding implies that oceanic involvement in interannual changes of CO_2 storage may be smaller than atmospheric studies[4,5] suggest. But the estimate is higher than that inferred in another study using oceanic data[6].

Conventional ocean studies estimate CO_2 air–sea exchange from the gradient in the CO_2 partial pressure (p_{CO_2}) across the sea surface and from a gas-transfer velocity, which depends on wind speed. One of the obstacles to this approach is the incomplete coverage, in both space and time, of surface-water p_{CO_2}. Instead, Loukos *et al.* take surface-water temperature and salinity as proxy measurements for dissolved inorganic carbon and alkalinity. Their rationale is that there is a close coupling between the physical and biological processes affecting these carbonate parameters in the surface waters of the equatorial Pacific Ocean. Satellite observations and compilations of shipboard data provide good coverage of temperature and salinity. From dissolved inorganic carbon and alkalinity, Loukos *et al.* calculate the p_{CO_2} in surface water, and finally CO_2 air–sea exchange across the equatorial Pacific Ocean for the years 1982–93.

A central question in this research is whether studies based on atmospheric CO_2 data overestimate, and those based on oceanic CO_2 underestimate, the variability of the ocean CO_2 sink. One unknown factor in studies using atmospheric ^{13}C values is the relative contribution of plants using specific photosynthetic systems (C_3 and C_4) to terrestrial plant growth[8], as they affect the isotopic composition of CO_2 in different ways. Another uncertainty is the badly quantified difference in the ^{13}C isotopic content of outgoing and incoming fluxes between the atmosphere and oceanic and terrestrial carbon reservoirs[8], as fossil-fuel CO_2 emissions alter the ^{13}C content of atmospheric CO_2 with time. Finally, possible errors in atmospheric transport models[4], the relatively short time series of atmospheric ^{13}C measurements, and the dominance of atmospheric monitoring stations at marine sites[4,9] contribute further to the uncertainty of estimates based on atmospheric CO_2 data.

A major unknown in the oceanic estimates is the relationship between air–sea gas transfer and wind speed. The oceanic uptake of the radioisotope ^{14}C, originating from natural sources and from atomic-bomb tests, provides a value for the global average transfer velocity. Otherwise there are too few direct measurements of air–sea gas exchange in the open ocean to allow the precise nature of the relationship between gas exchange and wind speed to be determined. Wind speed accounts for up to 30% of the year-to-year variation of net global oceanic CO_2 uptake[6], so here is a considerable source of error.

Further uncertainty stems from the techniques used to obtain coverage of surface-water p_{CO_2} for an entire ocean basin. The p_{CO_2} estimates of Loukos and colleagues have a slightly lower spatial variability than corresponding shipboard observations, resulting in an underestimation of the spatial, and possibly of the temporal, variability in CO_2 air–sea exchange. Lee and colleagues' study[6] probably underrates year-to-year variation by normalizing surface-water p_{CO_2} data to a