tion lines present in the spectrum which are due to gas ejection continuing after light maximum. For some novae there are great changes in this outflow which are manifested in dramatic changes in the light curve and spectrum.

Nova Cygni, being bright and easily accessible to all northern observatories, was expected to throw light on the various rival theories for different aspects of the nova phenomenon. Unfortunately it does not seem to be, in any sense, a typical nova.

From comparison of the strengths of the interstellar lines in the early spectrum of the nova with those in the supergiant 55 Cygni, Tomkin, Woodman and Lambert (*Astron. Astrophys.*, in the press) deduced a distance for the nova of somewhat greater than 1 kpc and an intrinsic visual luminosity at maximum of about -10 mag. A similar brightness was obtained from the rate of decline in brightness shortly after maximum by Lindegren and Lindgren (*Nature*, **258**, 501; 1975). This intrinsic luminosity is the highest known for a galactic nova, and the speed of decline is the fastest.

The spectrum during this rapid decline also changes very rapidly and does not show all the 'normal' absorption systems seen in typical novae spectra. Possibly this indicates that for Nova Cygni an instantaneous ejection model is more applicable than the continued ejection models which seem better suited to most novae.

While the decline from maximum light has, compared with many novae, been remarkably smooth, the confirmation (Marcocci, Messi, Natali and Rossi, page 186 of this issue of *Nature*) of a small periodic variation in brightness with a period of some 3 h suggests the possibility that some form of modulation of the nova's brightness is being caused by the orbital motion of the binary.

The observation of coronal lines in the near infrared by Grasdalen and Joyce (see page 187) indicates the presence of a very high temperature ($\sim 10^6$ K) region where apparently the degree of ionisation is still increasing. They suggest that there is the possibility of determining the physical conditions and abundances in this region which is presumably situated close to the site of the nova explosion. Abundance determination should also be possible for a series of high dispersion spectra obtained at the Royal Greenwich Observatory by Stickland within 8 h of discovery. A preliminary analysis by the author indicates that the strengths of the CII, OII and especially NII lines are greater than would be expected suggesting that these elements have greater than normal abundance. This would agree with the theoretical predictions of Starrfield, *et al.* (*Astrophys. J.*, **176**, 169; 1972) who indicated that these elements will be enhanced in this way by the nova mechanism discussed above.

Shortly after maximum light the spectrum became dominated by emission lines which became progressively stronger relative to the continuum as the nebular stage was approached. At the same time structure at the top of each emission line became apparent. Thus structure, initially seen as four peaks at each emission line, has subsequently developed so that by the end of September up to eleven peaks were seen and measured in each of several different spectral lines. The derived velocities were constant for each peak, where present, for all the spectral lines measured. The expanding nebula is thus strongly lobed in a very complex manner which together with observations of circular polarisation in Hα and Hβ by Kemp and Rudy (*IAU Circ.* 2837) perhaps supports Mustel's ideas that strong magnetic fields are present in novae.

There is, no doubt, an enormous amount of observational data on Nova Cygni accumulating. The first results are only now starting to appear, but Nova Cygni seems so far to have posed more problems than it has solved. □

Expression of eukaryotic genes in prokaryotes

from Giorgio Bernardi

MANY of the hopes, and some of the fears, associated with future developments in the field of genetic engineering rely upon the possibility of transcribing and translating eukaryotic genes in prokaryotic systems. It is not yet clear, however, whether the ambitious goal of converting bacteria into factories for the synthesis of medically, agriculturally and industrially important proteins of eukaryotic origin can be achieved without some additional advances in our technology.

Following the pioneering work of Berg and his colleagues at Stanford, restriction endonuclease-generated DNA fragments from various sources have been cloned in *E. coli* by linking them to a plasmid (or a phage) replicon, and then introducing the composite molecules into bacterial cells by transformation. After having been first used to introduce segments of other bacterial plasmids and phage DNA into bacteria (see for example Chang and Cohen, *Proc natn. Acad. Sci. U.S.A.*, **71**, 1030; 1974; Hershfield *et al.*, *Proc. natn. Acad. Sci. U.S.A.*, **71**, 3455; 1974; Tanaka and Weisblum, *J. Bact.*, **121**, 354; 1975), this method has been used to clone fragments of eukaryotic DNA, such as *Xenopus laevis* amplified rDNA (Morrow *et al.*, *Proc. natn. Acad. Sci. U.S.A.*, **71**, 1743; 1974), *Drosophila melanogaster* DNA (Wensink *et al.*, *Cell*, **3**, 315; 1974; Thomas *et al.*, *Proc. natn. Acad. Sci. U.S.A.*, **71**, 4579; 1974; Glover *et al.*, *Cell*, **5**, 149; 1975; Tanaka *et al.*, *Biochemistry*, **14**, 2064; 1975) and sea urchin histone genes (Kedes *et al.*, *Nature*, **225**, 535; 1975; Birnstiel *et al.*, *Xth FEBS Meeting*, **38**, 3; 1975).

In order to study the expression of eukaryotic genes in the prokaryotic host, advantage has been taken of the fact that plasmids segregating into minicells (which are devoid of bacterial chromosomal DNA) are capable of plasmid-specific synthesis of functional gene products (Frazer and Curtiss, *J. Bact.*, **115**, 615; 1973; Van Embden and Cohen, *J. Bact.*, **116**, 699; 1973). For both rDNA and histone genes, it has been shown (Morrow *et al.*, *op. cit.*; Kedes *et al.*, *op cit.*) that transcription takes place in minicells; the fidelity of transcription has not, however, been demonstrated in either case.

A novel approach to this problem has been tried by Chang, Lansman, Clayton and Cohen (*Cell*, **6**, 231; 1975) by looking at the expression of mouse mitochondrial DNA in minicells. This genome has a size of only 10×10^6 and can be cloned in its totality, thus providing an opportunity for studying transcription and translation of a complete eukaryotic genome in a prokaryotic system, in contrast to previous studies which only involved genome fragments.

Covalently closed, circular mtDNA was partially digested with *Eco*RI restriction endonuclease, (in order to obtain linear molecules broken at only one of the two *Eco*RI sites on mouse mtDNA), and annealed with *Eco*RI-digested pSC101 plasmid DNA; after ligation, the DNA mixture was used to transform *E. coli* cells which were grown in the presence of tetracycline, to allow selection of cells transformed by the antibiotic resistance-carrying plasmid. The total closed, circular DNA population was then isolated from transformed cells and fractionated by sucrose gradient certifugation. The 44S DNA molecules, corresponding to a molecular weight of 16×10^6, and therefore to the sum of mtDNA plus plasmid DNA, were amplified by a second cycle of transformation. Four different chimaeric molecules were recognised by heteroduplex analysis and by degradation with *Hind*III, a restriction enzyme having three sites on mtDNA and one (practically coinci-

dent with the *Eco*RI site) on pSC101. These molecules differed in the mtDNA *Eco*RI site used for plasmid insertion and in the relative orientation of plasmid and mtDNA.

MtDNA was then isolated from *Eco*RI digests of chimaeric plasmids by centrifugation in CsCl-netropsin density gradients, and separated into its heavy and light strand by centrifugation in alkaline CsCl. RNA labelled for 30 min with ^{3}H-uridine, obtained from minicells carrying chimaeric plasmids showed a continuous size distribution, as if unstable messenger-like RNA was being isolated in different stages of synthesis and degradation. In addition, most of the 30 min- and 3 min-labelled RNA hybridised to the L-strand, whereas little, if any, hybridised to the H-strand. Since the preferential transcription of the L-strand is not influenced by the relative orientation of pSC101 and mtDNA in the chimaeric molecules, it is improbable that transcription of the L-strand depended on a pSC101 promotor and read through at the mtDNA–pSC101 junction. Thus, one or more signals located within the L-strand are, indeed, recognised by the *E. coli* RNA polymerase. This does not, however, establish any structural identity between these sites and the promotor regions used inside functional mitochondria. In fact, as stressed by the authors, the opposite view is favoured by two lines of evidence. *In vivo,* mitochondrial rRNAs and most tRNAs and mRNAs are transcribed from the H-strand, only three tRNAs and one mRNA originating from the L-strand (Attardi *et al.*, in *The Biogenesis of Mitochondria,* 9, edit. by Kroon and Saccone, Academic Press, 1974). *In vitro,* mitochondrial RNA polymerase preferentially transcribes the H-strand (Wu and Dawid, *J. biol. Chem.*, **249**, 4412; 1974), whereas *E. coli* RNA polymerase preferentially transcribes the L-strand (Tabak and Borst, *Biochim. biophys. Acta* **217**, 356; 1970; Dawid, *Devl Biol.*, **29**, 139; 1972).

The total incorporation of ^{3}H-leucine into protein in minicells carrying the four types of chimaeric plasmids was found to be 2–4 times higher than that obtained from minicells containing pSC101 alone. The label was present in four polypeptides ranging in molecular weight from 2,500 to 5,000; four of the five pSC101-determined proteins were also seen. The protein synthetic pattern observed suggests that translation occurred from RNA transcripts of mtDNA; it differed, however, from that observed in mouse mitochondria, in which case seven proteins ranging in molecular weight from >40,000 to <10,000 were detected.

Among the additional findings of Chang *et al.*, one is of particular interest, namely that the mtDNA origin of replication is not used at a significant frequency in chimaeric plasmids. In fact, all the *Eco*RI fragments originating from them and having the electron microscopic appearance of replicative intermediates had the length of pSC101; furthermore, their replication origins mapped in positions known to be the pSC101 replication origin.

To sum up this remarkable work, it has been demonstrated for the first time that transcription of eukaryotic DNA in prokaryotes can actually start at signals located on the eukaryotic DNA itself instead of being the result of read through from the plasmid; such transcripts, which appear to be translated in minicells, do not seem, however, to start at natural promotor sites on mtDNA and, therefore, do not correspond to any of the natural transcripts of the mitochondrial genome. This conclusion, which may, perhaps, disappoint the proponents of the prokaryotic origin of mitochondria, suggests that only a mitochondrial RNA polymerase is able to recognise mitochondrial promotors. In this case, the successful expression of eukaryotic genes in prokaryotes may need selective resection of their control elements, which should be replaced by those of plasmid or phage vectors (Murray *et al., Xth FEBS Meeting,* **38**, 193; 1975). It should be recalled, however, that most of the mitochondrial transcripts are, in fact, derived from the processing of the single RNA copy of the H-strand (Aloni and Attardi, *J. molec. Biol.*, **70**, 363, 1972). Such a situation, implying the presence of a single promoter on the H-strand, makes animal mtDNA an unfavourable model to test its faithful transcription in minicells and leaves the door open, for the time being, to some optimism over the possibility of correct expression of eukaryotic genes in prokaryotes. An encouraging result along this line is the very recent finding by K. Struhl, J. Cameron and R. Davis of Stanford University of complementation of a *E. coli his*B mutation by a yeast DNA fragment inserted into λ DNA. □

B chromosomes in grasshoppers

from John Bishop

B CHROMOSOMES are chromosomes which appear in some but not in other members of a species, and as such are clearly not necessary for the survival of individuals. The designation B indicates a supernumerary chromosome, in contrast to the A chromosomes, the representatives of the normal complement.

B chromosomes occur in many species of animals and plants. They are heterochromatic, do not pair with A chromosomes, and have no simple Mendelian effects upon the phenotype. They have been observed, however, to have very sharp clinal distributions which suggests a greater tolerance of B chromosomes under more favourable environmental conditions. The evolutionary significance of B chromosomes is elegantly explored in a paper by Hewitt in *Chromosomes Today* (4, edit. by Wohrman and Lewis, Wiley, New York, 1972).

Several studies have pointed to homologies between different B chromosomes and autosomes, centric fragments and X chromosomes. This suggests the exciting possibility that B chromosomes might represent one of the routes by which the genomes of eukaryotes change radically in informational content to take advantage of changing environmental opportunities. The B chromosome, being heterochromatic and therefore at least largely inactive, would be freed from selective constraint; but could be called on again (perhaps randomly) when and if it fulfilled a newly required function. Because of this, it was both surprising and disquieting when Gibson and Hewitt (*Nature*, **225**, 67; 1970; *Chromosoma*, **38**, 121; 1972) suggested that the incidence of B chromosomes in the grasshopper *Myrmeleotettix maculatus* was correlated with the occurrence of a DNA satellite which accounted for as much DNA as the B chromosome(s) could be expected to comprise, if not more.

A satellite DNA is one with a different base composition and therefore a distinct buoyant density from the bulk of the DNA (the main-band DNA). Characteristically, a satellite DNA consists of very many similar short sequences (less than 20 nucleotide pairs) arranged tandemly in long continuous stretches (greater than 30,000 nucleotide pairs). On the other hand main-band DNA is largely composed of non-repetitive sequences intimately interspersed with repetitive sequences that have a much lower repetition frequency than satellite DNA.

If B chromosomes contained DNA that had such a degree of disparity with the A chromosome DNA then it would seem unlikely that the two were in any way related. This would very much weigh against the idea that B chromosomes are in the mainstream of the generation of new genetic function. If this were general, it would close, in eukaryotes, an enticing door which has only more recently