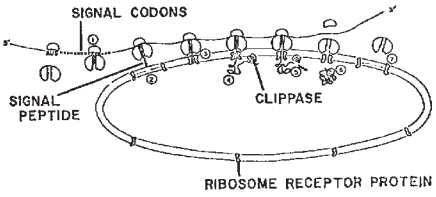
tion factors to viral messengers is favoured—hence their preferential translation.

In the section dealing with the topography of protein synthesis G. Blobel (Rockefeller University, New York) presented a very appealing model to explain the translation of mRNA for secretory proteins on membrane-bound ribosomes. It requires that mRNAs for secretory proteins code for an N-terminal peptide that is recognised by the membrane, resulting in the attachment of ribosomes to the membrane so that the growing protein is vectorially transported into the intracisternal space. This is illustrated in the diagram: the numbers indicate sequential steps in the synthesis of the protein. Bernard Mechler (University of Cambridge) proposes a slightly different scheme since he has to explain his observation that the binding of ribosomes to the membrane can be mediated by the mRNA directly rather than its translational product. H. Lodish (MIT, Cambridge) showed that translation of the polycistronic Sindbis virus RNA can take place in the cytoplasm or on membranes using the same ribosomes but producing different proteins. He concludes that the N-terminus of the nascent chain



must specify the binding of ribosomes to membranes.

The participants left the conference with the impression of many examples of cytoplasmic control of protein synthesis but few explanations of the mechanisms involved, an admirable situation sure to keep them in business for several years to come. □

Flow of superfluid ^{3}He

from P. V. E. McClintock

To push some of the newly discovered superfluid ^{3}He through a fine tube, in order to see how it behaved, might seem a particularly obvious experiment, were it not for the daunting difficulties inherent in working at temperatures near 2 mK. In fact, it is a quite remarkable achievement in terms of experimental design and technique that has enabled R. M. Mueller, E. B. Flint and E. D. Adams to report (*Phys. Rev. Lett.*, **36**, 1460; 1976) the first studies of equilibrium flow phenomena, only four years after observation of the superfluid phases was first suspected. Their experiments were carried out in the Physics Department at the University of Florida.

Because of extreme difficulties of making thermal contact in this ultra-low temperature range, it is usually necessary to design the cryogenics and the experimental cell as an integral unit. Accordingly, the Florida group have devised an ingenious arrangement whereby a pair of Pomeranchuk cells was used both for cooling the ^{3}He into the superfluid A-phase and also for inducing a flow of liquid through the narrow tube which connected them together.

The Pomeranchuk cooling technique depends on the fact that, at very low temperatures, ^{3}He gets colder if it is solidified by compression. Thus, by applying a mechanical force to reduce the volume of a sample of ^{3}He enclosed in a flexible chamber, thereby solidifying some of it, the remaining liquid may be cooled through the superfluid transition at 2.6 mK. The mechanical force is usually generated by way of a separate hydraulic system in which the working fluid is liquid ^{4}He.

The two Pomeranchuk cells of the Florida apparatus were fitted with completely separate hydraulic systems, so that their volumes could be varied independently of each other. By compressing both cells simultaneously the ^{3}He could be cooled without inducing any movement of liquid in the connecting tube. On the other hand, if one cell was compressed while the other was being expanded, then liquid could be made to flow through the tube at constant temperature. A nuclear magnetic resonance (NMR) technique was used for monitoring events in the flow tube.

In view of a calculation by Fetter (*Phys. Lett.*, **54A**, 63; 1975), it had been anticipated that the flow would result in a shift downwards of the NMR resonant frequency. It was surprising, therefore, that rather than changing its frequency, the resonance line in practice developed satellites, and then abruptly disappeared, as the flow velocity was increased. Although the behaviour was not always reproducible in detail, it was found that the satellite structure never appeared when the external magnetic field was applied at 90° to the flow direction. This may perhaps be related to Fetter's prediction that no frequency shift should be seen in this configuration. A further complication was that, before its final rapid fall, the resonance signal sometimes diminished in a series of steps as the flow velocity was increased.

The most definite and interesting observation, however, was the remarkably small velocity at which the signal disappeared. The exact value depended on temperature, but fell within the range 0.003–0.02 mm s^{-1}. These velocities are several orders of magnitude smaller than the Landau critical velocity at which the superfluidity would be expected to disappear as a result of pair-breaking, so that some other mechanism must presumably be involved. It is interesting that investigations of the heat conduction by R. T. Johnson, R. L. Kleinberg, R. A. Webb and J. C. Wheatley (*J. Low Temp. Phys.*, **18**, 501; 1975) also yielded multiple critical velocities for the superfluid which were remarkably small (0.2–0.8 mm s^{-1}), although considerably larger than the velocities reported here. The authors demonstrate, however, that the two sets of measurements may in fact be in quantitative agreement if it is assumed that the flow did not occur through a transfer of bulk liquid: if only the superfluid component of the liquid had moved, leaving the normal fluid component clamped in the tube by its own viscosity, it would have had to move relatively briskly to simulate a given rate of apparent bulk flow because it constitutes only a small proportion of the liquid at the temperatures in question. On this basis they deduce superflow critical velocities in the range 0.14–0.64 mm s^{-1}, in good agreement with those derived from the thermal conduction experiments.

Why should these critical velocities be so small? One is immediately reminded of the parallel situation in superfluid ^{4}He, where the actual critical velocities for superflow are usually about 1 mm s^{-1}, compared with the much larger Landau critical velocity for roton creation of around 50 m s^{-1}. In the case of ^{4}He, the discrepancy has been shown to arise from the relative ease with which quantised vortex lines may be generated in the liquid for flow velocities far below the Landau velocity. It seems very likely that something of a similar nature is happening in ^{3}He. The situation is, of course, infinitely more complicated because the liquid is governed by a vector order parameter, is anisotropic, and takes up complicated textures depending on the shape of the container, applied electric and magnetic fields, and so on. Furthermore, it is not yet by any means clear what would be the analogue of a vortex, in the case of ^{3}He: there may in fact be several different sorts of collective excitation of this general nature.

Assuming, however, that the low critical velocities observed in these experiments arise from the onset of

dissipative processes, it seems probable that at least one completely new type of collective excitation is being created, thus promising to add yet another whole dimension of complexity to our understanding of this extraordinarily complicated liquid. □

Two approaches to gene synthesis

from Maria Szekely

THE recent discovery that synthetic genes can be used to produce genetic recombinants (see *News and Views*, **260**, 189; 1976) opens wide possibilities in the field of genetic engineering. Well-characterised DNA sequences can be inserted into plasmid DNA thus providing specific probes with known sequences for studying the organisation and expression of eukaryotic genomes. Amplification of the inserted gene yields large amounts of specific sequences which can be used for the production or isolation of highly labelled mRNAs, for sequence studies, and so on. At the same time, the safety risk is lessened, as no unknown, harmful genetic information can be integrated into the recombinant.

So far only one eukaryotic gene, that of β-globin, has been inserted into bacterial plasmids. Almost simultaneously, four laboratories reported the production of recombinants from β-globin DNA, using different techniques but all of them starting with reverse transcription of globin mRNA into cDNA. (Rougeon *et al.*, *Nucleic Acids Res.*, **2**, 2365, 1975; Efstratiadis *et al.*, *Cell*, **7**, 279, 1976; Rabbits, *Nature*, **260**, 221, 1976; Higuchi *et al.*, *Abstract ICN-UCLA Winter Conference*, 1976). The advantages of this technique are obvious, there are, however, also some limitations to the method. It follows from the principle of the technique that the DNA inserted into the plasmid can contain only the sequences present in the mRNA: any additional, non-transcribed sequences that may be present in the original gene will be lost. It remains to be seen how their absence affects the function of the integrated gene. In practice, the limitations go even a step further: it depends on the efficiency of the reverse transcriptase how much of the genetic information present in the mRNA will be passed on to the synthetic gene. In many laboratories only fragments of cDNA copies could be obtained with this enzyme. Rougeon *et al.* used a 300–400 nucleotide long cDNA preparation to synthesise the double-stranded globin DNA and after amplification ended up with a 100 to 150 base-pair long integrated globin DNA, about one fifth of the length of the mRNA.

Maniatis' group reported last year the synthesis of a full-length copy of globin mRNA (Efstratiadis *et al.*, *Cell*, **4**, 367; 1975). A few months ago the same group described the sythesis of double-stranded globin DNA with the aid of DNA polymerase I, making use of the hairpin loop structure at the 3′-end of this cDNA, which served as primer in the synthesis of the anti-cDNA strand. (Efstratiadis *et al.*, *Cell*, **7**, 279; 1976). They succeeded in inserting this synthetic gene into PMB 9 plasmid DNA and in their recent paper (Maniatis, Kee, Efstratiadis and Kafatos, *Cell*, **8**, 163; 1976) they describe in detail the techniques of integration of the DNA, transformation of *Escherichia coli* cells and identification of the recombinant.

The synthetic gene is 580 base pairs long, about 80 nucleotides shorter than β-globin mRNA. It contains the full coding region plus 40 and 110 nucleotides of the untranslated regions at the 5′ and 3′ termini, respectively. Molecular hybrids were constructed by annealing the globin DNA which carried poly(dT) tails to *Eco*RI-treated plasmid DNA to which poly(dA) tails have been attached. Transformation of *E. coli* HB101 with this hybrid DNA yielded 600 tetracycline-resistant colonies, a high proportion of which contained sequences of the β-globin gene, as was shown in hybridisation assays. The size of the inserted DNA did not change upon construction of the molecular hybrid, transformation and cloning.

The method seems suitable for general application in the synthesis and amplification of eukaryotic genes. Maniatis' group has already synthesised a number of other genes by copying eukaryotic mRNAs.

It is essential for the usefulness of such techniques that the original structure of the gene should be retained in the recombinant. Fidelity in copying the mRNA and stability of the DNA structure during integration and transformation is therefore of vital importance. Maniatis *et al.* characterised the synthetic gene and the inserted globin DNA after amplification by comparing restriction maps of the DNAs and correlating them to the structure of globin mRNA. Using eight restriction enzymes with known recognition sequences, the restriction map of synthetic DNA was established and compared with the cleavage pattern predicted from the nucleotide sequence of β-globin mRNA. As only part of the nucleotide sequence has been determined so far, some cleavage sites were established by deducing the nucleotide sequence from the known amino acid sequence of β-globin. The restriction map of synthetic globin DNA was in perfect agreement with that expected from the mRNA structure.

The same restriction enzymes were used individually and in combination on the recombinant DNA. By using *Hha* endonuclease, an enzyme which does not cleave synthetic globin DNA but produces many fragments from PMB 9 DNA, one fragment, carrying the inserted globin gene could be isolated and characterised. The restriction map of this fragment was established, and the orientation of the inserted DNA and the distance between the restriction sites determined. These proved to be the same as in the synthetic gene before integration, confirming that the entire synthetic globin DNA molecule was amplified without sequence rearrangements. Some slight changes were detected in the poly(dA)–poly(dT) tails only.

The approach used by Maniatis' group yields very useful DNA probes which will greatly enhance progress in our understanding of the structure and function of the eukaryotic genome. The success of a different, more chemical approach has been reported recently from Khorana's laboratory: the complete gene of suppressor tyrosine tRNA of *E. coli* has been synthesised without using an RNA template. The techniques were worked out years ago by Khorana's group. Following a known nucleotide sequence, chemical synthesis is used to produce blocks of nucleotides and the ligase reaction to join them. The synthetic gene contains the total sequence of the precursor tRNA and includes the sequence of the promoter site and of the stop signal for transcription. Twenty-four scientists contributed to its synthesis which has taken 9 years. This gene has now been introduced into an *E. coli* amber mutant by way of a transducing phage (the details of the technique have not been published so far) and it was found to function correctly as a suppressor gene in the bacterial cell.

Khorana's method requires more time and work than the copying of an RNA template and its use is limited to the synthesis of genes the sequence of which has been fully established. Still, this approach has some great advantages over the reverse transcription of RNAs. The gene synthesised by Khorana contains also sequences which are not present in the final gene product, sequences lost during processing of the RNA and non-transcribed control signals. Such sequences cannot be incorporated into a reverse transcript. The purely synthetic method also makes it possible to introduce specific alterations into the gene structure and to study the effect of such "mutations". It would be interesting to