for human erythrocytes could not be demonstrated in these serums by the usual methods.

(2) Among 41 samples of anti-pneumococcic horse serums of types other than type XIV, only two agglutinated human erythrocytes in a dilution of 1:20 or higher. One concentrated bivalent type V and VII horse serum and one unconcentrated type IX horse serum agglutinated human group B erythrocytes in dilutions of 1:40 and 1:20, respectively. These forty-one serums included at least one specimen of serum for each of the twenty-nine other available specific pneumococcus types. Sixteen of them were concentrated preparations.

(3) Ten samples of different type XIV anti-pneumococcic rabbit serums were obtained from the same three laboratories as supplied the horse serum. These rabbit serums were prepared with the same strains of type XIV pneumococci as were used in making the type XIV horse serum. None of these rabbit serums agglutinated human group B or group O cells (1:5 dilution was the lowest tested), but four agglutinated both AB and A human erythrocytes in dilutions of 1:20 to 1:80. Among twenty rabbit anti-pneumococcic serums of eleven types other than type XIV, eight showed agglutinins for human AB and A in titers of 1:10 to 1:160, but none agglutinated either group B or group O cells. The agglutinins for groups AB and A human erythrocytes in the various anti-pneumococcic rabbit serums, both type XIV and those of other types, were associated with agglutinins and hemolysins for sheep erythrocytes and could be absorbed with sheep red blood cells.

(4) After sufficient absorption with type XIV pneumococci to remove the homologous pneumococcus agglutinins, no agglutinins could be demonstrated, in the type XIV anti-pneumococcus horse serums, for human erythrocytes of any of the four blood groups. Absorption with human erythrocytes of each of the four groups completely removed the agglutinins for human red blood cells of the homologous and of the three heterologous blood groups, but left the type XIV pneumococcus agglutinins essentially unchanged. Large amounts of erythrocytes were necessary to carry out these absorptions.

The details of these observations and of further experimental studies will be reported elsewhere.

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PREPARATION OF PURE D-ARGININE

ALTHOUGH the best method for preparation of pure arginine derivatives appears to be through the flavianate isolation, as introduced by Kossel and Gross,[1] and subsequently improved[2, 3] to give a high-purity arginine hydrochloride, none of these papers presents details for the obtainment of free base d-arginine. Another article[4] has described an unsuccessfull attempt to convert the hydrochloride into pure arginine base.

The essential precautions for obtaining pure d-arginine from the hydrochloride now appear to be the choice of a satisfactory protein source, and the removal of arginine-silver complex from the final free arginine solution, since the arginine-silver complex is soluble in solutions of arginine.[5] Satisfactory protein sources include salmine and gelatine of U. S. P. grade or better. Casein, hog's blood and defatted canned sardine spermatic tissue were found to be unsatisfactory.

For the preparation of d-argininium chloride, the classical method of Brand and Sandberg[3] was followed, except that it was imperative to dilute 39 ml of 5 N HCl in 300 ml of water in hydrolyzing 50 gm batches of benzylidene arginine. For the hydrolysis of commercial salmine, seven parts of concentrated HCl (S.G. 1.19) sufficed.

In an exemplary preparation of free base d-arginine, 8.03 gm of silver nitrate (3 per cent. excess over the arginine hydrochloride requirement) was dissolved in 50 ml of water and treated with 50 ml of 2N NaOH. The precipitated silver oxide was washed until the washings were neutral, and the silver oxide was then transferred to a solution of 9.66 gm of arginine hydrochloride in 50 ml of water, with aid of the wash-bottle. The mixture was stirred mechanically for ten minutes, and the silver chloride filtered off. The filtrate was saturated with hydrogen sulfide, boiled, and the coagulated silver sulfide filtered off. The filtrate from this operation was evaporated to dryness under reduced pressure on the water bath in a stream of carbon dioxide-free air. The residue was dissolved in 20 ml of boiling water, and placed in a desiccator over sodium hydroxide. When the solution had cooled, the desiccator was evacuated. The yield was 7.84 gm of arginine, melting at 228° (corr.) with dec. This corresponds to 98 per cent. recovery from the hydrochloride. Recrystallization from 16 ml of hot water and 40 ml of freshly boiled absolute ethanol gave a 96 per cent. recovery.

No difficulty was experienced in crystallizing arginine prepared from salmine or gelatine. One crystal of 3 cm length was obtained by slow evaporation of the solvent during two weeks. Arginine solutions

[1] A. Kossel and R. E. Gross, *Zeit. Physiol. Chem.*, 135: 167, 1924.

[2] P. Brigl, R. Held and K. Hartung, *Zeit. Physiol. Chem.*, 173: 151, 1928.

[3] E. Brand and M. Sandberg in F. C. Whitmore, "Organic Syntheses," 12: 4, 1932.

[4] E. Schulze and E. Steiger, *Zeit. Physiol. Chem.*, 11: 43, 1887.

[5] W. Gulewitsch, *Zeit. Physiol. Chem.*, 27: 178, 1899.

from the other sources mentioned above sometimes required addition of carbon dioxide-free ethanol in order to cause crystallization.

Arginine absorbs carbon dioxide from the air, but this may be driven off by boiling the solution of recrystallization.

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FAILURE OF ALFALFA TO PREVENT THE HEMORRHAGIC SWEETCLOVER DISEASE[1]

IN 1922, Schofield[2] showed that the feeding of poorly cured sweetclover hay may induce in the bovine a disease characterized by a diminished clotting power of the blood. The disturbance of the clotting mechanism has since been found to be due to a deficiency in prothrombin.[3,4] Continued administration of a toxic diet results in severe, usually fatal hemorrhage.

In a recent paper on one phase of this problem, Quick[5] concludes from experiments with rabbits that "Diet is the important means for controlling the disease. The incorporation of 5 per cent. of dehydrated alfalfa meal with the toxic hay was found sufficient to prevent the development of the disease or even any demonstrable reduction of prothrombin." It is also stated that "The animal appears to be able to store this accessory factor [from alfalfa], for it is very difficult to produce sweetclover disease in animals that have been fed relatively large amounts of alfalfa. This explains why some animals are far more resistant than others to the same lot of spoiled hay." Quick points out the practical significance of these conclusions and suggests the relation of this accessory factor to the anti-hemorrhagic vitamin K which is required by the chick for normal blood coagulation and is present in alfalfa. These conclusions, of significance in agriculture and in studies of factors affecting the mechanism of blood coagulation, are not in agreement with the results of our experiments with rabbits. The details of these experiments will be recorded elsewhere, but the salient features will be presented at this time.

We have found no indication that alfalfa exerts a protective action against the sweetclover disease. Rabbits have been fed alfalfa to the amount of 50 per cent. of the diet along with toxic hay and toxic extracts. Nevertheless the symptoms of the disease have appeared. Freshly cut alfalfa constituting 12.9 per cent. (dry weight) of the ration and kiln-dried alfalfa constituting 10 per cent. each in a diet of toxic hay failed to inhibit the action of the toxic principle. Likewise a commercial alfalfa hay of excellent quality when incorporated to the amount of 10 per cent. in a toxic hay was ineffective in checking the onset and fatal termination of the disease.

A marked variation within a group of rabbits of similar age in reaction to a given toxic hay has been observed in our experiments. Animals found susceptible in a preliminary test on a uniform diet have maintained their susceptibility in further subjection to the action of the toxic principle. In like manner resistant animals in as far as they have been tested have been resistant in later trials on the same hay. Furthermore, no significant differences have been observed between groups of rabbits having had alfalfa or no alfalfa during the period prior to a test on toxic hay. These and related considerations arising from experiments on some 150 rabbits suggest that the variation in a group of rabbits in reaction to a given toxic hay is due to the inherent characteristics of the animals; there is no evidence in our experiments that the variation results from previous feeding.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

STATIC ELECTRIC PROPERTIES OF A NEW BAKELITE PLASTIC

RECENTLY the Bakelite Corporation has put on the market a new plastic designated as polystyrene XMS–10023. It was described as an excellent electric insulator. In the search for some material that could be used as insulator in static electric experiments the writers tested this material by electrometric methods. While amber has all the desirable properties for an insulator it is rather expensive and, particularly, not easily obtainable in larger dimensions. XMS–10023 can be molded readily into any given shape without restriction as to size.

The electric resistivity was compared to that of amber and of a shellac-coated hard rubber, with the approximate relative results shown in the following table:

[1] Contribution from the Department of Genetics (Paper No. 226), Wisconsin Agricultural Experiment Station in cooperation with the Division of Forage Crops and Diseases, Bureau of Plant Industry, U. S. Department of Agriculture. Cooperative investigations with the Biochemistry Research Laboratory, Department of Agricultural Chemistry, Wisconsin Agricultural Experiment Station.

[2] F. W. Schofield, *Can. Vet. Record*, 3: 74–78, 1922.

[3] L. M. Roderick, *Am. Jour. Physiol.*, 96: 413–425, 1931.

[4] A. J. Quick, *Jour. Biol. Chem. Proc.*, 114: lxxxii, 1936.

[5] A. J. Quick, *Am. Jour. Physiol.*, 118: 260–271, 1937.