suspensions or the presence of some factor in rumen fluid that facilitates hydrogenation and explains the previous conclusions regarding the role of bacteria in ruminant hydrogenation. However, these results show there can be no doubt that rumen bacteria are capable of extensive hydrogenation, and emphasize the caution needed in the interpretation of results obtained under conditions far removed from those found *in vivo*.

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HÆMATOLOGY

Red Cell Agglutination by Stromal Material in Hæmolysates

DURING the preparation of hæmoglobin solutions a phenomenon was observed which, so far as is known, has not previously been described. The red cells of some species of animals are agglutinated by hæmolysates made from the cells of the same species. The degree of agglutination is sufficiently strong to be easily visible to the naked eye when tested on a slide.

Hæmolysates were prepared from the cells of a number of different animals by subjecting a 20 per cent suspension of washed cells to ultrasonic vibration. After centrifuging for 20 min. at 3,000 rev./min. to remove the larger particles of stroma, it was tested against washed cells of the various species. The results are shown in Table 1, where the degree of agglutination is recorded, following the usual practice of blood transfusion work[1]. From Table 1 it can be seen that the agglutination seems to be a property of the hæmolysate rather than of the intact cell; and that it is confined to the three rodent species.

Table 1

Cells of	Hæmolysates of cells of: Sheep	Rabbit	Dog	Horse	Rat	Guinea pig
Sheep	0	+	0	0	0	+
Rabbit	0	+++	0	0	++++	++
Dog	0	0	0	0	+	++
Horse	0	++++	0	0	0	0
Rat	0	+	0	0	+++	+
Guinea pig	0	0	0	0	+	+

Group *A* and *O* human cells were not agglutinated by any hæmolysate and hæmolysates of human cells of groups *A* and *O* caused no agglutination of cells of any species.

If packed washed rabbit cells are frozen and thawed with the minimum of disturbance a jelly-like material with the consistency of egg-white is produced. This material, which is probably stromatin[2], can be filtered off and washed. The filtered hæmoglobin, although very concentrated, produces no agglutination of intact red cells. On the other hand, the washed residue, although not entirely free of hæmoglobin, is very active in producing agglutination when dissolved in saline. This property can be removed by absorption with rabbit cells but is unaffected by treatment with fat solvents.

The properties of the hæmolysates are dependent on the method of preparation. Fresh washed rabbit cells were lysed: (1) by ultrasonic vibration; (2) by the addition of distilled water followed by the calculated quantity of 10 per cent saline to make the solution isotonic and (3) by freezing. The hæmolysates were handled with care to avoid shaking. On completion of lysis all three preparations produced a similar degree of agglutination. After centrifuging at 96,000*g* for 1 hr. it was found that the supernatant fluids varied considerably in their ability to agglutinate washed cells. The deposit was re-suspended in saline by shaking vigorously for at least 5 min. before centrifuging at 3,000 rev./min. for ½ hr. The results of testing the supernatant fluid and the deposit against whole cells are summarized in Table 2.

Table 2

Method of production of hæmolysate	Hæmolysate: Deposit	Supernatant
1. Ultrasonic vibration	+	+++
2. Distilled water	+++	+
3. Freezing	+++	0

The probable explanation of these findings is that the more violent method of disruption of the cells also disintegrates the red cell envelope and stroma, some product of which is added to the solution and not removed by high-speed centrifugation for 1 hr. The gentler methods, however, leave this substance attached to the 'cell ghosts' which are found in the deposit.

We can conclude, therefore, that the agglutination of homologous and some heterologous red cells by hæmolysates of rabbit, rat and guinea pig red cells is due to a substance liberated from the stroma. The amount of this substance in the solution is determined by the method of lysis and the amount of shaking during preparation. This variation in composition may explain the divergent results obtained by the earlier workers in experimental hæmoglobinuric nephrosis.

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Asymmetrical Recombination of Alkali-dissociated Hæmoglobin Mixtures

THE human adult hæmoglobins and canine hæmoglobin recombine asymmetrically, that is:

$$\alpha_2{}^X\beta_2{}^X + \alpha_2{}^Y\beta_2{}^Y = \alpha_2{}^X\beta_2{}^Y + \alpha_2{}^Y\beta_2{}^X$$

after both short and prolonged exposure to dilute acid[1–5]. At *p*H 11, hæmoglobin *A* dissociates into half-molecules[6]; furthermore, re-association of a mixture of labelled hæmoglobin *S* and hæmoglobin *A* after 24 hr. at *p*H 11 results in the exchange of the β_2 sub-units[7]. We now wish to report a series of experiments which indicate that hæmoglobin also recombines asymmetrically after short exposure to alkali.

In the first experiment a 2 per cent carbonmonoxy-hæmoglobin solution (all carbonmonoxyhæmoglobin