longer than 48 hr. This solution, diluted two-, four-, eight- and sixteen-fold and mixed with blood in the same proportion by volume, delayed coagulation for 20, 13, 10 and 8 min., respectively, but normal clotting took place in the presence of a × 32 dilution. Table 1 shows the coagulation times and corresponding approximate quantities of the extract. The substance extracted from the granules gave a metachromatic reaction with subsequent precipitation with toluidine blue. These observations strongly suggest a resemblance of the extracted material to heparin.

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Specificity of Complete Cold Auto-Agglutinins

Cold and warm auto-agglutinins exist. Both occur in the complete and incomplete forms. They are generally believed to be non-specific. Recently, doubt has been cast on their lack of specificity. I consider that (complete) auto-agglutinins might consist of multiple specific components[1-3]. Recently, Crawford, Cutbush and Mollison[4] have shown that incomplete cold auto-agglutinins present in all human sera have the specificity anti-*H*. The degree of sensitization of cells of various groups by these antibodies could be arranged in the following descending order, O—A_2—B—A_1—A_1B. Such reactions correspond to the relative amounts of *H* present in these various cells, *O* cells containing the most *H* and A_1B cells the least. Further, the action of incomplete cold auto-antibodies was inhibited by *H* substance, and their reactions closely agreed with those of an eel anti-*H* serum. Absorption of the antibodies in the cold by cells of various groups revealed that if A_1B cells are used the amount of antibody absorbed is inappreciable, whereas if *O* cells are used ability to sensitize group *O* cells or cells of groups A_1 and A_2 was completely abolished. These workers consider that the specificity of other non-specific antibodies needs to be re-examined. Davidsohn and Oyamada[5] have shown that the warm auto-agglutinins associated with hæmolytic anæmias are predominantly specific for the individual's own cells. Weiner *et al.*[6] have demonstrated that such an auto-agglutinin had the specificity anti-*e*.

The complete cold auto-agglutinins which are present in almost all Indian bloods were therefore re-investigated. For convenience *AB* bloods were examined as they do not contain iso-agglutinins. The sera were titrated against cells of groups A_1B, A_2B, A_1, A_2, *B* and *O*, and in each case against the patient's own cells. Typical results are reproduced in Table 1.

It is evident that the strengths of agglutination do not correspond to the expected amount of *H* agglutinogen in the various cells, and that these antibodies are not predominantly specific for the individual's own cells.

Table 1. Titres of Cold Agglutinins in an A_1B Serum against Various Cells at 4–6° C.

Cells	Activity of undiluted serum	Titre
A_1B	+++	8
A_2B	W	2
A_1	+++	8
A_2	+++	16
B	+++	128
O	+++	16
Own	+++	32

The antibodies did not give parallel reactions with an anti-*H* serum prepared by absorbing the serum of the Indian water buffalo with A_1B cells (water buffalo anti-*H* is more powerful than that prepared from Indian cattle). *H* substance (dried saliva of a group *O Le* (*a-b-*) secretor) produced some inhibition of the action of these antibodies on various cells. Similar inhibition was produced by the saliva of a group *O Le* (*a-b-*) non-secretor. The secretor saliva successfully inhibited the water buffalo anti-*H* but the non-secretor saliva did not. In some sera complete absorption with *O* cells failed to abolish activity upon other cells, and similar findings in certain other sera were obtained after absorption with the individual's own cells. Elution of the antibody from the cells used for the absorption gave inconclusive results.

The titration and absorption experiments indicate that the complete cold auto-agglutinins present in the sera of most Indians do not give the reactions expected of anti-*H*. This view is supported by the examination of the titration values of these cold agglutinins in a number of sera undertaken in connexion with another investigation (to be published). In this investigation complete cold agglutinins in the sera of Indians of all groups were titrated in parallel against *O* cells, compatible *A* or *B* cells and the individual's own cells. The results are summarized in Table 2.

Table 2

1st Series

Number of sera examined	100
Number of sera in which titres against *O* cells were higher than the individual's own	74
Number of sera in which the individual's own cells were agglutinated to higher titres than *O* cells	17
Number of sera showing equal activity against both *O* cells and the individual's own	6

2nd Series

Number of sera examined	81
Number showing highest titres against *O* cells	41
Number showing highest titres against compatible *A* or *B* cells	14
Number showing greatest activity against the individual's own cells	10
Number showing equal activity against all cells	16

While it appears that complete cold auto-agglutinins do not have the specificity anti-*H*, the possibility of anti-*H* being a component antibody cannot be excluded.

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