

All published spectra of isolated lignins appear to contain the 1,710 cm^{-1} absorption band ('appear to' because not all have been printed with adequate clarity for comparison). The band is also present in Freudenberg's dehydrogenation polymers, and Freudenberg and co-workers⁹ have pointed out that the strength of the band is influenced by the method of preparation of the polymer.

These observations are consistent with the conclusion that an acetal or hemi-acetal bond exists between the carbonyl groups of lignin and the hydroxyl groups of some portion of the holocellulose. This was first suggested as a hypothesis by Holmberg in 1925¹⁰, and is supported by the results of a number of chemical investigations¹¹. The work reported here is the first evidence obtained by purely physical means in such a way that the wood and pulp lignins were observed without having been modified by chemical reactions.

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¹¹ For example, ref. 1, 458, 616, 645, 646, 649, 650, 652.

BIOCHEMISTRY

Chemical Characterization of Kinins of Human Plasma

By the method of ammonium sulphate fractionation, human plasma can be separated into a part *A*, which is insoluble at 33 per cent saturation, and a part *B*, precipitated between 33 and 45 per cent saturation. When these precipitates are dissolved in water, both the solutions spontaneously develop the ability to cause contraction of the isolated rat uterus, apparently due to the liberation of plasma kinins. From the second preparation (precipitate *B*) the well-characterized kinins bradykinin and kallidin have already been isolated^{1,2}. We have now undertaken detailed investigations of the neutral '33 per cent precipitate', *A*, with results which may be summarized as follows.

(1) The ammonium sulphate precipitate (*A*) contains a substance, apparently a proteolytic enzyme, which has been isolated (by a method to be described), and which liberates a kinin from 'preactive' human plasma³. This kinin can be distinguished from both bradykinin and kallidin and has been provisionally called kinin *E*.

(2) When the ammonium sulphate precipitate (*A*) is allowed to activate spontaneously in neutral solution, two kinins can be obtained which are different from either kinin *E*, bradykinin or kallidin. These have been referred to as kinins *F* and *S*.

These substances have been classed as plasma kinins on the grounds that they can bring about contractions of the isolated rat uterus, cause pain when applied to the exposed blister base, are unstable in plasma and are digested by pancreatic enzymes. All five compounds can be separated either by chromatography or by electrophoresis on paper. For each method the paper has been washed with 2 mM 'Versene' solution and then with water, in order to reduce adsorption of the peptides. Despite this precaution, however, substances *E* and *F* are often 'lost' during the separation, though whether by adsorption on the support or through destruction by the solvent is not yet clear.

Table 1

Solvent	<i>F</i>	<i>E</i>	<i>S</i>	Kall.	Brad.
BuOH/AcOH/H ₂ O	12	19	23	23	26
BuOH/Pyrr./H ₂ O	Decomposed (?)	16	13	6	10

The distance moved (cm) after 18 h chromatography of five plasma kinins on Whatman No. 1 paper washed for 24 h in 2 mM 'Versene' followed by water for 48 h. The strips were cut into 1-cm segments and eluted with Ringer's solution. The eluate was then tested for its ability to contract the isolated rat uterus. Crystalline synthetic bradykinin and kallidin were used.

Paper electrophoresis is particularly useful for the separation of bradykinin and kallidin from substance *S*, which migrates the fastest of the three (thus, at a field strength of 100 V/cm in 2 N acetic acid, *S* moved 24 cm; kallidin, 16 cm; bradykinin, 7 cm in 30 min). Substances *E* and *F* can also be resolved by this method; for example, in pH 7.4 pyridine/acetic acid buffer, at 100 V/cm, *E* moved 2 cm; kallidin, 4 cm; and *F*, 6 cm in 10 min, but after 30 min substances *E* and *F* could not be detected. In all cases migration was cathodic.

The chromatographic behaviour of all five kinins is summarized in Table 1, from which it appears that substance *S* and kallidin can be resolved in butanol/pyridine/water (1 : 1 : 1), but not in butanol/acetic acid/water (4 : 1 : 5). However, the latter solvent is the better for the identification of compounds *F* and *E*, since it brings about their inactivation at a slower rate.

From these results it appears that human plasma has a potential for the endogenous formation of several kinins. Moreover, since the isolated enzyme of precipitate *A* produces kinin *E* when added to human plasma, it cannot be plasma kallikrein, which forms bradykinin under these conditions². Furthermore, substances *E* and *F* are formed at a rapid rate, whereas substance *S* reaches its maximum concentration only after about 3 h. Thus it is unlikely that substance *S* appears in reactions that produce 'kinin' rapidly; but it may be an important product of systems, such as those of acid or acetone-treated plasma⁴⁻⁷, which react relatively slowly.

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Blood- and Serum-levels of Water-soluble Vitamins in Man and Animals

VITAMIN-LEVELS in human blood and serum have recently been reported¹. Except for cyanocobalamin (vitamin B₁₂)², the titre of vitamins of the B-group in animal bloods and sera has not been documented. Comparative values would be an aid in determining the influence of these vitamins as animal growth factors, and also for evaluating their influence when these fluids are included in microbial and tissue culture growth media. Here we report the vitamin titre of freshly drawn blood and serum of cattle, sheep, chicken, rabbit and horse, the biological fluids of which are commonly used for experimental work. Eight pooled samples from each species were obtained from Cappel Laboratories of West Chester, Penn. These fluids were assayed for: (a) thiamine with *Ochromonas malhamensis*³; (b) pantothenic acid with *Lactobacillus plantarum*⁴; (c) nicotinic acid and amide with *Tetrahymena pyriformis*⁵; (d) B₆ group (pyridoxine, pyridoxal, pyridoxamine) with *T. pyriformis*⁶; (e) uncon-