JOURNAL OF VIROLOGY, May 1988, p. 1741–1745
0022-538X/88/051741-05$02.00/0

Vol. 62, No. 5

Identification of the Gene Coding for the Precursor of Adenovirus Core Protein X

JOSEPH M. WEBER[1]* AND CARL W. ANDERSON[2]

Département de Microbiologie, Faculté de Médecine Université de Sherbrooke, Sherbrooke, Québec, Canada J1H 5N4,[1] and Biology Department, Brookhaven National Laboratory, Upton, New York 11973[2]

Received 7 October 1987/Accepted 26 January 1988

Nucleoprotein complexes extracted from infected cells (J. Weber and L. Philipson, Virology 136:321–327, 1984) or *ts*1 virions grown at 39°C (J. M. Weber, G. Khitto, and A. R. Bhatti, Can. J. Microbiol. 29:235–241, 1983) contain, in addition to known virion proteins, a polypeptide with a molecular weight of 11,000 (11K polypeptide). We identified the gene for this polypeptide by sequence analysis of the radioactively labeled 11K polypeptide isolated from *ts*1 virions and comparison of this sequence with the nucleotide sequence of the adenovirus type 2 genome. The 11K polypeptide was encoded by an open reading frame of 80 residues located between nucleotides 17,676 and 17,915 in late transcription region 2 of adenvirus type 2; the initiating methionine residue was removed leaving a 79-residue product. The late transcription region 2 that encoded the 79-residue polypeptide (11K) was arginine rich (21%) and had a predicted molecular weight of 8,715. It was cleaved by the viral endoprotease to give two products which comigrated on sodium dodecyl sulfate-polyacrylamide gels as virion polypeptide X. Our data suggest that additional cleavage of the carboxy-terminal 48-residue fragment generates a 19-amino-acid fragment with the amino acid composition of mu (K. Hosokawa and M. T. Sung, J. Virol. 17:924–934, 1976).

Adenovirus particles contain 10 structural polypeptides whose location and function are relatively well defined (6, 19). There remain, however, several poorly defined virion components whose functions and origins are unknown. Three of those components are present in mature wild-type virions and are designated X, XI, and XII (1). These are small-molecular-weight cleavage products of undetermined precursor proteins. Component X forms part of the core, or viral nucleoprotein complex, along with polypeptides V and VII (21, 23). In studies on virus maturation (22) aided by use of the viral protease-deficient mutant *ts*1, a nucleoprotein complex has been identified that is present in infected cells or *ts*1 virions grown at the nonpermissive temperature and that is composed of polypeptides V, the precursor to component VII (P-VII), and a polypeptide with a molecular weight of 11,000 (11K polypeptide). Here we show by amino-terminal sequence analysis that the 11K polypeptide is a late transcription region 2 (L2)-encoded protein that is the precursor of two virion polypeptides. These two polypeptides migrated together during sodium dodecyl sulfate (SDS)-polyacrylamide gel electrophoresis (PAGE) as virion component X.

MATERIALS AND METHODS

Cells and viruses. Viruses were propagated in HEp-2 cells (12). The *ts*1 mutation maps in the virus-encoded protease and prevents proteolytic cleavage of at least five adenovirus precursor polypeptides at the nonpermissive temperature (20, 24).

HPLC purification of virion component XI. Purified adenovirus type 2 (Ad2) virions labeled with [^{35}S]methionine were prepared as described below. The virions were disrupted in 4 M guanidine hydrochloride and chromatographed on Sephadex G75 adjusted to 4 M guanidine hydrochloride. Two peaks eluted, with the second peak containing exclusively protein components XI to XII. This peak was dialyzed and further purified by reverse-phase high-pressure liquid chromatography (HPLC) on a chromatograph (model 5020; Varian Instruments). The octadecylsilane-3 column was used with a linear gradient of buffer A (15% acetonitrile, 0.1% trifluoroacetic acid [pH 2.4]) and buffer B (95% acetonitrile, 0.1% trifluoroacetic acid [pH 2.3]). Radioactive peaks were checked by SDS-PAGE, which showed a component XI that was suitable for sequencing.

Protein sequence analysis. The radioactively labeled 11K polypeptide was prepared from purified *ts*1 virions grown at 39°C; component X was prepared from wild-type Ad2 virions. Infected cells were labeled separately with [^{35}S] methionine (250 μCi/ml), [^{3}H]leucine (50 μCi/ml), or [^{3}H] arginine (50 μCi/ml) as follows. At 18 h after infection, the labeled amino acid in 0.2 ml of Dulbecco modified essential medium was added to a petri dish (diameter, 10 cm), and at 22 h after infection 10 ml of complete Dulbecco modified essential medium was added. Labeled cultures were harvested at 48 h after infection, and purified virions were prepared by CsCl gradient centrifugation.

Appropriate amounts of methionine-labeled virions were mixed with each of the tritium-labeled virus preparations, and the virion proteins were fractionated on 12 or 20% SDS-polyacrylamide gels (12) or on 20% gels prepared as described by Thomas and Kornberg (18). Gels were dried without fixation and exposed to film to localize the polypeptides of interest. These were excised, recovered by electroelution, and subjected to amino-terminal sequence analysis with a protein sequencer (890C; Beckman Instruments, Inc., Fullerton, Calif.) as described previously (2). In some cases core polypeptides were obtained from pyridine-disrupted virions as described previously (5).

RESULTS

Identification of the 11K polypeptide gene. Previously, nucleoprotein complexes derived from Ad2 *ts*1 virions produced at the nonpermissive temperature were found to contain a novel 11K polypeptide not found in wild-type

* Corresponding author.

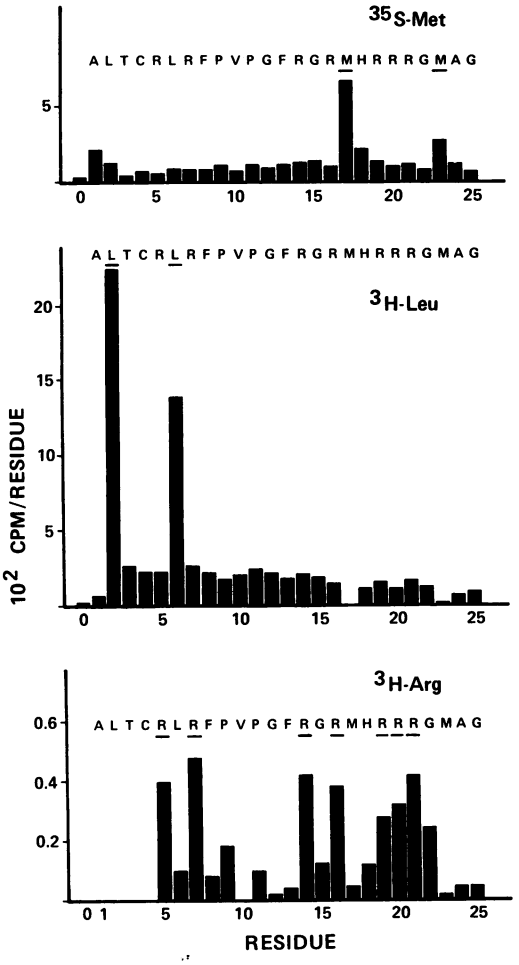
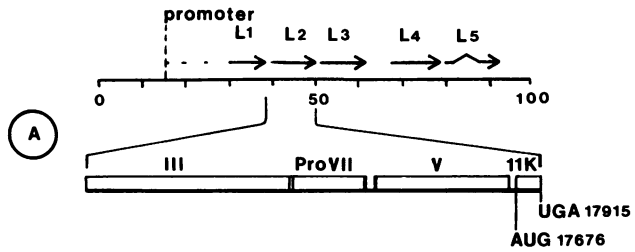


FIG. 1. Partial amino-terminal sequence analysis of the *ts*1 11K polypeptide. The 11K polypeptide was prepared from the *ts*1 virions labeled with [^{35}S]methionine, [^{3}H]leucine, or [^{3}H]arginine described in the text and applied to a protein sequencer (890C; Beckman). The profile of radioactivity released after each Edman degradation cycle is shown. The first cycle (labeled 0) was performed without the addition of phenylisothiocyanate. The predicted amino acid sequence of the L2-79R reading frame is given at the top of each panel. Radioactivity applied to the sequencer was 28,700 cpm of [^{35}S] methionine, 63,200 cpm of [^{3}H]leucine, or 2,300 cpm of [^{3}H] arginine.

virions (22, 23). To determine the origin of this polypeptide, we prepared *ts*1 virions labeled with different amino acids, isolated the 11K polypeptide by SDS-PAGE, and subjected these preparations to amino-terminal sequence analysis. Edman degradation of the [^{35}S]methionine-labeled 11K polypeptide indicated that methionine was present at residues 17 and 23; leucine was found at residues 2 and 6; and arginine was found at residues 5, 7, 14, 16, 19, 20 and 21 (Fig. 1). A computer search of the sequence of the Ad2 genome (16) revealed a single open reading frame (ORF) of 80 amino acids that could encode this pattern of amino acids (Fig. 2). Immediately prior to leucine at position 2 in the 11K polypeptide, the ORF predicted the sequence Met-Ala. Polypeptides that begin with the sequence Met-Ala- frequently are processed during synthesis to remove the amino-terminal methionine (10). The 11K polypeptide ORF is found at the end of L2, which also encodes virion components III (penton base), P-VII, and core component V. Just prior to the first methionine codon in the 11K polypeptide ORF, and following the termination codon for component V, is the sequence TTCTTGCAG. This sequence is homologous to the mRNA splice acceptor consensus sequence (13). Together, these facts suggest that at late times, an mRNA is produced from L2 that encodes the 79-residue (L2-79R) 11K polypeptide found in *ts*1 virions.

The L2-79R sequence does not contain tyrosine. This fact was previously predicted from an inability to label the *ts*1 11K polypeptide with ^{125}I (7).

In vitro cleavage of the *ts*1 11K polypeptide. The methionine-labeled L2-79R (11K) polypeptide was prepared by SDS-PAGE, and after removal of SDS, the polypeptide was incubated with extracts of Ad2-infected cells that are capable of cleaving P-VII to VII (3). Figure 3 (lane e) shows the best cleavage obtained. About half of the 11K polypeptide was cleaved to yield three products: a major intermediate fragment that comigrated with a band present only in incompletely cleaved *ts*1 (in Fig. 3, compare lane e with lanes f and g), a band that comigrated with component X, and a band that comigrated with component XI. Attempts to produce complete cleavage of the purified 11K polypeptide were unsuccessful. Incomplete or inappropriate cleavage in vitro might result from the incomplete removal of detergent or from incomplete refolding of the polypeptide. Complete cleavage of the 11K polypeptide was obtained when *ts*1 virion proteins were digested with protease (Fig. 3, lane b). Component X was a discernible cleavage product (in Fig. 3, compare lane a with lane b). These results suggest that the 11K polypeptide may undergo cleavage at several sites and that components X and XI might be cleavage products.



B

11K Sequence

```
                5                  10                  15
ALA LEU THR CYS ARG LEU ARG PHE PRO VAL PRO GLY PHE ARG GLY
               20                  25                  30
ARG MET HIS ARG ARG ARG GLY MET ALA GLY HIS GLY LEU THR GLY
               35                  40                  45
GLY MET ARG ARG ALA HIS HIS ARG ARG ARG ARG ALA SER HIS ARG
               50                  55                  60
ARG MET ARG GLY GLY ILE LEU PRO LEU LEU ILE PRO LEU ILE ALA
               65                  70                  75
ALA ALA ILE GLY ALA VAL PRO GLY ILE ALA SER VAL ALA LEU GLN
               80
ALA GLN ARG HIS
```

C

AMINO ACID COMPOSITION

11	ALA	A	2	GLN	Q	8	LEU	L	2	SER	S
17	ARG	R	0	GLU	E	0	LYS	K	2	THR	T
0	ASN	N	11	GLY	G	4	MET	M	0	TRP	W
0	ASP	D	6	HIS	H	2	PHE	F	0	TYR	Y
1	CYS	C	5	ILE	I	5	PRO	P	3	VAL	V

FIG. 2. Map location, sequence, and amino acid composition of the 11K polypeptide. (A) Representation of the Ad2 genome with an enlargement of L2. The 80-residue 11K polypeptide ORF lies at the 3′ end of L2 between nucleotides 17,676 and 17,915. L2 also encodes three other virion components: penton base (III), the precursor to component VII (Pro-VII), and core component V. (B) Predicted 79-amino-acid residue sequence of the 11K protein after removal of the amino-terminal methionine. (C) Amino acid composition of the 11K polypeptide.

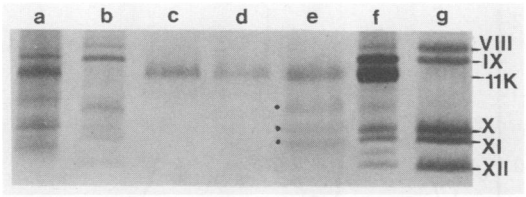


FIG. 3. In vitro cleavage of the L2-79R (11K) core precursor. The L2-79R polypeptide, which was labeled with [^{35}S]methionine, was purified from *ts*1 virions grown at 39°C and incubated with buffer (lane c), uninfected nuclei (lane d), nuclei from cells infected with wild-type virus for 24 h (lane e). The dots denote cleavage fragments. The enzyme assay was performed as described previously (20). Additional controls consisted of *ts*1 virion proteins grown at 39°C and disrupted (lane a) and the sample from lane a incubated with proteinase (infected cell nuclei) as described for lane e (lane b). The marker lanes of the SDS-polyacrylamide gel show *ts*1 virion proteins showing partial cleavage (lane f) and wild-type virion proteins (lane g).

Sequence analysis of virion components X, XI, and XII. To determine whether one or more of the small-molecular-weight virion polypeptides might be the product of L2-79R processing, these polypeptides, which were labeled with one of several tritiated amino acids and [^{35}S]methionine, were isolated from wild-type Ad2 virions and subjected to amino-terminal sequence analysis. Results of the analysis of component X isolated from pyridine cores labeled with [^{35}S]methionine are given in Fig. 4. Methionine was found at positions 1, 16, 17, and 23. Note that the amino terminus of the 11K polypeptide gave methionines in positions 17 and 23. Thus, this result strongly suggests that component X includes the amino terminus of the 11K polypeptide.

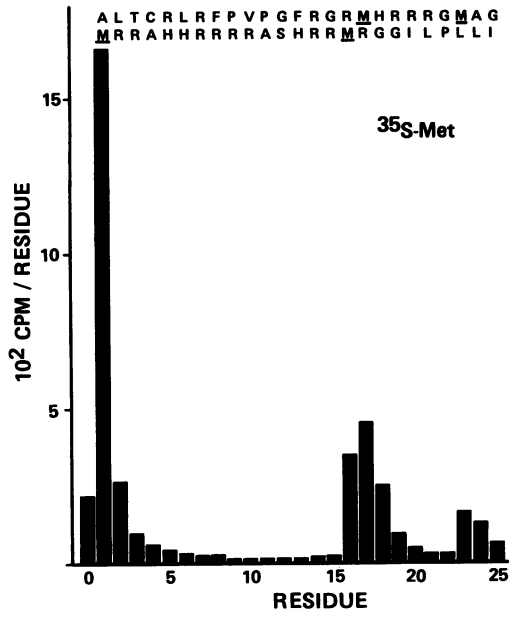


FIG. 4. Amino-terminal sequence analysis of virion component X. Pyridine cores were prepared from [^{35}S]methionine-labeled wild-type virions. Component X was excised from SDS-polyacrylamide gels, eluted, and applied to the sequencer. The release of radioactivity after each Edman cycle is shown. Phenylisothiocyanate delivery was omitted during the first cycle to wash out any unincorporated radioactivity. Residues 1 to 24 (beginning with alanine) and 32 to 56 of the L2-79R (11K) protein are shown at the top of the figure in single-letter code. Radioactivity applied to the sequencer was 17,610 cpm of [^{35}S]methionine.

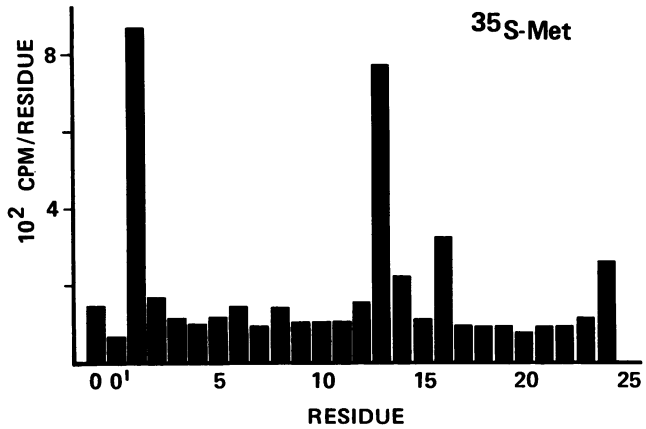


FIG. 5. Amino-terminal sequence analysis of HPLC-purified virion component XI. Virion component XI, which was labeled with [^{35}S]methionine, was purified from wild-type virions by reverse-phase HPLC, as described in the text, and 17,500 cpm was applied to a protein sequencer (890C; Beckman). As described in the legend to Fig. 1, phenylisothiocyanate was omitted from the first two cycles (labeled 0 and 0'). Consideration of repetitive yields suggested the presence of two methionine-labeled components, one with methionines as residues 1 and 16 and the second with methionine as residue 13. The latter component was present in about a twofold molar excess over the former.

A computer search of both strands of the Ad2 sequence revealed only 25 sites that could directly encode (excluding splicing) two methionine residues 15 codons apart (i.e, in residues 1 and 16; see Fig. 4). All but two of these sites can be excluded from consideration because of intervening termination or other methionine codons. One site was in a short reading frame located on the early strand between nucleotides 17,935 and 17,445 that is not known to be expressed during infection. The second site fell in the 11K polypeptide reading frame beginning with residue 32; residue 47, 15 amino acid residues beyond residue 32, was also predicted to be methionine. This finding suggests that the L2-79R protein is cleaved after the Gly-Gly sequence of residues 30 and 31 to yield two fragments that migrate with virion component X.

To provide further support for cleavage of L2-79R after residue 31, [^{3}H]arginine-labeled components X and XI were prepared and subjected to amino-terminal sequence analysis. Arginine was found in residues 2, 3, 5, 7, 8, 9, 10, 14, 15, 16, 17, 19, 20, and 21, as expected (see Fig. 2), if the sample consisted of an equal molar mixture of the two predicted fragments of the L2-79R protein (data not shown).

Analysis of methionine-labeled component XI that was purified by reverse-phase HPLC yielded a major component with methionine as residue 13 and a lesser amount of the carboxy-terminal component of L2-79R beginning with residue 32 (Met at positions 1 and 16; Fig. 5). Cleavage between residue 34 and alanine would account for the methionine at position 13. This result suggests that the carboxy-terminal 48-amino-acid fragment of L2-79R may be further processed near its carboxy terminus, to produce products that migrate both with components X and XI.

Sequence analysis of component XII yielded a methionine at position 8 and an arginine at residue 3; a small amount of leucine was found in residues 2 and 5 (data not shown). Although a peptide with leucine as residue 2, arginine as residue 3, and methionine as residue 8 could be derived from the 24-residue amino-terminal fragment of P-VII that was

removed by the Ad2 proteinase, our data were not sufficient to establish the identity of component XII.

DISCUSSION

The data we have presented identify a new adenovirus virion protein precursor that is processed by the adenovirus endoproteinase. This 79-residue basic polypeptide, previously referred to as the 11K polypeptide (22, 23), is encoded by L2. Previously, it was shown (23) that the 11K component is associated in virions with the chromatin core of the virus particle. A small L2 mRNA spanning the region in which the 79R ORF lies has been mapped by electron microscopy (4).

In wild-type infected cells, the 79R protein is cleaved, to form virion core component X. Results of our analyses indicate that component X itself represents at least two polypeptides, both of which are derived from L2-79R. Cleavage of L2-79R occurs after residue 31 in the sequence -Thr-Gly-Gly:Met-Arg-Arg-. If this were the only site of cleavage, the two fragments would be 31 and 48 residues and would have predicted molecular weights of 3,422 and 5,311, respectively. Although small peptides frequently run with highly anomalous mobilities during SDS-PAGE, we suspect that the carboxy-terminal 48-amino-acid fragment may be further processed by the proteinase. Indeed, component XI contained a peptide with the amino-terminal sequence of the 48-residue L2-79R fragment, plus another unidentified peptide, but did not contain the amino-terminal 31-amino-acid fragment (Fig. 5). Both Gly-Gly and Gly-Ala sequences, both of which are potential cleavage sites, occurred in the carboxy-terminal portion of the 48-residue fragment (Fig. 2B). Since the carboxy-terminal fragment would not contain methionine, cleavage at either of these sites would not have been easily detected.

The cleavage site in the L2-79R protein resembled the two other known viral endoproteinase cleavage sites in that for all three, cleavage occurred after adjacent glycine residues and prior to a moderately hydrophobic residue. The sequence surrounding the cleavage site in P-VI is -Asn-Met-Ser-Gly-Gly:Ala-Phe-Ser-Trp-, while that in P-VII is -Lys-Met-Phe-Gly-Gly:Ala-Lys-Lys-Arg- (15, 17). The recognition specificity of the Ad2 endoproteinase is unlikely to be a simple Gly-Gly-Ala or Gly-Gly-Met, as these elements occur in Ad2 proteins that are not substrates for the endoproteinase including hexon, fiber, and the early E1B 495-residue protein. The dipeptide Gly-Gly, in addition to appearing at the three known cleavage sites, also appeared three times in the carboxy-terminal sequence of P-VIII, at least six times in the sequence of the terminal protein precursor, and once in the endoproteinase precursor itself. Thus, Gly-Gly may be an element in the recognition sequence. Complete elucidation of the endoproteinase specificity will require sequence analysis at additional cleavage sites as well as the construction of cleavage site mutants.

Examination of the predicted sequence of L2-79R permits the identification of several structural features. The protein is highly basic, having 21% arginine and 8% histidine but, curiously, no lysine. The acidic residues aspartic acid and glutamic acid were absent, as was asparagine and the aromatic residues tryptophan and tyrosine. There were 4 methionine residues, 2 in the amino-terminal 31 residues and 2 in the amino-terminal half of the 48-residue fragment; there was only 1 cysteine residue located near the L2-79R amino terminus. The basic groups were divided almost evenly between the amino-terminal and carboxy-terminal fragments, but the basic residues in the carboxy-terminal fragment were highly clustered; 12 of the first 17 residues of the 48-amino-acid fragment were arginine or histidine. The hydropathy analysis identified two pronounced hydrophilic domains separated by the cleavage site at residue 31 (Fig. 6). A computer search of the protein sequence data banks has not revealed highly significant homologies with other proteins, although the central segment does resemble the protamines.

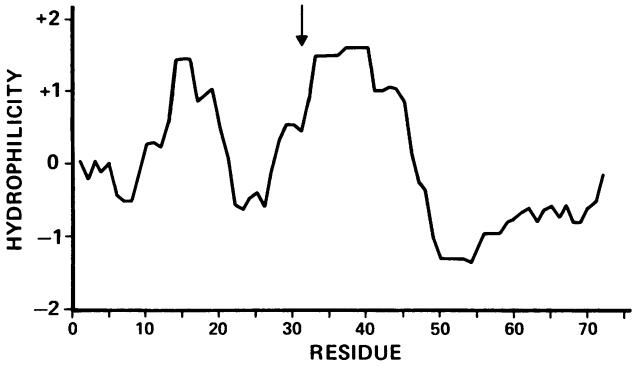


FIG. 6. Hydrophilicity profile of the L2-79R core precursor. The average hydrophilicity over a seven-amino-acid segment plotted by the method of Hopp and Woods (8). The arrow marks the site of cleavage at residue 31.

The basic nature of the L2-79R polypeptide is consistent with its location in the chromatin core. Based on a comparison of the [^{35}S]methionine-labeled *ts*1 virion proteins P-VI and P-VII, there appear to be about 340 copies of the precursor per virion, and virions contain approximately 180 nucleosome-like structures (14). This would provide two copies of the 79R precursor per nucleosome. It is tempting to suggest that the role of the precursor might be to condense the viral prochromatin for encapsidation by virtue of the two basic domains. Subsequent cleavage within the particle after residue 31, and perhaps also after residues 50 and 64, would release the cross-link and would prepare the viral chromatin for the relaxed conformation required during subsequent infection and uncoating. The importance of cleavage for infectivity is borne out by the fact that *ts*1 virions are not infectious. The block to infection is manifested as a failure to uncoat (11, 12). Although it is uncertain whether this effect can be ascribed solely to cleavage of the 79R precursor, with the coding location established, it should now be possible to construct mutants with altered cleavage sites to test this hypothesis.

Hosokawa and Sung (9) reported that Ad5 virions contained an extremely basic protein, with a mobility about the same as those of components X to XII, that they called mu. Mu is located in the chromatin core (9, 21); thus, we considered whether mu might be derived from the L2-79R precursor. The composition of mu reported by Hosokawa and Sung (9) was quite different from the predicted compositions of either the amino-terminal 31-amino-acid fragment or the 48-amino-acid carboxy-terminal fragment, but was quite close to that predicted for a fragment beginning at residue 32 (Met) and extending through the hypothetical Gly-Gly cleavage site at residue 49. This 19-amino-acid fragment would be composed of only 6 different amino acids and would have the following composition, in mole percent: Met, 10.5; Arg, 47.4; Ala, 10.5; His, 15.8; Ser, 5.3; Gly, 10.5. Except for a higher value for methionine, this composition is very close to the composition reported for mu (9). Although

we have been unable to obtain sufficient mu purified as described by Hosokawa and Sung for sequence analysis, we suggest that this is a likely identity for mu. Thus, mu probably is the carboxy-terminal L2-79R fragment observed in our preparation of component X and identical to the component X observed in core preparations (21–23). Efforts to confirm this hypothesis are in progress.

Components X to XII have been presumed to be cleavage fragments resulting from proteolytic processing of several virion components by the viral endoproteinase; however, their identities have remained elusive. Although we have now identified the origin of two of these peptides, results of our analyses suggest that at least half a dozen small peptides are found in virions. We did not obtain sufficient sequence information to identify the origin of the remaining peptides. Several cleavage products are not expected to contain methionine and thus would not have been observed by most investigators. Several cleavage products, such as the 24-amino-acid fragment removed from P-VII, may have blocked the amino termini and thus would not have been detected in our experiments. It remains to be seen whether any of these fragments have a function in virion structure after cleavage from their precursors by the endoproteinase.

ACKNOWLEDGMENTS

We thank Richard Feldman, Jeanne Wysocki, and Lise Clavet for expert technical assistance.

C.W.A. is supported by the Office of Health and Environmental Research of the U.S. Department of Energy. J.M.W. is supported by a grant from the Medical Research Council of Canada and a career award from the National Cancer Institute of Canada.

LITERATURE CITED

1. **Anderson, C. W., P. R. Baum, and R. F. Gesteland.** 1973. Processing of adenovirus 2-induced proteins. J. Virol. **12**:241–252.
2. **Anderson, C. W., and J. B. Lewis.** 1980. Amino-terminal sequence analysis of adenovirus type 2 proteins: hexon, fiber, component IX, and early protein 1B-15K. Virology **104**:21–41.
3. **Bhatti, A. R., and J. Weber.** 1979. Protease of adenovirus type 2: partial characterization. Virology **96**:478–485.
4. **Chow, L. T., J. M. Roberts, J. B. Lewis, and T. R. Broker.** 1977. A map of cytoplasmic RNA transcripts from lytic adenovirus type 2, determined by electron microscopy of RNA:DNA hybrids. Cell **11**:819–836.
5. **Déry, C. V., G. De Murcia, D. Lamarre, N. Morin, G. G. Poirier, and J. Weber.** 1986. Possible role of ADP-ribosylation of adenovirus core proteins in virus infection. Virus Res. **4**:313–329.
6. **Ginsberg, G.** 1984. The adenoviruses, p. 605. Plenum Publishing Corp., New York.
7. **Hannan, C., L. Raptis, C. Déry, and J. Weber.** 1983. Biological and structural studies with an adenovirus type 2 temperature-sensitive mutant defective for uncoating. Intervirology **19**:213–223.
8. **Hopp, T. P., and K. R. Woods.** 1981. Prediction of protein antigenic determinants from amino acid sequences. Proc. Natl. Acad. Sci. USA **78**:3824–3828.
9. **Hosokawa, K., and M. T. Sung.** 1976. Isolation and characterization of an extremely basic protein from adenovirus type 5. J. Virol. **17**:924–934.
10. **Jornvall, H.** 1975. Acetylation of protein N-terminal amino groups structural observations on α-amino acetylated proteins. J. Theor. Biol. **55**:1–12.
11. **Miles, B. D., R. B. Luftig, J. A. Weatherbee, R. R. Weihing, and J. Weber.** 1980. Quantitation of the interaction between adenovirus types 2 and 5 and microtubules inside infected cells. Virology **150**:265–269.
12. **Mirza, M. A. A., and J. Weber.** 1979. Uncoating of adenovirus type 2. J. Virol. **30**:462–471.
13. **Mount, S. M.** 1982. A catalogue of splice junction sequences. Nucleic Acids Res. **10**:459–472.
14. **Nermut, M. V.** 1984. The architecture of adenoviruses, p. 5–34. *In* H. S. Ginsberg (ed.), The adenoviruses. Plenum Publishing Corp., New York.
15. **Pettersson, U.** 1984. Structural and nonstructural adenovirus proteins, p. 205–270. *In* H. S. Ginsberg (ed.), The adenoviruses. Plenum Publishing Corp., New York.
16. **Roberts, R. J., G. Akusjarvi, P. Alestrom, R. E. Gelinas, T. R. Gingeras, and D. Sciaky.** 1986. A consensus sequence for the adenovirus-2 genome, p. 1–51. *In* W. Doerfler (ed.), Adenovirus DNA. The viral genome and its expression. Martinus Nijhoff Publishing, Boston.
17. **Sung, M. T., T. M. Cao, R. T. Coleman, and K. A. Budelier.** 1983. Gene and protein sequences of adenovirus protein VII, a hybrid basic chromosomal protein. Proc. Natl. Acad. Sci. USA **80**:2902–2906.
18. **Thomas, J. O., and R. D. Kornberg.** 1978. The study of histone-histone associations by chemical cross-linking, p. 429–440. *In* G. Stein, J. Stein, and L. J. Kleinsmith (ed.), Methods in cell biology, vol. 18. Academic Press, Inc., New York.
19. **Tooze, J.** 1980. DNA tumor viruses. Part 2, p. 443–546. Cold Spring Harbor Laboratory, Cold Spring Harbor, N.Y.
20. **Tremblay, M. L., C. V. Déry, B. G. Talbot, and J. Weber.** 1983. In vitro cleavage specificity of the adenovirus type 2 proteinase. Biochem. Biophys. Acta **743**:239–245.
21. **Vayda, M. E., A. E. Rogers, and S. J. Flint.** 1983. The structure of nucleoprotein cores released from adenovirions. Nucleic Acids Res. **11**:441–460.
22. **Weber, J., and L. Philipson.** 1984. Protein composition of adenovirus nucleoprotein complexes extracted from infected cells. Virology **136**:321–327.
23. **Weber, J. M., G. Khittoo, and A. R. Bhatti.** 1983. Adenovirus core proteins. Can. J. Microbiol. **29**:235–241.
24. **Yeh-Kay, L., G. Akusjarvi, P. Alestrom, U. Pettersson, M. Tremblay, and J. Weber.** 1983. Genetic identification of an endoproteinase encoded by the adenovirus genome. J. Mol. Biol. **167**:217–222.