in order to determine the direct effect of the drug upon the dehydrogenase activity of resting cells of the parent and drug-fast strains. These results are shown in Table II.

Sulfapyridine, in a final concentration of 1:8,000 in the reacting system, does not inhibit the glucose dehydrogenase of either strain. However, the same concentration of the drug greatly inhibits the dehydrogenase-activity of the parent cells for glycerol, lactate, and pyruvate.

The relation of peroxide formation to the carbohydrate metabolism of pneumococcus is not entirely clear. However, suspensions of the parent cells incubated in a shallow layer in the presence of glycerol and coenzyme produce much more peroxide than if the glycerol is replaced by an equal concentration of glucose, indicating that hydrogen peroxide is produced during the metabolism of glycerol.

*Summary.* The acquisition of "sulfapyridine-fastness" by a strain of pneumococcus Type I is associated with a marked diminution in the production of hydrogen peroxide in cultures of this strain. Cell suspensions of the parent and drug-fast strains dehydrogenate glucose equally well. On the other hand, the acquisition of sulfapyridine-fastness is associated with a marked loss of dehydrogenase-activity for certain 3-carbon compounds (glycerol, lactate, and pyruvate). When sulfapyridine is added directly to the reacting system the dehydrogenase-activity of the parent cells for the same 3-carbon compounds is likewise much decreased.

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## Specific Absorption of Antibody with Extracts Containing the Rabbit Papilloma Virus (Shope).

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The papilloma virus, or a substance that has proved inseparable from it, reacts with specific immune sera to fix complement, as previous studies have shown;[1] and tests made recently with more than a score of such sera have borne out a finding already obtained, namely that the virus-neutralizing ability of any serum is directly proportional to its complement-fixing capacity. The present work was

[1] Kidd, John G., *J. Exp. Med.*, 1938, **68**, 703, 725, 737.

undertaken to learn whether the virus-neutralizing and complement-fixing antibodies can be absorbed from the immune sera; and, if so, whether they are absorbed together, and what is responsible for their removal.

A number of immune sera have been used, some obtained from cottontail rabbits with naturally-occurring growths and others from wild and domestic rabbits with experimental papillomatosis. Various dilutions of the sera were made with saline, from 1:2 to 1:64 or higher, and these were mixed in equal parts with clear, saline extracts of the virus-induced growths, either as such or after Berkefeld filtration, in dilutions up to 1:640. The mixtures were put into the water-bath at 37°C for 2 hours, then kept overnight in the refrigerator. The relative amount and character of the specific visible flocculation, which was regularly present in the tubes containing antigen and antibody in optimal proportions, were then recorded, and the mixtures were spun at 4400 rpm for 20 minutes in an International centrifuge with 51°-angle head. The supernatant fluids thus obtained were now tested for content of virus, complement-fixing antigen and antibody, by means of standard pathogenicity, neutralization, and complement-fixation tests which have been already described.[2]

In every one of 7 experiments, in which extracts containing the virus were mixed with immune sera in optimal proportions, tests of the sort described showed that absorption of antibody had occurred; and the complement-fixing and virus-neutralizing capacities of the sera were always proportionately reduced. After the mixtures had been centrifugalized, the supernatant fluids were regularly found to be neutral when optimal or near optimal proportions of antigen and antibody had been used; that is to say, they contained no detectable amounts of virus, antigen, or antibody, as manifested by the *in vivo* and *in vitro* tests. When an excess of antigen or antibody had been used, on the other hand, a proportionate excess of the one or the other remained in the supernatant fluids after absorption and centrifugation; and whenever there was a large excess of either no visible flocculation took place in the mixtures. It was observed that a given amount of antibody could be absorbed completely by very much less of a virus-filtrate than it would neutralize, and that the amount of a virus-filtrate required to absorb antibody was even less than that required to fix 2 units of complement.

The findings thus far indicate that the virus itself is involved in the antibody-absorption. Filtered extracts of the naturally-occurring

[2] Kidd, John G., Beard, J. W., and Rous, P., *J. Exp. Med.*, 1936, **64**, 63, 79; Kidd, John G., *J. Exp. Med.*, 1938, **68**, 703, 725, 737.

papillomas of some cottontails regularly contained much of the virus, and these always absorbed antibody readily and in great amount. Extracts or filtrates of the virus-induced papillomas of other cottontails, which contained much less of the virus as indicated by their infective titer, had much less power to absorb antibody; and still others, made from the experimental growths of cottontail and domestic rabbits, which yielded no pathogenic virus, failed completely to absorb antibody in the tests, even when large amounts were used repeatedly. Extracts of the Brown-Pearce tumor, tested concurrently, had no power to absorb the antibody.

When a potent virus filtrate was spun at 30,000 rpm for 60 minutes in the air-driven centrifuge, the supernatant fluid—which contained practically none of the virus, but much protein as determined roughly by the sulphosalicylic acid test—proved devoid of capacity to absorb antibody; whereas a suspension of the pellet of sediment in the original volume of saline—which contained little protein in comparison with the supernatant fluid, but almost as much virus as the whole filtrate—absorbed antibody quite as well as the latter.

A centrifugalized virus filtrate that absorbed much antibody when used unheated or after heating at 56°C for 30 minutes, failed to do so after it had been heated at 66°C for 30 minutes, a procedure that abolished the capacity of the filtrate to fix complement and rendered it completely non-infectious.

The findings make it plain that the complement-fixing and virus-neutralizing antibodies can be absorbed together, and readily, from the sera of rabbits bearing virus-induced papillomas, when these are mixed with extracts or filtrates containing the papilloma virus; and they indicate that union of the antibody with the virus itself, or an integral part of it, is responsible for the absorption. The significance of these results, notably in relation to the findings of Salaman in similar absorption experiments with the elementary bodies and soluble antigens of vaccinia,[3] will be discussed later when the facts are reported in detail.

[3] Salaman, M. H., *Brit. J. Exp. Path.*, 1937, **18**, 245; 1938, **19**, 192.