Tubulin synthesis

Controlling mRNA lifespan

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MESSENGER RNA in animal cells is generally very stable, with half lives of the order of several hours being the norm. Thus, the level of ribonuclease in cytoplasm must be extremely low, with what little there is probably held in check by RNasin, the RNase inhibitor which is found in liver, reticulocytes and placenta (probably all cells, in fact). This being so, it is the scattered cases of unstable mRNA that require explanation. The case of β-tubulin mRNA is a well-known example. It has been known for many years that tubulin synthesis is shut off by drugs that disassemble microtubules, and is enhanced by microtubule stabilizers like taxol[1]. Neither transcriptional nor translational control plays a significant part in the regulatory circuit; colchicine causes tubulin mRNA to become unstable and, conversely, taxol stabilizes it. The effect is specific for tubulin mRNA, and all the evidence suggests that unpolymerized αβ dimers regulate their own synthesis by a feedback mechanism acting on their mRNA. Studies of β-tubulin mRNA in Cleveland's laboratory at Johns Hopkins University pinned down the regulatory sequences to the 5′ end of the gene[2]. This in itself is unusual; destabilizing sequences usually lie in the 3′ untranslated regions (well documented examples are found in histones[3], GM-CSF (granulocyte-monocyte colony stimulating factor)[4], c-*fos*[5,6] and the transferrin receptor[7,8]). But, even more surprisingly, the latest work from Cleveland's group, reported on page 580 of this issue[9], shows that the instability of the tubulin mRNA depends on recognition of the first four amino acids of β-tubulin, Met-Arg-Glu-Ile (MREI), as they emerge from the ribosome, and not on the sequence or structure of the mRNA as such.

The evidence for this view is comprehensive and elegant. Alterations to the sequence of the mRNA permit normal regulation as long as the N-terminal amino-acid sequence stays the same, whereas single nucleotide changes that alter the protein sequence stop the regulation. Regulation works only when the sequence is translated in the correct frame, and the peptide sequence must be located at the N terminus of the nascent chain. Finally, the sequence must be followed by a reasonable length of polypeptide, long enough for it to emerge from the ribosome. If the protein containing the sequence is too short, or if peptides are released prematurely by puromycin, the mRNA is stabilized. These observations suggest a model in which something, presumably a nuclease or a nuclease activator, binds to ribosomes via the N terminus of nascent β-tubulin when the concentration of αβ-tubulin dimers gets too high. Perhaps free tubulin dimers bind to the sequence MREI, carrying a nuclease with them, but how such a mechanism might work is unclear.

How do these findings fit with what is known of other examples of regulated mRNA stability? The mechanism of mRNA turnover is still poorly understood, but several recent observations begin to suggest a fairly solid outline. The first significant result is that intermediates in the destruction of the mRNA are rarely, if ever, seen. This is most easily understood if the cell contains exonucleases that rapidly degrade the fragments of mRNA produced by the first (rare) endonucleolytic cut. Why do these enzyme(s) not attack intact mRNA? Almost certainly it is protected at both ends, at the 5′ end by the 'cap' structure and at the 3′ end by the poly(A) tail. Thus, a single endonucleolytic clip renders it vulnerable to the scavenging exonucleases. Ross and his colleagues have now identified a 3′ → 5′ exonuclease which is probably responsible for part of this process, and show that histone mRNA is degraded 3′ → 5′ *in vitro*[10]. Histone mRNA lacks a poly(A) tail and should therefore be highly vulnerable to destruction by this activity; however, it contains a structure at the 3′ end which provides partial and regulated protection[3]. The story is complicated, with echoes of the tubulin story in that destruction of the mRNA requires active translation of the mRNA and the accumulation of free (that is, not bound to DNA) histones in the cytoplasm[11]. But there is as yet no evidence for any involvement of the nascent chain in this case; although like Cleveland, Marzluff and his colleagues propose[11] that the translating ribosomes carry a nuclease with them.

Two other recent papers provide support for the basic idea that the ends of mRNAs are sensitive targets for destruction. Thus, Harland and Misher made a circular (endless) mRNA and injected it into amphibian (*Xenopus*) embryos[12]. In agreement with the model, such RNA molecules are exceptionally stable. These circles should be a useful test bed for potential destabilizing sequences and the factors that attack them. The only flaw in this idea is that circular mRNAs cannot be read by ribosomes[13], so the stability of these structures might arise as much from their failure to be translated as from their endlessness. Again using *Xenopus,* Hyman and Wormington[14] report that the translational inactivation and disappearance of the mRNAs for ribosomal proteins that accompanies oocyte maturation is preceded by removal of their poly(A) tails. Something of the same sort happens to c-*myc* and c-*fos* mRNA, where degradation is apparently preceded by shortening of the poly(A) tail[5,15]. This is a specific process which requires sequences in the 3′ untranslated region of the mRNAs; stable mRNAs like globin do not show poly(A) shortening. Other mRNAs in *Xenopus* oocytes suffer precisely the opposite fate, getting longer poly(A) tails at the same time that the ribosomal

100 years ago

I HAVE read with extreme interest the abstract of a paper by Prof. Ewart, on the "Structure and Development of the Electric Organ of *Raia radiata.*" Organic nature is full of organs which are either wholly or partly functionless. The Darwinian philosophy almost invariably explains them as structures which must have once been useful, and have become functionless by atrophy or disuse.

This is a natural and necessary consequence of the doctrine which ascribes all organic structures to utility as a physical cause. Utility as a mental purpose is kept out of sight.

I have always thought that if the doctrine of development be true, functionless organs must be the germs of potential use, and not necessarily at all the remains of past actual use. Here we have a case in which a distinguished physiologist detects, or thinks he can detect, an organ in process of being built up for the discharge of a very definite and peculiar function.

This fact does not tell against development or evolution. But it does tell, and tells fatally, against the element of fortuity, which is inseparable from the idea of "natural selection." ARGYLL

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