higher resistance of Hb-F to dissociation at both low and high *p*H has been well established. The present knowledge of the tertiary and quaternary structure of haemoglobin, and of the differences in the amino-acid sequences of the polypeptide chains of a large number of haemoglobin species, has now been used by Perutz (see page 341 of this issue) to provide an explanation for the large variations in their rates of alkaline denaturation.

In accordance with earlier views that the rate-limiting step is likely to be the formation of monomeric subunits from the dimer, Perutz has looked at sequence differences located at the $\alpha_1\beta_1$ interface for evidence in support of his hypothesis that denaturation by alkali is activated by the ionization of buried, weakly acidic side chains. Comparing Hb-A with bovine haemoglobin, which is almost indefinitely stable at *p*H 12.7, Perutz concludes that the significant structural difference between these two haemoglobin species in the region of the $\alpha_1\beta_1$ contact is the presence of three buried weakly acidic groups—two cysteines and one tyrosine—which will ionize above *p*H 12 in Hb-A, and which are replaced by neutral or more weakly acidic groups in bovine haemoglobin. The ionisation and hydration of these groups perturbs the tertiary structure and therefore shifts the equilibrium towards the unfolded state and favours dissociation across the $\alpha_1\beta_1$ interface. Comparing Hb-A and Hb-F, the non-α or γ chains of the latter species provide one less cysteine and one less tyrosine in the same locations. These are two of the three changes, relative to Hb-A, found in bovine β chains. It is in fact possible to list several haemoglobin species in order of increasing resistance to alkaline denaturation, and to show that this order follows the progressive substitution of β-130 tyrosine, β-112 cysteine and, finally, α-104 cysteine by neutral or more weakly acidic residues.

Thus both qualitatively and semi-quantitatively, Perutz's suggestion that the ionisation of buried weakly acidic side chains determines the relative susceptibilities of different haemoglobin species to alkaline denaturation seems to be well founded. Certainly electrostatic effects involving exposed ionising groups are likely to be involved in the denaturation of monomers, but the ionisation of buried weakly acidic groups, notably cysteines, in the dimer interface region seems to be the crucial factor in promoting monomer formation at high *p*H and initiating the sequence of events leading to denaturation.

From a Correspondent

Herpes not so simplex

THE most recent discovery of Sabin and his colleague Tarro may open up at least two interesting avenues of research, both of which will be pursued at the University of Chicago, but each of which lead in an entirely different direction. Sabin announced early last year the finding, now published (Sabin and Tarro, *Proc. natn. Acad. Sci. U.S.A.*, **70**, 3225; 1973), that the serum of patients with certain types of tumour contains antibody to a substance produced by cells infected with herpes simplex virus (HSV). The discovery, which swells the growing body of evidence for the involvement of herpes viruses in human cancer, (see, for example, *Oncogenesis and Herpesvirus*, edit. by P. M. Biggs, G. de Thé and L. N. Payne; International Agency for Research on Cancer, 1972) is of immediate practical interest because the presence of antibody is associated with metastasis and may thus constitute a diagnostic tool for monitoring the progress of malignant disease. But its more far reaching implications depend on the possiblity that the antigen is specified by a fragment of the HSV genome expressed in the tumour cells. From the practical point of view, this would establish the rationale for developing the anti-viral vaccine which Sabin has envisaged as a prophylactic against human cancer. From the point of view of basic research, the antigen seems at last a palpable tool for probing the mechanism of carcinogenic transformation of human cells.

But although Sabin and Tarro refer to it as a nonvirion antigen of HSV, no definitive evidence yet exists that it is specified by viral DNA. The evidence linking the tumour antigen with HSV is strong but circumstantial: all tumour types which have so far produced positive serum reactions in complement fixation tests occupy sites which would be accessible to HSV in a typical infection of the mouth, by HSV type I, or the genitals, by HSV type II.

Lac repressor and operator sequenced

How does repressor select one out of several million nucleotide sequences and bind to it to prevent the expression of the operon genes? In order to understand repression one must understand this and the stereochemistry underlying it. Two signal advances to this end have been reported by Gilbert and Maxam and by Beyreuther, Adler, Geisler and Klemm in the current issue of the *Proceedings of the National Academy of Sciences* (**70**, 3576 and 3581; 1973). Briefly Gilbert and Maxam have sequenced the operator, a stretch of DNA which binds the repressor of the *lac* operon in *Escherichia coli*; Beyreuther *et al.* have sequenced the repressor itself.

Gilbert and Maxam made use of the fact that the purified repressor will bind to cellulose nitrate those fragments of sonicated DNA which carry the operator. After binding in this manner the washed fragments were then eluted with a synthetic inducer. The repressor was found to bind to these purified fragments with a half life of 15 min so a quick treatment with DNase could be used to digest away all sequences except those protected by the repressor. The result was a double stranded DNA with a T_m of 67° C in SSC and containing twenty-seven base pairs as revealed by a calibrated polyacrylamide gel.

The sequence of this DNA was ascertained by copying one or other strands using RNA polymerase and complementary oligonucleotides of different length of sequence to effect initiation at various points. Fingerprinting of the transcription products yielded the complete sequence.

A feature of some interest in the final result was an ACAATT sequence near the 3′ end of each strand (with a complementary sequence near both 5′ ends), which gave the operator an element of symmetry. The two regions giving this symmetry are separated by nine base pairs and four of these show the same pattern of symmetry; the two regions are about one helix turn apart and thus could be approached from one side by a repressor showing a two-fold symmetry.

The repressor itself turns out to consist of four identical subunits each of 347 residues. No similarities with the sequence of histones or the known part of the β galactosidase were detected. Exactly how the two bind together must await further investigation, but Gilbert and Maxam suggest possibilities like opening up the DNA to see the bases directly or feeling out the edges of the bases in the grooves.

From a Correspondent