TUMOUR VIRUSES

New Assays

from our Cell Biology Correspondent

ASSAYS which allow the experimenter to measure the number of infectious units in a stock of virus particles are a *sine qua non* of quantitative virology. For that good reason alone the reports of Kawai and Hanafusa and of Kelloff, Hatanaka and Gilden in the current issue of *Virology* (**48**, 126 and 266; 1972) are bound to attract widespread attention, for both groups have devised new ways of assaying the infectivity of certain RNA tumour viruses. Kawai and Hanafusa's experiments are particularly interesting because they have hit upon a rapid and comparatively simple plaque assay for at least some strains of avian leukosis virus.

Most strains of avian leukosis virus infect and replicate in chick embryo fibroblasts without either transforming the cells or causing anything more than an occasional and transient cytopathic effect. As a result these viruses have to be assayed by comparatively laborious methods based on viral interference, viral antigenicity or on the fact that the avian leukosis viruses act as helpers for the replication of certain Rous sarcoma viruses, which can be assayed because they transform cells and give rise to foci or transformants.

Until recently much the same could be said of assays for murine leukaemia viruses, but in 1970 Rowe *et al.* devised the now widely used XC cell plaque assay for mouse leukaemia viruses, which is ingeniously based on the interaction between mouse cells infected with mouse leukaemia virus and rat XC cells transformed by the Prague strain of Rous sarcoma virus. The assay which Kawai and Hanafusa have devised for some avian leukosis viruses in some respects resembles Rowe's XC cell assay.

Kawai and Hanafusa find that when lawns of chick embryo cells infected by a temperature sensitive mutant of Rous sarcoma virus (ts-68) are held at the non-permissive temperature (at which they do not have a transformed cell phenotype) and superinfected with avian leukosis viruses of subgroups B and D (RAV 2, RAV 50 and AMV 2), plaques develop within 4 days as the leukosis viruses kill their hosts. Moreover, estimates of virus infectivity obtained in this way are very close to those obtained by conventional interference assay. By contrast the strains of avian leukosis viruses of subgroups A, C and E that have so far been tested do not cause plaques to develop and cannot therefore be assayed at present by this procedure.

The results of several control experiments prove that the plaques which develop after the ts-68 transformed cells have been superinfected with RAV 2 preparations are indeed the result of RAV 2 infection and not the result of infection by some other agent contaminating the RAV 2 stocks. For example, plaque formation can be prevented by neutralizing the RAV 2 stocks before they are used for superinfection with anti-RAV 2 antisera or by overlaying the lawns of ts-68 transformed cells after infection with RAV 2 with sloppy agar containing this antiserum.

Why only certain strains of avian leukosis viruses are able to form plaques in these conditions and how the plaque forming leukosis viruses kill their hosts remain to be elucidated. When answers to these questions are forthcoming it may be possible to arrange conditions such that leukosis viruses of subgroups A, C and E can also be plaque assayed.

The assay which Kelloff *et al.* have devised is apparently of general use for the measurement of infectivity of any C type RNA tumour virus. It is based on calibrating the amount of reverse transcriptase activity in the supernatant media of cultures of cells infected by these viruses with the amount of the progeny virus present and the infectivity of this virus as assayed by the so-called COMUL and COHAL assays. At least with the several viruses that Kelloff *et al.* have used the enzymatic assay gives 9 days after infection an estimate of the number of infectious virus particles in such supernatants comparable to those obtained at 21 days by existing assays. The new assay, if it proves as reliable as these preliminary results suggest, should at least be a valuable time saver.

SELENOLOGY

Luna 20 Samples

from our Soviet Correspondent

PRELIMINARY analysis of the lunar rock recovered by the Luna 20 spacecraft has revealed the presence of more than seventy elements. Comparison with the Mare specimens recovered by Luna 16 from a site some 120 km away from where the Luna 20 specimens were obtained shows that there is less iron oxide but a small increase in the proportion of alkalis in the Luna 20 specimens (*Pravda*, May 4; 1972).

Component	Rock fraction: Basalt Luna 16	Rock fraction: Anorthosite olivine Luna 20	Soil Luna 16	Soil Luna 20
SiO_2	42.95	42.40	41.90	44.40
Al_2O_3	13.88	20.20	15.33	22.90
FeO	20.17	6.40	16.66	7.03
CaO	10.80	18.60	12.53	15.20
MgO	6.05	12.00	8.78	9.70
TiO_2	5.50	0.38	3.36	0.56
Na_2O	0.23	0.40	0.34	0.55
K_2O	0.16	0.52	0.10	0.10

A comparison of the percentages of the chief components of the Luna 20 specimens with the revised figures for Luna 16 (which differ slightly from those quoted in *Nature* (**228**, 492; 1970)) is given in the table.

Codon-anticodon Specificity

THE nucleotide sequences of another pair of tRNAs from *Escherichia coli*—those which translate the two glutamine codons CAA and CAG—are reported in *Nature New Biology* next Wednesday (June 7) by Folk and Yaniv who add their voices to the growing speculation that the replacement of uridine by 2-thiouridine in anticodons may be an evolutionary device for preventing miscoding. Apparently Folk and Yaniv intend ultimately to characterize the nature of a new suppressor mutation of *E. coli*, su_7, which leads to the translation of the UAG amber codon as glutamine. Preparatory to that they decided to sequence the two glutamyl tRNAs in wild type *E. coli*, $tRNA_1^{Gln}$ which is specific for the codon CAA and $tRNA_2^{Gln}$ which is specific for the CAG codon.

As expected, the sequences of these two tRNAs can be drawn as a clover leaf structure and they are very similar indeed, differing only by seven nucleotides, which suggests to Folk and Yaniv that the genes for these two tRNAs may during evolution have diverged from duplicate copies of a single gene.

Of the seven nucleotide differences, six occur in double-stranded stem regions of the molecule; the seventh, however, is of much greater interest, being at the 5′ position of the presumptive anticodon. In $tRNA_2^{Gln}$, which is specific for the codon CAG, the anticodon sequence is CUG as expected from the rules of base-pairing. In the $tRNA_1^{Gln}$ molecule, however, which reads the CAA codon the anticodon sequence NUG.N is almost certainly 2-thiouridine or a derivative in which the oxygen atom at position 2 in the uridine molecule is replaced by a sulphur atom.

The selective advantage of this change becomes apparent when possible "wobble" base pairing is taken into account. Uridine at this position could base pair with A but also by wobble with G or U. But 2-thiouridine is much less likely to form these wobble base pairs and so the chances of the CAA specific glutamine tRNA translating the CAG glutamine codon or the CAU histidine codon are reduced.

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