position of the nucleoside end, the other to the 5′-phosphate end. The latter chain would have to end with a triphosphate.

Both possibilities of chain growth were at one time considered by Kornberg (*2*), but later the "limited reaction" experiments (*3*) showed that new units are added only to the nucleoside ends of the primer chains. Thus the growth of DNA chains may well be unidirectional, in which case the diagrams in Fig. 1 are erroneous, and Fig. 2 more nearly represents the process.

In consequence of unidirectional chain growth, the formation of replicate double structures is sequential (Fig. 2, left) or semisequential (Fig. 2, right); that is, the formation of one of the new chains, or a different half-length of each new chain, does not begin until the initial double structure has been completely separated. In either case the addition of nucleotides proceeds at twice the rate per chain that would be surmised from the corresponding simultaneous model. Unlike simultaneous models, sequential models predict that up to one-third of the unit DNA is in the single-stranded form at some time in the replication cycle, the observable amount depending on the degree of synchrony among units. If this is true, it may relate to changes in function and in sensitivity to certain agents during the division cycle. If separation of the chains always begins at the same end (for example, where the other end is attached to another structure), then the single chain exposed in successive divisions is always the same member of the complementary pair.

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Seleno-Amino Acid Found in Astragalus bisulcatus

Abstract. Ion-exchange and filter-paper columns were used in a separation of amino acids from an extract of *Astragalus bisulcatus*. Two amino acids were identified, S-methylcysteine and Se-methylselenocysteine.

The identification of naturally occurring selenium compounds has been a subject of investigation since the discovery of selenium in plant material (*1*). Horn and Jones (*2*) reported the isolation from *Astragalus pectinatus* of a crystalline substance containing sulfur and selenium, which analyzed as a complex of 2 parts of selenocystathionine and 1 part of cystathionine.

Table 1. R_F values of S-amino acids, with solvent mixtures A (*3*) (ethanol, 1-butanol, water, and dicyclohexylamine, 10:10:5:2 by volume) and B (formic acid, *tert*-butyl alcohol, and water, 15:70:15 by volume).

Amino acid	$R_F \times 100$	
	Solvent A	Solvent B
Cystine	31.3	9.4
Homocystine	30.5	22
Lanthionine	22.2	8.0
Cystathionine	20.6	12.3
Djenkolic acid	26.6	10.4
Methionine	67.1	67.3
Methionine sulfoxide	37.7	47
S-methylcysteine sulfoxide	41.2	52.3
S-methylcysteine, synthetic	68.2	57.6
Se-compound from *A. bisulcatus*	67.8	58

It was our aim to isolate the selenium compound in *Astragalus bisulcatus* (two-grooved milk vetch) in a manner which would avoid any rearrangements. We used ion-exchange and filter-paper columns for the separation.

The gross separation of the amino acids was made with Amberlite resins. About 80 percent of the selenium was found in the neutral amino acid fraction. Two-dimensional paper chromatograms were made of this fraction of the *A. bisulcatus* extract. By analyzing these paper chromatograms we found that the selenium appeared in only one spot. The spot did not coincide with the sulfur amino acids cystine, homocystine, lanthionine, cystathionine, djenkolic acid, methionine, methionine sulfoxide, and S-methylcysteine sulfoxide (see Table 1). The unknown selenium spot from *A. bisulcatus* had the same R_F values as S-methylcysteine and its selenium analog. In fact, when *A. bisulcatus* extract was chromatographed with either of these acids the same amino acid map was obtained.

The *A. bisulcatus* used for this work was collected in Wyoming and Montana. Dried leaves and stems were ground in a Wiley mill; they analyzed 0.1- to 0.3-percent selenium. A 10-percent water extract was made with isopropyl alcohol as preservative. The neutral amino acids were separated from most of the other compounds by the use of the Amberlite resins IR-4B, buffered IRC-50, and IR-120 (acid form). Absorbing these neutral amino acids on Dowex 50×4 (200 to 400 mesh) and eluting with $0.01M$ NH_3 gave a partial separation into a sulfur and a selenium fraction. The sulfur fraction contained proline and small amounts of other amino acids, but no selenium. The selenium fraction contained some sulfur amino acid, alanine, valine, proline, glycine, and all of the selenium compound. These two fractions were treated separately to isolate the S- and Se-amino acids.

The sulfur fraction was passed through filter-paper columns, with solvent mixtures A (*3*) (see Table 1) and C (formic acid, *tert*-butyl alcohol, acetone, and water, 5:40:40:15 by volume). The portions containing only the sulfur compound were pooled, flash evaporated, and crystallized several times from ethanol. The infrared spectrum for this crystalline sulfur amino acid was identical with that of synthetic S-methylcysteine (*4*). The analysis: calc. for $C_4H_9O_2NS$: C, 35.54; H, 6.71; N, 10.36; S, 23.72. Found: C, 35.80; H, 6.42; N, 10.10; S, 23.55. The agreement of analysis, infrared spectra, and R_F values indicates that the sulfur amino acid in *A. bisulcatus* is S-methylcysteine.

The selenium amino acid fraction, when subjected to the same procedures, yielded a crystalline solid which analyzed for 38.5 percent selenium (calc. for Se-methylselenocysteine: 43.37). The infrared spectrum of the crystalline selenium compound was very close to but not exactly the same as that of pure synthetic Se-methylselenocysteine and seemed to indicate the presence of both S-methylcysteine and Se-methylselenocysteine (*4*). The R_F values with solvent mixtures A and B of synthetic Se-methylselenocysteine and of the natural seleno-amino acid are identical and are also identical with those of S-methylcysteine; they differ from those of methionine and selenomethionine. The results seem to indicate that the seleno-amino acid found in *A. bisulcatus* is Se-methylselenocysteine, at this stage of the work not yet completely separated from S-methylcysteine (*5*).

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* Deceased.

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