containing atoms of only one velocity (perhaps obtained with laser cooling) the atoms can maintain the superposition of two phase-shifted fields; each interaction merely recreates the pre-existing superposition. Like Schrödinger's cat, the two fields are macroscopic states, and they are distinct states. Conventional Schrödinger cats rapidly die on interaction with a dissipative environment: their decay depends on how distinct are the components. But these cats are fed 'quantum food' in the form of the injected atoms in superposed states[5].

Brune *et al.* show that their cats can be made with existing experimental technology: the field damping time in cavities currently available is 0.1 seconds. Schrödinger cat states involving as many as 100 photons could be generated and observed, using a modified interference technique. If realized, they could form the basis of much-needed experimental investigation of the creation of quantum coherence and its destruction by the environment, a subject of intense theoretical speculation. □

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RNA CATALYSIS

The universe expands

Peter B. Moore

FOR over a decade, RNA enthusiasts have been claiming that a race of self-replicating entities that used RNA molecules both for genetic purposes and as catalysts for their metabolic reactions were the progenitors of us all. Their advocacy stems, of course, from the knowledge that some RNAs — ribozymes — possess catalytic activity. The chemistry catalysed by the ribozymes identified so far is not all that exciting, however, in that they promote nucleophilic attacks by either hydroxyl groups or water on phosphates. The faithless have questioned whether such a limited catalytic palette could sustain self-replicating entities, and so have cast doubt on the whole concept.

Two papers in last week's *Science*[1,2] should silence some of the critics. They show that ribozymes can catalyse nucleophilic addition and elimination reactions involving carbonyl groups, and that a ribozymal activity of that sort is almost certainly responsible for the formation of peptide bonds during protein synthesis.

The peptidyl transferase activity of the ribosome is intrinsic to its large subunit[3], and those in the business have known for several years that the Noller laboratory at Santa Cruz was trying to find out whether that activity is RNA-driven. The first formal report[1] on what has been accomplished is one of the two papers in question here.

The fragment reaction assay for peptidyl transferase activity[4] is the key to this work. What is measured is the transfer to puromycin of amino acids esterified to an oligonucleotide isolated by nuclease digestion from the 3′ end of an aminoacylated transfer RNA. Its virtue is that it can use isolated ribosomal subunits, independent of tRNA binding, messenger RNA binding, interactions with the small subunit, and all the other complications of protein synthesis.

Reconstitution studies carried out in the 1970s suggested that large ribosomal subunits from *Escherichia coli* containing less than their normal complement of protein retain fragment reaction activity[5], and what Noller and his colleagues wanted to see was whether all the protein could be removed without loss of activity. Their best results were achieved using large subunits from the extreme thermophile, *Thermus aquaticus*. Proteins were stripped from them by treatment with sodium dodecyl sulfate (SDS), proteinase K and phenol (at 4° C). Done singly or in combination these treatments had no effect on activity, but any treatment that disrupted RNA tertiary structure abolished activity completely. The 50S subunits from *E. coli*, which presumably are not as tough, retained some activity in the face of SDS and proteinase K treatment, but would not tolerate phenol extraction in the cold. The activity remaining in stripped subunits had the same sensitivity to antibiotics as the activity that was seen in intact subunits.

These observations do not constitute absolute proof that the ribosomal peptidyl transferase is ribozymal because 5 per cent of the protein cannot be removed from *T. aquaticus* subunits by these means. It nevertheless suggests how bets should be placed in the future.

The second of the two papers, which also involves Noller, comes from Cech's laboratory at Boulder[2]. It describes the conversion of the self-splicing intron from *Tetrahymena thermophila* from a catalyst of phosphodiester transesterifications into a catalyst of carboxylate ester hydrolysis. The guide sequence of the *Tetrahymena* nucleotide intervening sequence (IVS), which normally aligns its RNA substrates in the activity site by base pairing, was altered by mutation so that it would bind the 3′ hexanucleotide of $tRNA^{fMet}$. The site was engineered so that *N*-formyl-L-methionine esterified to the hexanucleotide's 3′ ribose would occupy the active site. The mutated enzyme retains the ability to cleave substrate RNAs whose sequences have been altered so that they will bind to the mutated guide sequence, but now it also weakly catalyses the hydrolysis of the amino acid ester in question.

'Weakly' is very weak indeed — a five-fold increase in rate is all that is observed. Nevertheless, tests with variants of the IVS demonstrated that mutants lacking nuclease activity are also inactive as carboxylic acid esterases. In addition, the esterase activity depends on magnesium, as does the IVS nuclease activity, and it can be saturated by raising the substrate concentration. Thus this esterase activity is likely to be due to chemistry occurring at the normal active site of the IVS, an important point given the small effect observed.

The IVS esterase activity differs from the normal IVS transesterification activity in its nucleophile specificity. The latter accepts either guanosine (well) or water (poorly) as its nucleophile, but only water serves for the former. As the authors point out, the transesterification reactions of (tetrahedral) phosphate groups depend on in-line attack by nucleophiles. Carbonyl groups are trigonal, and nucleophiles attack them normal to the plane. Thus a guanosine positioned in the IVS active site for participation in a phosphate transesterification reaction will not be in a position to attack a carboxylate ester, if the carboxylate ester binds in the position normally occupied by phosphate groups.

Taken together, the two new papers deliver a simple message: we must expand our view of what RNAs can catalyse. That expansion is not a large one chemically; nucleophilic additions to phosphates are not all that different from nucleophilic additions to carbonyl groups. But it is of the utmost significance biologically. The type of RNA catalysis demonstrated in the papers is that on which the entire translation process depends. The possibility that there once were RNA entities capable of both replicating RNA and synthesizing protein seems a lot more reasonable now than it did a few months ago. □

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