CXXXI. THE FOURTH COMPONENT OF COMPLEMENT AND ITS RELATION TO OPSONIN.

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THE question of the nature of opsonins in normal serum has engaged the attention of many workers during the last twenty years. Broadly speaking, there appear to be two schools of thought. Many workers, including Neufeld and Hühne [1907] and Levaditi and Inmann [1907], consider that opsonin is intimately related to, if not identical with, the haemolytic complement in serum, whilst others, including Wright and Douglas [1904], believe that opsonins are distinct bodies not identical with complement or anti-bodies. Briefly, the evidence in favour of the first view is that processes that absorb complement from serum generally remove the opsonic power. Thus any system such as bacteria, serum or red cells plus the immune body will take up complement from a normal serum, and the serum at the same time loses its opsonic action. Browning [1925], however, shows that bacteria may remove most of the opsonins from serum and still leave a considerable amount of haemolytic complement. This he explains either by the existence of a natural immune body which combines with the organism and leads to the fixation of a definite amount of complement or to the existence of multiple complements, some of which only are concerned with opsonic action. The effect of heat upon the opsonic and complementary properties of serum seems in favour of the identity of the two properties; both are definitely destroyed at 56°.

It seemed to us that some advance would be made if the question of the identity or non-identity of complement and opsonin could be settled. In the first place it must be pointed out that even if some process, which removes complement activity from serum, removes opsonic activity at the same time, it is not necessarily proved that the two substances are one and the same.

For since complement involves several factors, all of which must be present to bring about haemolysis, opsonin may be similarly complex and we may destroy one factor which happens to be common to both. Thus the evidence mentioned above must be taken to indicate that one factor of complement at least is also concerned in opsonic action; other complement

factors may not be concerned. If complement could be inactivated by destroying one factor and the opsonic power of the serum remained unimpaired then the non-identity of the two properties or substances would be established. In a foregoing paper we have described a simple and easily applied method for destroying or inactivating a relatively heat-stable component of complement and it was considered of interest to determine whether this factor had any connection with opsonic action.

Serum was treated with one-quarter of its volume of $N/6.5$ NH_4OH and incubated for various times ranging from half-an-hour to 2 hours. It was neutralised with acid and then tested for complementary and opsonic power against a control which had been incubated for the same time and diluted to the same extent with normal saline. The opsonic tests were carried out by mixing equal volumes of serum, suspension of leucocytes and suspension of organisms, and incubating the mixture for half-an-hour at 37° . Slides were stained and the presence or absence of phagocytosis was determined. The experiments were performed with rabbit and guinea-pig serum, the respective leucocytes and with staphylococci and tubercle bacilli. Controls with washed leucocytes and bacteria alone showed practically no phagocytosis.

The treatment with ammonia described above never failed to give a serum with no complement activity, due simply to lack of the fourth component, and on the other hand such serum proved to have lost none of its opsonin. In no experiment could it be distinguished from the control of unheated serum so far as phagocytosis was concerned.

Thus it is definitely established that while complement and opsonin may be closely related the whole complement system is not essential for opsonic action.

SUMMARY.

Serum in which the fourth component of complement had been inactivated by means of ammonia proved to have lost none of its opsonic activity. The fourth component of complement therefore plays no part in opsonic action.

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