Physicochemical Properties and Restriction Maps of Simian Adenovirus Type 38 DNA

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SUMMARY

The sedimentation constant of simian virus type 38 (SV-38) DNA was estimated to be 31.6S. The intrinsic viscosity of DNA was on average 86.5 dl/g and the length of the molecule determined by electron microscopy was 10.6 μm. The average mol. wt., as determined by sedimentation and viscometry, was 21.5 × 10^6, which agreed well with the value derived from the length of the molecule (21.4 × 10^6) and with the value of 21.2 × 10^6 determined by the relative electrophoretic mobility of the DNA fragments produced by restriction endonucleases EcoRI, SalI and BglII.

The buoyant density of the DNA in caesium chloride and caesium sulphate was 1.7185 and 1.4295 g/ml respectively. The melting temperature of the DNA in 1 × SSC was 93.5 °C. The GC content calculated from ρ and Tm values was 59.3%.

BglII cleaves SV-38 DNA at three sites producing four fragments with mol. wt.: A, 9.3 × 10^6; B, 5.6 × 10^6; C, 3.3 × 10^6; and D, 2.9 × 10^6. After treatment with EcoRI and SalI, SV-38 DNA is cleaved into five and six fragments respectively, with mol. wt. for EcoRI fragments: A, 8.2 × 10^6; B, 6.5 × 10^6; C, 4.0 × 10^6; D, 1.27 × 10^6; and E, 1.07 × 10^6, and for SalI fragments: A, 6.5 × 10^6; B, 5.4 × 10^6; C, 4.2 × 10^6; D, 2.8 × 10^6; E, 2.5 × 10^6 and F, 0.25 × 10^6. The sequence of fragments within the SV-38 DNA molecules for BglII was deduced to be BDCA, and for EcoRI was BCEAD.

INTRODUCTION

Until now the only simian adenovirus investigated with regard to its physicochemical properties and genome fragmentation with restriction endonucleases was adenovirus type 7 (SA-7; Burnett et al. 1975; Naroditsky et al. 1978). Concurrently with studies on this, the most oncogenic simian adenovirus, we chose to investigate another, also oncogenic, SV-38. This allows comparative studies within the group of simian viruses with respect to physicochemical properties of their DNA as well as the fine structure of their genome. The specific fragmentation of simian adenovirus DNA and the restriction endonuclease maps proved to be valuable tools for studying the structural, functional and comparative features of the adenovirus genomes. The restriction endonuclease maps also provide the first necessary step in constructing transcription maps and analysis of virus gene expression.

METHODS

Cells. Primary green monkey kidney cells were grown in monolayer cultures containing 199 medium supplemented with 45% lactalbumin hydrolysate and 10% calf serum.
Virus. SV-38 was obtained from Dr A. D. Altstein and new seed stocks were prepared by propagation in monolayer cultures. In vivo $^{32}$P-labelled SV-38 was prepared by the addition of H$_3^{32}$PO$_4$ 1 h after infection (p.i.) to a phosphate-free medium supplemented with 2% embryonic calf serum to a final specific activity of 80 to 100 $\mu$Ci H$_3^{32}$PO$_4$ per 1 ml medium. Virus was harvested 72 h p.i. SV-38 was purified by a modified method of Green & Piña (1963) including two sedimentation equilibrium bandings in CsCl density gradient by the method of Maizel et al. (1968). The purity of the virus preparation was controlled by electron-microscopy. Sometimes adenovirus particles with a density of 1.34 g/ml were detected in the purified virus preparations. This adeno-associated virus could be tentatively identified as belonging to serotype 4 which is present as a contaminant in the primary green monkey kidney cells (R. S. Dreizin, unpublished observation). If DNA is released from such contaminated preparations and then fractionated by electrophoresis in agarose gels two DNA bands are detected which could be eluted separately from the gels. SV-38 DNA was further used for cleavage with restriction endonucleases (Fig. 1).

DNA. SV-38 DNA was released from viruses by the method of Bello & Ginsberg (1969). In order to obtain a DNA--protein complex, DNA was released from SV-38 with guanidinium chloride as described previously by Naroditsky et al. (1978).
Nick translation for labelling DNA in vitro was performed by the method of Maniatis et al. (1975) with certain modifications, using 5-32P-deoxynucleoside triphosphates (Radiochemical Centre, Amersham, Bucks, U.K.) and DNA polymerase (Miles, Elkhart, Ind., U.S.A.). Nick translation produced DNA preparations with a specific activity of $3 \times 10^6$ ct/min/1 μg DNA as compared with $3 \times 10^5$ ct/min/1 μg DNA obtained by in vivo labelling.

Enzymes. Endonuclease EcoR1 was prepared from E. coli strain RY-13 by a modified method of Yoshimori (1971). BglII was prepared by a modified method of R. J. Roberts (unpublished observations). SalI was prepared by the method of B. S. Naroditsky (unpublished observations). DNA polymerase, prepared from Micrococcus lysodeikticus was purchased from Miles. Pronase was purchased from Calbiochem (San Diego, Calif., U.S.A.) and dissolved in 0.01 M-tris, pH 7.9, 0.01 M-EDTA, pH 7.9, to make 20 mg/ml stock solution which was autodigested for 3 h at 37 °C.

Digestion with restriction endonucleases. Reaction conditions for endonuclease digestion were as follows: digestion reactions with EcoR1 contained 70 mM-tris-HCl, pH 7.5, 50 mM-NaCl, 10 mM-MgCl₂, and 7 mM-β-mercaptoethanol; with SalI – 8 mM-tris, pH 7.6, 150 mM-NaCl, 6 mM-MgCl₂, 0.2 mM-EDTA and 50 μg/ml bovine serum albumin; with BglII – 20 mM-tris-HCl, pH 7-5, 7 mM-MgCl₂ and 7 mM-β-mercaptoethanol. For double cleavage with BglII and EcoR1 the first enzyme was incubated with the DNA for 3 h and then NaCl was added to a final concentration of 50 mM and the DNA was further digested with EcoR1. Digestion was carried out for 3 h at 37 °C; the volume of the reaction mixture varied from 20 to 50 μl. The DNA content varied from 0.5 to 2.0 μg and the volume of the added endonuclease varied from 0.5 to 5 μl. In order to obtain partial digestion, SV-38 DNA was digested with diluted enzymes and the time of incubation was decreased. The reaction was stopped by addition of EDTA or by heating for 5 min at 65 °C. Digestion of the DNA protein complex carried out after previous splitting with a restriction endonuclease was accomplished by the addition of 10 μg Pronase for 10 min at 37 °C to the pre-existing reaction mixture.

Gel electrophoresis. Agarose gels (0.8 to 1.4%) were prepared and run as described by Sharp et al. (1973). Tube gels containing 32P-labelled DNA were sliced into 1 mm sections and Cerenkov radiation was counted in an Intertechnique scintillation counter. In the case of slab gels containing 32P-DNA the gels were dried and bands of 32P-labelled DNA were located by autoradiography using Kodak NS-5T or ORWO X-ray films. All gels containing non-labelled material were stained by immersion in electrophoresis buffer containing 0.5 μg/ml ethidium bromide and fluorescence of the bound dye in u.v. light was photographed through a red filter on Svema 250 or ORWO plates. To estimate the mol. wt. of the obtained restriction fragments we used as an inner standard the values of the mol. wt. of the EcoR1 λ DNA fragments estimated by Thomas & Davis (1975).

Recovery of DNA and DNA fragments from agarose gels was performed by the method of Mulder et al. (1975) with certain modifications.

Electron microscopy. Electron microscopy of DNA and virus particles was performed as previously described (Naroditsky et al. 1978).

Ultracentrifugation. Ultracentrifugation was carried out in the An-D rotor of the Spinco model E Ultracentrifuge in standard 12 mm aluminium cells at 16000 rev/min at 20 °C. DNA was dissolved in 1 x SSC (0.15 M-NaCl, 0.015 M-sodium citrate) to a final concentration ranging from 10 to 25 μg/ml. The sedimentation velocity patterns were photographed in u.v. light on Kodak plates. The plates were analysed with a Chromoscan 200 densitometer. The sedimentation coefficient ($s_{20,w}^o$) was calculated by graphical extrapolation of the value $10^2/s_{20,w}^o$ towards infinite dilution.

Viscometry was performed in a low-gradient rotational Zimm viscometer (Zak &
Fig. 2. Extrapolation of $10^3/s_{20,w}$ towards infinite dilution of SV-38 DNA.

Fig. 3. Thermal denaturation profile of SV-38 DNA in $1 \times$ SSC. $A_A/A_{ss}$ is the ratio of the absorption at the listed temperature to the absorption at $25^\circ$C.

Tikchonenko, 1966) at $20 \pm 0.1^\circ$C with a DNA concentration of $2.0 \mu g/ml$ and tension ($\tau$) of $1.2 \times 10^{-3} \text{ dl/cm}^2$.

**Thermal denaturation.** DNA was denatured in a thermostated cell of the Unicam SP 800 spectrophotometer. Temperature rise and absorption at 260 nm were measured using a double channelled self-writing mechanism, Varian A-25.

**Density gradient ultracentrifugation.** CsCl and Cs$_2$SO$_4$ density gradient centrifugations were done in the Spinco Model E ultracentrifuge, as previously described (Dubitchev et al. 1978). We used *Micrococcus lysodeikticus* DNA (CsCl density = $1.731 \text{ g/ml}$ and Cs$_2$SO$_4$ = $1.435 \text{ g/ml}$) and *Clostridium perfringens* DNA (CsCl = $1.6911 \text{ g/ml}$ and Cs$_2$SO$_4$ = $1.4212 \text{ g/ml}$) as markers (Szybalski, 1968).

**RESULTS**

**Physicochemical properties of DNA**

The value of the sedimentation coefficient was calculated to be $31.6 \pm 0.4 \ s_{20,w}^0$ (Fig. 2). The value of intrinsic viscosity ($\eta$) was estimated to be $86.5 \pm 5 \text{ dl/g}$. Using different empirical equations between the $s_{20,w}^0$ and the $\eta$ values and the semi-empirical equation of Mandelkern–Flory or its variants, as in our previous work (Dubitchev et al. 1978), we estimated values for the mol. wt. of SV-38 DNA of $18$ to $24 \times 10^6$. By this means, the average value of the mol. wt. of SV-38 DNA was estimated to be $21.5 \times 10^6$. From a histogram of the length of SV-38 DNA determined by electron microscopy (Dubitchev et al. 1978) the average length of the above DNA was estimated to be $10.4 \pm 0.4 \mu m$.

The pattern of thermal denaturation of SV-38 DNA is shown in Fig. 3. The characteristics of the melting profile are typical of a double stranded DNA. The melting temperature
Restriction maps of simian adenovirus

Fig. 4. (a) Distribution of SV-38 DNA in CsCl density gradients. 1, Marker DNA of Micrococcus lysodeikticus, 1.721 g/ml, 71% GC; 2, SV-38 DNA; 3, marker DNA of Clostridium perfringens, 1.6911 g/ml, 31% GC; 4, SV-38 DNA. (b) Distribution of SV-38 DNA in Cs2SO4 density gradients. 1, Marker DNA of Micrococcus lysodeikticus, 1.435 g/ml, 71% GC; 2, SV-38 DNA; 3, marker DNA of Clostridium perfringens, 1.4212 g/ml, 31% GC; 4, SV-38 DNA.

of SV-38 DNA was calculated to be 93.5 ± 0.1 °C, which corresponds to a GC content of 58.9% (De Ley, 1970).

The distribution of SV-38 DNA and the marker DNAs in a CsCl and Cs2SO4 gradient is illustrated in Fig. 4(a and b). The buoyant density of SV-38 DNA in CsCl was calculated to be 1.7185 g/ml and in Cs2SO4 1.4295 g/ml, which, according to Szybalski (1968) and De Ley (1970), corresponds to 59.1 and 60.0% GC respectively. The value for the base composition estimated by the melting temperature and by the buoyant density agree well within the limits of error characteristic for the methods described.

Fragmentation of SV-38 DNA by specific endonucleases EcoR1, BglII and Sall

The restriction endonuclease EcoR1 cleaves SV-38 DNA at four sites and on agarose gel electrophoresis, five separate bands are detected by autoradiography or by the conventional ethidium bromide technique (Fig. 5). The DNA fragments are denoted by their electrophoretic mobility.

Cleavage of 32P-labelled and non-labelled SV-38 DNA with BglII led to the appearance of four separate fragments when fractionated by electrophoresis in agarose gels (Fig. 5). By cleavage of 32P-labelled SV-38 DNA with Sall and subsequent fractionation by electrophoresis six fragments were detected on radioautograms (Fig. 5). When non-labelled material was used fragment F was faint or absent.

In order to estimate the mol. wt. of the restriction fragments we carried out co-electrophoresis of EcoR1 fragments of bacteriophage λ DNA with SV-38 DNA fragments produced by all the above-mentioned enzymes. The apparent size of each fragment was calculated from a calibration curve and the mol. wt. is listed in Table 1. We have assumed that the relative electrophoretic mobility of the fragments reflects the relative mol. wt.
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Fig. 5. Agarose-gel electrophoresis of SV-38 DNA cleaved with restriction endonucleases EcoRI, BglII and Sall. DNA was fractionated by 0.8% agarose slab gel electrophoresis, 1.5 V/cm, 18 to 22 h, at 4°C. A, B, C, D, E, F – restriction fragments; I, II, III and IV – products of partial hydrolysis. (a) EcoRI fragments of SV-38 DNA; (b) EcoRI fragments and products of partial hydrolysis of SV-38 DNA; (c) Sall fragments of SV-38 DNA; (d) Sall fragments of 32P-labelled SV-38 DNA; (e) BglII fragments of SV-38 DNA; (f) BglII fragments and products of partial hydrolysis. The slab gels were stained by the conventional ethidium-bromide technique and photographed; the photographs were cut in arranging the figure to facilitate identification of the bands – tracks (a), (b), (c), (e) and (f). By this technique Sall fragment F is not detected. The 32P-labelled Sall fragments of SV-38 DNA were radioautographed, track (d).

To exclude the possibility of the existence of small mol. wt. fragments of SV-38 DNA and overlapping of two fragments with the same or similar mol. wt., 32P-labelled DNA with a sp. act. of 3 to 4 x 10^5 ct/min per sample was restricted and fractionated in 1.4% agarose tube gels. From the distribution of the 32P-labelled DNA across the gel it was apparent that the number of labelled peaks and their radioactivity after restriction with EcoRI, BglII and Sall corresponds to the number of fragments estimated on non-labelled preparations.

By the correlation of the specific activity of the restriction fragments, taking as a base the average mol. wt. of 21.4 x 10^6, we have calculated their mol. wt. (Table 1). The values of the mol. wt. of the fragments calculated by these means exclude the overlapping of two frag-
Table I. Mol. wt. of the SV-38 DNA restriction endonuclease fragments obtained by cleavage with EcoR1, SalI and BglII

<table>
<thead>
<tr>
<th>DNA fragments</th>
<th>BglII</th>
<th>SalI</th>
<th>EcoR1</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mol. wt*</td>
<td>Mol. wt. by 32p X 10^-6†</td>
<td>Mol. wt. by 32p X 10^-6</td>
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<tr>
<td>A</td>
<td>9.3</td>
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<td>6.5</td>
</tr>
<tr>
<td>B</td>
<td>5.6</td>
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<td>5.4</td>
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<tr>
<td>C</td>
<td>3.3</td>
<td>6.1‡</td>
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<td>D</td>
<td>2.9</td>
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</tr>
<tr>
<td>E</td>
<td>—</td>
<td>—</td>
<td>2.5</td>
</tr>
<tr>
<td>F</td>
<td>—</td>
<td>—</td>
<td>0.25</td>
</tr>
</tbody>
</table>

* The mol. wt. estimated by the relative electrophoretic mobility.
† The mol. wt. estimated by the specific activity of the 32p-labelled DNA.
‡ The value of the mol. wt. of fragments situated close to each other is given as a sum of their masses.

Restriction endonuclease maps

To establish the sequence of fragments within the DNA molecule we have used the following approaches. We found that the SV-38 DNA-protein complex is retained almost quantitatively on the surface of the gel, which is in accordance with our previous results on simian adenovirus 7 (Naroditsky et al. 1978). If the DNA-protein complex is cleaved, both terminal fragments are retained on the gel surface which agrees with the previous results of Sharp et al. (1976) concerning human adenovirus 5 DNA-protein complex.

When SV-38 DNA protein complex is restricted with BglII, fragments A and B are retained at the gel surface and after restriction with EcoR1, fragments B and D are retained. After digestion of this material prior to electrophoresis with Pronase the retention of BglII A and B fragments and of EcoR1 B and D fragments is eliminated and after electrophoresis they migrate to reach their theoretically expected places (Fig. 6). These results establish the terminal position of the BglII fragments A and B and the EcoR1 fragments B and D respectively.

Using selected conditions we have examined products of partial digestion with restriction enzymes. Under such conditions we also found products of partial digestion I, II, III and IV in addition to the main A, B, C and D BglII fragments (Fig. 5), with mol. wt. corresponding to the theoretically estimated values of complex fragments DCA, AC, BD and DC (Table 2).

The BD fragment labelled with 32p was eluted from the gel and then re-digested with BglII. The restricted labelled BD fragment was mixed with non-labelled SV-38 DNA previously restricted with BglII, which served as a marker and fractionated on a 1% agarose tube gel. It is apparent that the radioactivity is present only in fragments B and D (Fig. 7). Thus the analysed fragment of partial hydrolysis is BD which shows that B and D are adjacent fragments. Bearing in mind that BglII fragments A and B are terminal, we have
Fig. 6. Agarose-gel electrophoresis of SV-38 DNA-protein complex cleaved with restriction endonucleases EcoRI and BglII. As can be seen EcoRI fragments B and D (2), and BglII fragments A and B (4) are retained at the gel surface. After digestion of the restricted DNA-protein complex with Pronase prior to electrophoresis, the EcoRI B and D fragments (1), and BglII A and B fragments (3) are not retained.
Table 2. Mol. wt. of SV-38 DNA restriction endonuclease fragments obtained by cleavage with EcoRI and BglII and by double cleavage with EcoRI and BglII

<table>
<thead>
<tr>
<th>Partial hydrolysis with EcoRI</th>
<th>DNA fragments</th>
<th>Mol. wt. × 10^-6</th>
<th>Partial hydrolysis with BglII</th>
<th>DNA fragments</th>
<th>Mol. wt. × 10^-6</th>
<th>Double cleavage with EcoRI and BglII</th>
<th>DNA fragments</th>
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<tbody>
<tr>
<td>I</td>
<td>I</td>
<td>19.5</td>
<td>I</td>
<td>I</td>
<td>15</td>
<td>EcoRI A</td>
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<td></td>
</tr>
<tr>
<td>II</td>
<td>II</td>
<td>12.4</td>
<td>II</td>
<td>II</td>
<td>12</td>
<td>BglII B</td>
<td>5.6</td>
<td></td>
</tr>
<tr>
<td>III</td>
<td>III</td>
<td>9.4</td>
<td>A</td>
<td>A</td>
<td>9.3</td>
<td>I</td>
<td>2.2</td>
<td></td>
</tr>
<tr>
<td>A</td>
<td>III</td>
<td>8.2</td>
<td>III</td>
<td>III</td>
<td>8.4</td>
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<tr>
<td>B</td>
<td>IV</td>
<td>6.5</td>
<td>IV</td>
<td>IV</td>
<td>6.15</td>
<td>EcoRI D</td>
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<td></td>
</tr>
<tr>
<td>IV</td>
<td>B</td>
<td>5.1</td>
<td>B</td>
<td>B</td>
<td>5.6</td>
<td>EcoRI E</td>
<td>1.07</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>C</td>
<td>4.0</td>
<td>C</td>
<td>C</td>
<td>3.3</td>
<td>--</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>D</td>
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<td>D</td>
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<td>--</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>E</td>
<td>E</td>
<td>1.07</td>
<td>--</td>
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</tbody>
</table>

Fig. 7. Distribution of specific activity of 32P-labelled BglII complex fragment BD, after elution from agarose gel, re-cleavage with BglII and fractionation in agarose gels. A, B, C and D denote the positions of the simultaneously fractionated unlabelled BglII marker fragments of SV-38 DNA.

deduced that the order of the BglII fragments within the SV-38 DNA molecule is BDCA (Fig. 9).

Under conditions permitting partial digestion of SV-38 DNA with EcoRI along with the main EcoRI fragments A, B, C, D and E, we have found partial digestion products I, II, III and IV (Fig. 5) with mol. wt. corresponding to the theoretically estimated values of complex fragments BCEA, BCE, AD and CE (Table 2).

A double cleavage of SV-38 DNA was performed with BglII and EcoRI. In this case we found six fragments (Fig. 8) among which we detected EcoRI A and D and E fragments and BglII B fragment. Fragments I and II were products of the double cleavage. It is apparent that BglII A fragment is restricted while EcoRI A fragment remains intact. Taking into consideration the mol. wt. of fragments produced as a result of the double
Fig. 8. Agarose-gel fragmentation of SV-38 DNA fragments obtained by double cleavage with EcoRI and BglII (see Methods). Conditions of electrophoresis are the same as listed for Fig. 5. (1) BglII fragments of SV-38 DNA; (2) EcoRI fragments of SV-38 DNA; (3) Double cleavage of SV-38 DNA with EcoRI and BglII.

cleavage (Table 2), we have deduced that the EcoRI site of restriction is located inside the BglII A fragment and that EcoRI fragments A and D are located within BglII A fragment (Fig. 9). Thus, EcoRI fragments A and D are neighbours. As seen from Fig. 8 EcoRI fragment C is not detected among the double cleavage products and it follows that the BglII site of restriction is located inside EcoRI fragment C whereas BglII cleaves EcoRI fragment C into two parts which apparently are the products of the double cleavage I and II. Meanwhile EcoRI fragment E remains intact. Thus, it follows that EcoRI fragments B
and C are neighbours. The assumption that EcoRI fragments B and E are neighbours is not possible, because, as seen from Fig. 8, BglII fragment C is absent after the double cleavage.

From the fact that EcoRI fragments B and D are terminal, that A and D are adjacent on the one hand and B and C are neighbours on the other, we have concluded that the restriction endonuclease map for EcoRI is BCEAD (Fig. 9).

**DISCUSSION**

According to Hull *et al.* (1965), SV-38 is oncogenic. According to Piña & Green (1965) the mol. wt. of the oncogenic adenovirus type 12 DNA is $21 \times 10^6$, as compared to the mol. wt. of DNA from non-oncogenic adenoviruses which is $23 \times 10^6$. This agrees with our data regarding the mol. wt. of the oncogenic SV-38 DNA. Our estimation that the GC content of SA-7 is 58% (Dubitchev *et al.* 1978) agrees with the value estimated by Goodheart (1971) of 57.2% GC. In the present work, using CsCl and Cs$_2$SO$_4$ density gradient ultracentrifugation and thermal denaturation of SV-38 DNA, we have estimated that the GC content of SV-38 DNA corresponds to 59.3%. Goodheart (1971), using only ultracentrifugation in CsCl density gradient, estimated the GC content of SV-38 DNA to be 55.4%. Probably the former value, based on three independent estimations, is more reliable. Piña & Green (1965) have grouped the human adenoviruses on the bases of their GC content, biological characteristics and nucleic acid hybridization. According to them, the 47 to 49% GC group are highly oncogenic, the 50 to 53% GC group are weakly oncogenic and the 56 to 60% GC group are non-oncogenic.

Goodheart (1971) has shown that the oncogenic simian adenoviruses have a statistically significant tendency to higher percentages of GC than the non-oncogenic simian adenoviruses. It was shown that the mean molar ratio for the oncogenic simian viruses is 56.92% GC as compared to 54.26% GC for the non-oncogenic simian adenoviruses. We have shown that the oncogenic SA-7 and SV-38 have a higher GC content than the mean molar ratio characteristic for the oncogenic simian adenoviruses. This is consistent with the general tendency of the simian oncogenic adenoviruses to show a higher GC content (Goodheart, 1971).

Sharp *et al.* (1973) have shown that when adeno type 5 DNA protein complex, isolated by treating viruses with guanidinium chloride, is cut by restriction endonuclease, both terminal fragments are retained on the gel surface, while all internal fragments migrated through the gel at the rate expected from their mol. wt. Digestion with pronase of the restriction endo-
nuclease cleavage products of the DNA-protein complex eliminates the anomalous retention on the gel surface of the terminal fragments. Determination of the terminal fragments by the described procedure provides a simple and inexpensive method for the establishment of terminal fragments in the case of adeno or other viruses with terminal protein (see also Naroditsky et al. 1978). After digestion with EcoRI, SalI, BamHI and BglII the SA-7 DNA is cleaved into fragments with mol. wt. ranging from 12 to \(0.6 \times 10^6\). SA-7 DNA has one site of restriction for EcoRI, as compared to four sites of restriction for SV-38 DNA. SA-7 DNA has four sites of restriction with BglII (B. S. Naroditsky, unpublished observations), while SV-38 DNA has three. The sequence of the EcoRI and BglII fragments within the DNA molecules of SV-38 and SA-7 is different.

SA-7 was isolated from African green monkey while the simian adenoviruses from the SV series were of Rhesus or cynomolgus monkey origin (Hull et al. 1965) and although the SA-7 and SV-38 are both oncogenic, obviously there are differences in their genomes.

REFERENCES


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