The Cosmical Constant

I should like to discuss a little further one of the questions raised by R. H. Dicke (*1*) in his very generous review of my book *General Relativity and Cosmology*. The point raised refers to the cosmical constant. Dicke made a comment, which I read with great pleasure, on the book as a whole, namely, that I was engaged in "drawing from the observational data, meager as they are, the vitality needed to convert formal mathematics into theory." I believe this statement also gives the reason why I included the cosmical constant in Einstein's equations. If the cosmical constant is omitted from the equations, the predicted average density of matter in the universe comes out to be some 100 times greater than the observed. We may, with Sandage (*2*), simply notice the problem with the words "the reason for this discrepancy is not understood at present"; or say, with Dicke, that the observations are not yet reliable enough. Neither of these ways of escape is open to me if Dicke's description of my attitude to general relativity is accurate. An effort must be made, using the theory in its entirety, to find out if the discrepancy can be avoided. I have, I believe, shown that a negative cosmical constant does resolve the difficulty on the basis of the data we possess, even if they are somewhat inaccurate.

The objection raised by Dicke to the presence of the cosmical constant in Einstein's equations is that, when they are derived from a variational principle, the integral to be varied consists of two parts arbitrarily added together, of which one produces the cosmical constant term. Let us call these integrals A and B, of which A produces the Einstein equations without the cosmical constant. Now A has itself to be carefully chosen by the mathematician; an arbitrarily selected integral will not do the trick. And the only way of choosing A is to know the required answer beforehand. In other words, Einstein's equations must have already been obtained by some alternative method. Since A is thus artificially constructed, it does not seem to me that the combination $A + B$ is any more, or any less, artificial than was A itself. In any case, cosmology requires the Einstein equations valid in a region containing matter or energy. To deduce them from a variational principle, a third integral, also carefully selected to give the predetermined answer, must be added *ad hoc* to either A or $A + B$ (*3*). I conclude that the deduction of Einstein's equations from a variational principle is an ingenious exercise in formal mathematics which does nothing to increase or decrease the validity of the equations.

The point at which readers may be more seriously misled is Dicke's statement that the presence of the cosmical constant introduces into the theory "a large characteristic constant length." If this were a necessary interpretation, the situation would be odd indeed, especially as the standard length, compared with which the cosmical constant "length" is "large," is not indicated. However, general relativity can be developed in terms of ordinary units of mass, length, and time, cgs units for example. When this is done it becomes obvious that proper-times should be expressed in time-units and not as lengths, and the method also enables the reader to keep the physical dimensions of all variables clearly before him. It then turns out that the cosmical constant is the inverse square of a time-interval (*4*). In the static Einstein universe this time-interval is the radius of this universe divided by the local velocity of light. In the expanding universe models it is the reciprocal of the Hubble constant. In this second class of models the magnitude of the cosmical constant is of the order of 10^{-35} sec^{-2}. Whether this is "large" or "small" depends on the standard of comparison, a standard that must, of course, be expressed in the same units. For example, the inverse square of the time for $(2\pi)^{-1}$ revolutions in the first Bohr orbit is of the order of 10^{-33} sec^{-2}, compared with which the cosmical constant is exceedingly small. The acceleration of gravity due to the sun at the distance of the earth divided by the radius of the earth's orbit is 4×10^{-14} sec^{-2}, which is again much larger than the cosmical constant. The expression of the cosmical constant as the inverse square of a time-interval characteristic of the physical situation envisaged shows why, in the Newtonian approximation, this constant manifests itself as a universal force (*5*). If the cosmical constant is negative, the force is a general binding force which, like gravitation, serves to bind matter together on a cosmical scale. If the constant is positive, the force is one of repulsion, and it tends to accelerate the expansion of the universe.

Until recently I had fondly imagined that my method of introducing the cosmical constant as a "constant of integration" in the establishment of Einstein's equations was original. I am indebted to Windsor L. Sherman (*6*) for drawing my attention to the fact that the method had long ago been used by Einstein (*7*). At that time Einstein was in his creative stage and was undoubtedly converting formal mathematics into theory. It was only later that he turned to other methods by which esthetic considerations were to abolish the cosmical constant, or the deity's alleged unwillingness to play games of chance with humanity was to abolish the quantum theory.

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References

1. R. H. Dicke, *Science* **149**, 1493 (1965).
2. A. Sandage, *Astrophys. J.* **133**, 355 (1961).
3. R. Adler, M. Bazin, M. Schiffer, *Introduction to General Relativity* (McGraw-Hill, New York, 1965), sec. 10.4.
4. G. C. McVittie, *General Relativity and Cosmology* (Univ. of Illinois Press, Urbana, 1965), pp. 151 and 227.
5. ———, *ibid.*, sec. 6.5.
6. W. L. Sherman, *NASA (Natl. Aeron. Space Admin.) Tech. Note D-2601* (1965).
7. A. Einstein, *The Principle of Relativity*, W. Perrett and G. B. Jeffery, Trans. (Dover, New York, 1923), pp. 191–198.

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Reconstruction of Protein and Nucleic Acid Sequences: Alanine Transfer Ribonucleic Acid

Holley *et al.* (*1*) have proposed the complete sequence of Ala-sRNA (*2*) shown in our Table 1 (sequence II). The sequence was reconstructed by an elegant logical process from the most extensive data on fragments of polynucleotides yet published. While reconstructing this sequence from the data with a digital computer we have observed some inconsistencies between the data and the published sequence. These might be considered trivial were it not for the fact that it is possible to construct slightly different sequences which resolve them. Precedent has already been established for the idea that a reconstructed sequence evolves as more extensive and refined data become available (*3–7*). We wish to

Table 1. Alternative sequences of Ala-sRNA. The final A was added to all sequences, as it was known by Holley to be at the end of the molecule. (*1*, note on p. 1462). The correspondence between the 12 nucleotide designations and letters used by the computer program is as follows: A, A; C, C; MeG, D; DiMeG, F; G, G; I, I; MeI, J; T, T; U, U; DiHU, V; U*, W; ψ, X.

I.	GGGCJXGUGAU(CCU)GC(CCCUU)IGCGGTXCGCGUDGCGGGAGAGUAGCCGUGG?AGCFCGGAC(CU)GUCCACCA
II.	GGGCGUGUDGCGCGUAGVCGGVAGCGCFCUCCCUUIGCJXGGGAGAGWCUCCGGTXCGAUUCCGGACUCGUCCACCA
III.	GGGCGUGUDGCGCGUAGVCGGVAGCGCFCUCCUUIGCJXGGGAGAGGWCUCCGGTXCGAUUCCGGACUCGUCCACCA
IV.	GGGCGUGUDGCGCGUAGVCGGVAGCGCFCUCCUUIGCJXGGGGAGAGWCUCCGGTXCGAUUCCGGACUCGUCCACCA
V.	GGGCGUGUDGCGCGUAGVCGCGGVAGCFCUCCUUIGCJXGGGAGAGGWCUCCGGTXCGAUUCCGGACUCGUCCACCA

show that the sequence can also be refined with the aid of a digital computer to carry out the logical steps in the reconstruction from any given set of data.

Any change in sequence will have implications in terms of the conformation of the polymer if current theories of the role of Watson-Crick base pairing in determining structure are correct. Certain changes may also have implications in terms of the genetic code if they occur at the anticodon or amino-acid-acceptor loci.

Inconsistencies can easily be recognized by comparison of the experimental fragment data with a data matrix (*8*) or fragment table, which is a list of the fragments that should be produced under given experimental conditions from the proposed sequence. In Table 2 is a listing of the fragments which should be produced from sequence II by complete digestion with pancreatic and bacteriophage T1 ribonucleases. Also listed is the group of fragments which should be found upon complete digestion with the T1 enzyme of each of the T1 partial digest fragments (*a* through *k*) reported by Holley *et al.* (*1*) and in our Table 2. Holley *et al.* using certain logical arguments decided upon the sequences *a* through *k* by arranging their complete digest fragments according to certain logical arguments.

Comparison of the entries in Table 2 with the recently published data (*1, 7, 9, 10, 11*) shows the following six inconsistencies: (i) Nine moles of Gp per mole of RNA should be produced by complete digestion of sequence II by T1 ribonuclease; the experimental value is 10.5 (*7*, table 5). (ii) The W-ending (U*) fragment in the complete pancreatic ribonuclease digest should have five G residues; the experimental value is 5.7 (*7*, table 1). (iii) Fragments *j* and *k* should give 5 and 4 mole of Gp, respectively, upon complete digestion with T1 ribonuclease; the experimental data show there are at least as many Gp's in *k* as in *j* [*9*, table 1, which shows that (Gp in *k*)/(Gp in *j*) = 5.7/5.4]. (iv) Thirteen moles of Cp should be produced but only 12.4 were found (*7*, table 1). (v) the C(CCU)CUUI fragment in the complete T1-ribonuclease digest should have four C residues; the experimental value is 3.3 (*7*, table 5). (vi) Six moles of Up should be produced, but only 5.0 were found (*7*, table 1). These inconsistencies represent differences between the experimental values and the integer number of residues in the fragment table which cannot be resolved by normal procedures for rounding off values.

As an illustration of the manner by which inconsistencies can be resolved by modifying a sequence, we

Table 2. An Ala-sRNA fragment table constructed from sequence II (Table 1) and in a form for input into the computer program which reconstructs sequence. Letters B and E denote the beginning and end of the molecule. The number at the left of each fragment indicates the chemical breaking method used to obtain the fragment. For example, 1 denotes pancreatic ribonuclease, which breaks the molecule after the nucleotides C, T, U, V, W, and X, and 2 and 12 denote T1 ribonuclease which breaks after D, F. G, and I. Single digits (as 1 and 2) denote that the fragment resulted from a complete enzyme digestion, whereas two digits (as 12) denote a fragment obtained from partial hydrolysis. The dashes indicate that the fragment containing them is unresolved with respect to the order of the subfragments between dashes. This use of dashes differs from that of Holley (*1*), who used them to denote linkage between nucleotides. The sequence of letters in fragments and subfragments is known unless enclosed in parentheses, as (CCU) in fragment *b*. The computer tries all permutations of subfragments between dashes and of enclosed letters within a fragment in search of a uniquely allowable sequence of nucleotides in the fragment. A plus after a fragment, as in fragment *f*, indicates that the following fragment or fragments (which are always of known sequence) must occur between the first and last letters of any permutation of the original fragment. It is not necessary to indicate the 3′-phosphate terminals and the 3′-5′ phosphodiester linkages of the fragments in the computer input.

Complete digests (ribonuclease)

Pancreatic				Takadíastase			
1 CE	1 C	1 U	1 GGGAGAGW	2 G	2 G	2 CG	2 CJXG
1 C	1 C	1 AC	1 DGC	2 G	2 G	2 AG	2 TXCG
1 C	1 C	1 JX	1 AGC	2 G	2 BG	2 AG	2 ACUCG
1 C	1 C	1 FC	1 AGV	2 G	2 CF	2 UG	2 UCCACCE
1 C	1 X	1 GC	1 GAU	2 G	2 UD	2 UAG	2 WCUCCG
1 C	1 U	1 GC	1 IGC	2 G	2 CG	2 VCG	2 AUUCCG
1 C	1 U	1 GU	1 GGT	2 G	2 CG	2 VAG	2 CUCCCUUI
1 C	1 U	1 GU	1 GGV		2 CG		
1 C	1 U	1 GU	1 GGAC				
1 C	1 U	1 GU	1 BGGGC				

Partial Digests

a 12BG-G-G-G-UG-UD-CG-
b 12-CF-C(CCU)CUUI-
c 12-G-G-AG-AG-CJXG-
d 12-ACUCG-UCCACCE
e 12VCG-VAG-G-CG-CF-C(CCU)CUUI-+ 1 AGC
f 12-G-G-AG-AG-CJXG-WCUCCG-TXCG-G-+ 1 GGT
g 12-AUUCCG-G-ACUCG-UCCACCE
h 12-UAG-VCG-VAG-G-CG-CF-C(CCU)CUUI-+ 1 AGC
i 12-AUUCCG-G-G-AG-AG-CJXG-WCUCCG-TXCG-G-
j 12BG-G-G-G-CG-UG-UD-UAG-VAG-G-CG-CF-CG-CG-C(CCU)CUUI-G-VCG-
k 12-G-G-G-G-AG-AG-CJXG-ACUCG-WCUCCG-TXCG-AUUCCG-UCCACCE

present sequences III and IV (Table 1), which contain one more G residue and one less C residue than sequence II. Both III and IV give fragment tables with (i) ten Gp's in the complete T1 digest, (ii) six G residues in the W-ending fragment in the complete pancreatic-ribonuclease digest, (iii) the same number of Gp's in the complete T1 digest of fragments *j* and *k*, (iv) 12 Cp's in the complete pancreatic-ribonuclease digest, and (v) three C residues in a CUCCUUI fragment. Both would also yield an additional Gp in the complete digests of the fragments *f* and *i*. The question of whether a G should be added to fragments *f* and *i* cannot be answered until the publication of the quantitative analysis of those fragments. In sequence IV, Table 1, the sub-sequence corresponding to fragment *c* would yield three Gp, two AGp, and one CJXGp upon complete T1 digestion. However, Apgar *et al.* (*10*) report that fragment *c* yields Gp, AGp, and CJXGp in molar ratios of 2 : 2 : 1, respectively. Therefore, with the data now available, sequence III has five fewer inconsistencies than II and one less than IV.

The sequence of the W-ending fragment was apparently determined by analysis of the elution profile after partial digestion of the fragment with snake-venom phosphodiesterase (*11*, fig. 2). The chromatographic peaks numbered 5, 6, and 7 contained fragments ending in A, G, and W, respectively, from which the terminal sequence AGW was deduced. The chromatographic data do not exclude the possibility of a terminal sequence AGGW. There is no necessity that the stepwise degradation products appear in comparable amounts, and any subsequence which is particularly labile to the enzyme might not be seen as a separate peak. However, inspection of the published elution profiles does reveal a definite asymmetry between peaks 6 and 7, and this asymmetry suggests the presence of an additional species which could correspond to a GGGAGAGG oligonucleotide.

Although to each sequence there corresponds a particular fragment table, the converse is not necessarily true. Alternative sequences which can be reconstructed from a given fragment table are called "isotomers" (*12*). From a proposed sequence it is a simple matter to construct a fragment table, but the search for all other sequences which would give the same fragment table is much more complex and is facilitated greatly by a computer program (*13*). When the fragment table (not shown) for sequence III is used as computer input, only one "isotomer" of III is found (Table 1, sequence V). Sequences III and V are the only "isotomers" even if the large fragments *h*, *i*, and *k* are not used. These large fragments are not needed, although they provide some confirmation of sequences III and V by adding redundancy. Sequence V differs from III in having the sub-sequence VCGCGGVAGCFCUCCUUI rather than VCGGVAGCGCFCUCCUUI. These subsequences correspond to fragment *e*, and (with VCG removed) would give two and one trinucleotides, respectively, upon complete pancreatic-ribonuclease digestion. Since Holley *et al.* (*1*) found one trinucleotide, sequence III has one less inconsistency than V.

As noted above, the existence of six Up's in the fragment table of sequence II is inconsistent with the observed value of 5.0 Up's. The inconsistency remains for III through V. In principle it might be possible to eliminate this inconsistency by a suitable sequence modification. Preliminary results showed that it is not easy to modify the sequence by trial and error, as was the case with the deleted C and added G, without introducing new inconsistencies. Therefore, an alternative approach using the computer was employed. The fragment tables for sequences II and III were modified by removing one Up and used as input for the computer. The computer was not able to find any sequences consistent with 5 Up and the rest of the fragment tables which did not increase the number of inconsistencies. We were, therefore, led to conclude that this particular inconsistency must be due to experimental error, either in chemical analysis of, or incomplete recovery of, the Up fragments (*14*, *15*).

There remain some uncertainties—that is, differences between experimental values and the fragment tables—which can be resolved by normal rounding procedures, for all sequences in Table 1. The most prominent uncertainties arise from analysis of the peaks from chromatography of the complete pancreatic-ribonuclease digest of Ala-sRNA on DEAE-sephadex (*7*, table 1) which reveals 0.4 AUp, 4.5 GUp, and 0.5 JXp mole per mole of RNA. Analysis of the peaks from chromatography of the complete T1 digest of Ala-sRNA (*7*, table 5) showed 3.5 mole of CGp per mole of RNA and peaks 78 to 82 showed 1.4 mole of Cp per mole of VCG fragment.

Until much more is known about the role which the particular sequence of an sRNA plays in its biological functions, there are few a priori grounds on which to decide whether a proposed sequence is valid. In the absence of detailed x-ray diffraction or electron-microscopy evidence as to the sequence, the only criterion for preferring one sequence to another is a higher degree of consistency with the experimental fragment data. By this operational criterion, sequence III is to be preferred to any other in Table 1, on the basis of the published data. Nevertheless, because III itself exhibits an inconsistency with the data and because there are a number of uncertainties in the data, III cannot be considered as a unique representation of the order of the residues in Ala-sRNA but must be viewed as subject to refinement.

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References and Notes

1. R. W. Holley, J. Apgar, G. A. Everett, J. T. Madison, M. Marquisee, S. H. Merrill, J. R. Penswick, A. Zamir, *Science* **147**, 1462 (1965).
2. Abbreviations: A, adenosine; C, cytidine; U, uridine; G, guanosine; DiHU, 5, 6-dihydrouridine; U*, a mixture of U and DiHU-; DiMeG, N^2-dimethyl guanosine; I, inosine; MeG, 1-methyl guanosine; MeI, 1-methyl inosine; ψ, pseudouridine; T, ribothymidine; p, 3′-phosphate, such as Ap; DEAE, diethylaminoethyl. In a sequence of letters, such as AUG, neighbors are linked by 3′5′ phosphodiester bonds.
3. For example, originally Ala-sRNA was thought to have a chain length of 85 nucleotides based on alkaline hydrolysis data (*4*) but subsequently this value was refined to 81 (*5*). Sequence I (Table 1) proposed as "one of thousands" consistent with the experimental data (*6*) contains approximately 72 nucleotides while sequence II contains 77. As a further example, earlier published data (*5*) showed the occurrence of the fragments GGGTp, AAGp, (AC)Gp, and IGGCp, in complete enzyme digests of the Ala-sRNA, which do not appear in the most recently published digest data (*7*). These earlier data (*5*) also call for 12 Gp, 2 UGp, and 3 AGp fragments in a complete T1 digest of the Ala-sRNA rather than 10.5 Gp, 0.9 UGp, and 2.3 AGp found more recently (*7*).
4. R. W. Holley, J. Apgar, S. H. Merrill, P. L. Zubkoff, *J. Amer. Chem. Soc.* **83**, 4861 (1961).
5. R. W. Holley, J. Apgar, G. A. Everett, J. T. Madison, S. H. Merrill, A. Zamir, *Cold Spring Harbor Symp. Quant. Biol.* **28**, 117 (1963).
6. R. W. Holley, G. A. Everett, J. T. Madison, M. Marquisee, A. Zamir, *Abstr. I. Nucleic Acids, Intern. Congr. Biochem.*, 6th Congr., 26 July to 1 Aug. 1964, New York, pp. 9–10.
7. R. W. Holley, G. A. Everett, J. T. Madison,

A. Zamir, *J. Biol. Chem.* **240**, 2122 (1965).
8. J. E. Mosimann, M. B. Shapiro, C. R. Merril, D. F. Bradley, submitted for publication.
9. J. R. Penswick and R. W. Holley, *Proc. Nat. Acad. Sci. U.S.* **53**, 543 (1965).
10. J. Apgar, G. A. Everett, R. W. Holley, *ibid.* **53**, 546 (1965).
11. R. W. Holley, J. T. Madison, A. Zamir, *Biochem. Biophys. Res. Commun.* **17**, 389 (1964).
12. C. R. Merril *et al.*, *ibid.* **19**, 255 (1965).
13. D. F. Bradley, C. R. Merril, M. B. Shapiro, *Biopolymers* **2**, 415 (1964).
14. The single change in G which we have proposed to eliminate three inconsistencies occurs adjacent to the intriguing W(U*) residue which consists of a mixture of uracil and dihydrouracil. It is not clear whether the two sequences, one containing U and the other containing DiHU, are synthesized separately or whether the one is converted to the other (*15*) by a redox reaction before, after, or during the protein-synthesizing step in which the RNA is involved. If the latter were true, then knowledge of the precise sequence about W might be very important for an understanding of the RNA function.
15. P. Roy-Burman, S. Roy-Burman, D. W. Vissar, *Biochem. Biophys. Res. Commun.* **20**, 291 (1965).
16. We thank J. Vinton for help in preparing many of the computer runs which were made as part of this work.

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Shapiro *et al.* (*1*), after an extensive review of our experimental data, suggest changes of two nucleotides in the nucleotide sequence of the alanine RNA (*2*). One change involves deleting a nucleotide from the octanucleotide C-U-C-C-C-U-U-I- to give a heptanucleotide, and the second change involves adding a nucleotide to the octanucleotide G-G-G-A-G-A-G-U*- (the asterisk indicates that the position is occupied by a mixture of uridine and dihydrouridine) to give a nonanucleotide. The basis for the suggestion is that, even though the observed nucleotide composition analyses (*3*) are within experimental error of calculated values for octanucleotides, the agreement would be improved if the fragment were a heptanucleotide and a nonanucleotide.

Until we determined the sequences of the two octanucleotides it was our assumption also that the fragments were a heptanucleotide and a nonanucleotide, respectively. However, during determination of the sequences, it was found that both fragments were octanucleotides. Thus, Shapiro *et al.* have retraced paths that we took ourselves. In our judgment, they have not considered adequately the additional experimental data that exclude their suggestions.

It may be worthwhile to summarize the evidence that was considered conclusive, particularly since it serves to emphasize the power of the method (*4*) that was used for sequence determination of oligonucleotides composed of five to eight nucleotide residues. In the sequence determinations, an oligonucleotide was first treated with alkaline phosphatase to remove the terminal 3′-phosphate. Snake-venom phosphodiesterase was then used to obtain a mixture of stepwise degradation products, formed by the successive removal of nucleotide residues from the 3′-end. Chromatography of the mixture, under conditions that separate the products primarily on the basis of charge, gave a series of peaks. The size of the oligonucleotide was obtained by counting the number of peaks. The nucleotide sequence was obtained from analyses of the successive peaks for their terminal nucleosides. With each of the above octanucleotides, the method gave a set of evenly spaced peaks, and the number of peaks indicated that the starting material was an octanucleotide.

The suggestion that the octanucleotide C-U-C-C-C-U-U-I- is perhaps a heptanucleotide requires that one ignore the fact that the chromatographic pattern shows that there is one more stepwise degradation product than would be obtained from a heptanucleotide.

The suggestion that the octanucleotide G-G-G-A-G-A-G-U*- is a nonanucleotide requires that the chromatogram contain one more peak than was observed, or that a peak corresponding to a stepwise degradation product be missing. In the chromatogram (*4*), the observed peaks are approximately evenly spaced, with no indication of a gap corresponding to a missing peak. The spacings between peaks are 7, 8.5, 7, 7.5, and 8.5 tubes, respectively, starting at the trinucleoside diphosphate and proceeding to the octanucleoside heptaphosphate. The last peak in the series is known to be the octanucleoside heptaphosphate because it contains the terminal pyrimidine. Actually this peak contains two components, because of the mixture of terminal uridine and dihydrouridine. As an added control in the chromatogram, the presence of a terminal 3′-phosphate is shown to displace the chromatographic peak 11.5 tubes to the right (comparison of the position of the octanucleoside heptaphosphate with that of the octanucleotide), which is exactly what would be predicted for the addition of two negative charges without the addition of any nucleoside. The removal of two nucleotide residues without the presence of a detectable intermediate would result in a gap in the pattern with a width of more than 11.5 tubes. The chromatogram shows no such feature. In conclusion, the chromatographic pattern is exactly what would be expected from an octanucleotide and is clearly inconsistent with a nonanucleotide pattern.

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References

1. M. B. Shapiro, C. R. Merril, D. F. Bradley, J. E. Mosimann, *Science* this issue.
2. R. W. Holley, J. Apgar, G. A. Everett, J. T. Madison, M. Marquisee, S. H. Merrill, J. R. Penswick, A. Zamir, *ibid.* **147**, 1462 (1965).
3. R. W. Holley, G. A. Everett, J. T. Madison, A. Zamir, *J. Biol. Chem.* **240**, 2122 (1965).
4. R. W. Holley, J. T. Madison, A. Zamir, *Biochem. Biophys. Research Commun.* **17**, 389 (1965).

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Adenovirus "Helpers"

In their report [*Science* **149**, 754 (1965)] on adenovirus-associated defective virus particles (AAV), Atchison, Casto, and Hammon establish three important facts: (i) AAV multiplies only in the presence of mulitplying simian adenovirus type 15 (SV 15). (ii) Only SV 15 and human adenovirus type 2, of several viruses tested, can serve as "helper" for AAV. (iii) AAV and SV 15 are serologically unrelated. The authors then draw an analogy between AAV and Rous sarcoma virus, another defective virus. The Rous sarcoma defect involves the inability of the virus to manufacture its own coat protein; in the presence of helper, Rous sarcoma is produced by utilizing coat protein provided by the helper. Consequently Rous sarcoma and its helper virus are serologically identical. A better analogy can be drawn between AAV and a defective plant virus, the so-called satellite of tobacco necrosis virus (TNV). The satellite cannot multiply alone, is helped only by strains of TNV, and is serologically unrelated to TNV. Concerning both AAV and the TNV satellite, the major question that remains unanswered is: what restricts the choice of helper virus?

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