

sera against viruses was discussed.

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1. Enders, J. F., Peebles, T. C., *Proc. Soc. Exp.*

*BIOL. AND MED.*, 1954, v86, 277.

2. Fogh, J., Lund, R. O., *ibid.*, 1957, v94, 532.

3. Hayflick, L., *Exp. Cell Res.*, 1961, v23, 14.

4. Stokes, J., Jr., Reilly, C. M., Hilleman, M. R., Buynak, E. B., *New Eng. J. Med.*, 1960, v263, 230.

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## Amino Acid Composition of Rat and Mouse Tumors.\* (27554)

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It has been shown(1) that there is no significant difference in the total amino acid composition of 2 rat tumors, Sarcoma R-1 and Walker carcinosarcoma 256, grown in ethionine-treated or untreated, male or female, Wistar rats. It was also observed(1,2) that the composition of these tumors was in excellent agreement with that found for the U.C.L.A. fibrosarcoma(3) and for 4 other rat tumors studied by Sauberlich and Baumann (4). It was of interest, therefore, that the content of 6 amino acids (arginine, aspartic acid, lysine, phenylalanine, proline, and serine) reported by Mickelson and Barvick(5) for the Sarcoma 180 mouse tumor differed markedly from that observed for rat tumors (1-4). In all of these experiments amino acids were determined by microbiological assay.

Greenstein(6) and Roberts and Tanaka(7) have stressed the biochemical similarity of tumors and Dunn *et al.*(8) reported that there was no significant difference in total amino acid content of normal rat and mouse tissues. It appeared desirable, therefore, to analyze both types of tumor tissue by column chromatographic and paper chromatographic techniques.

**Materials and methods.** Two samples of Walker carcinosarcoma 256 grown in rats, one sample of Sarcoma 180 grown in mice, and 2

samples of the Taylor mouse tumor, one grown in mice and one grown in incubating eggs, were treated for 3 minutes in boiling water, trimmed free of necrotic tissue and connective tissue, dried at 50-70° in a vacuum oven, extracted with ether-alcohol in a Soxhlet apparatus, redried to constant weight, and hydrolyzed by refluxing in 6N HCl for 24 hours. After filtering and washing the residue, the samples were evaporated to dryness under reduced pressure, dissolved in a minimum of hot water, filtered, and made up to a known volume.

An additional sample of Sarcoma 180 was prepared as above with the exception that it was not treated with boiling water. This control sample was necessary since the treatment with boiling water was included in the procedure of Baginsky, Murphy and Dunn(1) but not in that of Mickelson and Barvick(5).

Two amino acids, phenylalanine and tyrosine, not included in the previous study(1), were determined microbiologically using *Leuconostoc mesenteroides* P-60 (8042) for the phenylalanine and *Lactobacillus casei* (7469) for the tyrosine assays. The general assay procedures were those described previously(1). The column chromatographic methods used were modifications of those of Moore and Stein(9), Ishii(10), and Chinard(11), and the paper chromatographic method was a modification of the procedures of McFarren and Mills(12) and Baker and Khan(13).

**Results.** Since no significant differences were observed between the results for the 2 samples of rat tumor, or the 4 samples of

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TABLE I. Total Amino Acid Content of Rat and Mouse Tumors.

| Amino acid    | Microbiological assay                   |                                     |        |                       | Column chromatography |               | Paper chromatography |               |
|---------------|-----------------------------------------|-------------------------------------|--------|-----------------------|-----------------------|---------------|----------------------|---------------|
|               | Rat*                                    | Mickelson and Barvick <sup>10</sup> | Mouse† | Barvick <sup>10</sup> | Rat†                  | Mouse‡        | Rat†                 | Mouse‡        |
|               | This Laboratory <sup>10</sup>           |                                     |        |                       |                       |               |                      |               |
|               | (%)                                     | (%)                                 | (%)    |                       | (%)                   | (%)           | (%)                  | (%)           |
| Arginine      | 5.3 <sup>§</sup> (5.0-5.7) <sup>§</sup> | 4.12 ± .16                          |        |                       | 5.5 (5.4-5.6)         | 5.0 (4.9-5.1) | 4.6 (4.1-5.1)        | 4.8 (3.4-6.0) |
| Aspartic acid | 7.2 (7.1-7.3)                           | 9.32 ± 1.40                         |        |                       | 7.6 (7.5-7.7)         | 8.4 (8.2-8.8) | 8.1 (7.7-8.5)        | 7.4 (5.3-9.7) |
| Glutamic acid | 9.7 (9.5-10)                            | 10.75 ± .18                         |        |                       | 11 (11-11)            | 11 (11-12)    | 8.6 (7.5-9.2)        | 11 (9.7-13)   |
| Glycine       | 4.6 (4.3-4.7)                           | 4.30 ± .130                         |        |                       | 5.1 (5.0-5.3)         | 4.5 (4.0-4.8) | 5.8 (5.1-6.3)        | 6.4 (4.5-8.9) |
| Histidine     | 2.0 (1.9-2.1)                           | 2.00 ± .070                         |        |                       | 2.0 (1.9-2.1)         | 2.1 (2.0-2.2) | 2.6 (2.3-2.9)        | 2.3 (1.9-2.8) |
| Isoleucine    | 3.6 (3.5-3.8)                           | 3.78 ± .06                          |        |                       | 3.7 (3.4-3.9)         | 3.9 (3.8-4.0) | 7.1 (6.6-7.4)        | 6.7 (5.7-7.5) |
| Leucine       | 6.4 (6.4-6.5)                           | 6.58 ± .12                          |        |                       | 6.6 (6.4-6.8)         | 7.6 (7.3-7.9) | 5.2 (4.5-6.0)        | 4.9 (3.0-7.0) |
| Lysine        | 7.3 (7.0-7.8)                           | 2.77 ± .11                          |        |                       | 7.1 (7.0-7.2)         | 7.4 (7.0-7.7) | 6.0 (5.6-6.3)        | 6.4 (5.5-7.2) |
| Methionine    | 1.8 (1.7-1.8)                           | 1.83 ± .108                         |        |                       | 1.6 (1.5-1.6)         | 1.9 (1.8-2.0) | 0.94 (0.91-0.99)     | 1.4 (1.0-2.0) |
| Phenylalanine | 3.8 (3.1-4.1)**                         | 6.24 ± .13                          |        |                       | 3.5 (3.2-3.8)         | 4.2 (3.8-4.6) | 2.3 (2.0-2.6)        | 2.7 (2.2-3.4) |
| Proline       | 3.6 (3.6-3.7)                           | 4.62 ± .188                         |        |                       | 4.6 (4.5-4.7)         | 4.0 (3.9-4.1) | —                    | —             |
| Serine        | 4.0 (3.8-4.2)                           | 6.68 ± .252                         |        |                       | 4.2 (4.0-4.3)         | 4.5 (4.3-4.7) | 6.2 (5.5-6.9)        | 6.5 (5.8-7.6) |
| Threonine     | 3.5 (3.4-3.6)                           | 3.68 ± .07                          |        |                       | 3.6 (3.5-3.6)         | 3.8 (3.5-4.0) | 11 (10-12)           | 11 (10-14)    |
| Tyrosine      | 3.1 (2.9-3.3)**                         | 7.04 ± .21                          |        |                       | 3.1 (2.8-3.4)         | 3.6 (3.4-3.9) | 1.6 (1.3-1.8)        | 1.9 (1.3-2.2) |
| Valine        | 4.5 (4.3-4.6)                           | 4.30 ± .18                          |        |                       | 4.2 (4.0-4.3)         | 4.9 (4.6-5.2) | 5.1 (4.6-5.6)        | 4.0 (3.2-4.9) |

\* % of dry wt. 4 pooled samples.

† % of dry, fat-free wt. 2 pooled samples.

‡ % of dry, fat-free wt. 4 pooled samples.

§ Average.

|| Range.

\*\* This paper. Values for mouse tumor were: phenylalanine, 3.2 (2.9-3.3); and tyrosine, 2.9 (2.5-3.2).

mouse tumor, determined by any one method, only the average and range of results for each type of sample have been listed in Table I. The data of Mickelson and Barvick(5) has been included for purposes of comparison. The values for rat tumor obtained in this laboratory by microbiological assay and column chromatography are in excellent agreement except for glutamic acid, glycine, and proline which are slightly higher by the chromatographic method. The values determined by paper chromatography differ considerably from those by the other 2 methods but in many cases are of the same order of magnitude.

The small difference in aspartic acid content observed in the microbiological assay data for the 2 types of tissue is confirmed by column chromatography, while that for 6 other amino acids is not confirmed (Table I). The lack of agreement with the results of Mickelson and Barvick(5) may stem from a difference in the method of hydrolysis employed. Although the absolute values differ with the method used, it may be concluded from the results of both paper and column chromatography that there is no significant difference in total amino acid composition between the rat and mouse tumors.

**Summary.** 1. Percentages of total amino acids in viable tissues of the Walker carcinoma 256 (rat), Sarcoma 180 (mouse), and Taylor tumor (mouse) have been determined by both column and paper chroma-

tography. 2. Values obtained by column chromatography agreed satisfactorily with those determined in the same laboratory by microbiological assay while the majority of values determined by paper chromatography were in poor agreement with those of the other two methods. 3. There was no significant difference in the total amino acid content of tumors from rats or mice.

1. Baginsky, M. L., Murphy, E. A., Dunn, M. S., *Proc. Soc. Exp. Biol. and Med.*, 1960, v103, 427.
2. Baginsky, M. L., Thesis, Univ. of Calif., at Los Angeles, 1958.
3. Dunn, M. S., Feaver, E. R., Murphy, E. A., *Cancer Res.*, 1949, v9, 306.
4. Sauberlich, H. E., Baumann, C. A., *ibid.*, 1951, v11, 67.
5. Mickelson, M. N., Barvick, L., *J. Nat. Canc. Inst.*, 1956, v17, 65.
6. Greenstein, J. P., *Biochemistry of Cancer*, Academic Press, Inc., New York, 1954.
7. Roberts, E., Tanaka, T., *Cancer Res.*, 1956, v16, 204.
8. Dunn, M. S., Camien, M. N., Malin, R. B., Murphy, E. A., Reiner, P. J., *Univ. of Calif. Publ. in Physiol.*, 1949, v8, 293.
9. Moore, S., Stein, W. H., *J. Biol. Chem.*, 1951, v192, 663.
10. Ishii, S., *J. Biochem. (Japan)*, 1956, v43, 531.
11. Chinard, F. P., *J. Biol. Chem.*, 1952, v199, 91.
12. McFarren, E. F., Mills, J. A., *Anal. Chem.*, 1952, v24, 650.
13. Baker, B. E., Khan, N. A., *J. Sci. Food Agri.*, 1957, v8, 217.

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### Studies on Measles Viral Hemagglutination.\* (27555)

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The authors have demonstrated(1) the hemagglutinating (Fig. 1) and hemolytic activities of measles virus. Hemagglutination was specifically inhibited by convalescent sera but not by sera from persons with no previous his-

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