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The advantage for certain experiments *in vitro* of suspending trypanosomes in serum.

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The amount of trypanocidal substance contained in a given solution is sometimes estimated *in vitro* by determining the degree to which the solution may be diluted before it ceases to immobilize the parasites.

For want of a better diluent, trypanosomes have usually been suspended in physiological salt solution (with or without the addition of citrate), although salt solution not infrequently immobilized the parasites in 30 to 60 minutes.

In 1910, the writer made observations which caused him to substitute serum for the salt solution he had previously employed in suspending *T. brucei*. Serum (when not bound by the medicament under investigation) had the following advantages:

1. The motility of the control parasites was greatly prolonged. This enabled the observations to be continued over a longer time.

2. The motility of the control parasites was accelerated. This enhanced the delicacy of the tests *in vitro* by rendering more striking the contrast between the poisoned and the non-poisoned trypanosomes.

It was found, moreover, that poisons not infrequently immobilized more quickly trypanosomes suspended in serum than they did those suspended in salt solution. This also seemed to give serum a slight advantage over salt solution as a medium in which to suspend trypanosomes.

Rabbit, ox, horse, goat, sheep, pig, chicken, rat, and mouse sera were tested and were found to be efficient in prolonging the motility of trypanosomes.

It soon became easy to keep on hand a large supply of serum, for experiment showed that cattle serum, filtered through a Berkefeld filter, bottled aseptically, and preserved in the ice-box, retained its activity for many months.

That the motility of trypanosomes is preserved longer in serum than in salt solution was noted years ago and has recently been emphasized by Schern,[1] but the writer is not aware that anyone has previously recommended suspending trypanosomes in serum for experiments *in vitro*.

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The action of atoxyl.

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The action of atoxyl is paradoxical. *In vivo* it is effective against certain parasites, *in vitro* it has little or no action. Ehrlich believes that the medicament must be reduced in the body before it becomes active. This view has been strengthened by Levaditi and Yamanouchi who have shown that emulsions of liver, muscle, and lung, when incubated with atoxyl, transform this medicament into a toxic substance. Levaditi apparently believes that the transforming agent is in the liver and other organs, while Yamanouchi concludes that it is in the red blood cells only. My results confirm much of the experimental work of Levaditi and Yamanouchi, but lead to a conclusion that, in its entirety, is apparently held by neither of these investigators.

In my experiments, both liver and blood when incubated with atoxyl (10 per cent.) at 37 degrees for 3 hours, transformed this medicament into a toxic substance.

The transforming agent in liver had characteristics, however, which in some respects were quite different from those of the active agent in blood.

The active agent in liver was soluble in salt solution, was filterable through collodium, and was quite resistant. Liver emulsion ground with sand in a mortar, or heated to 100 degrees for 10 m., lost little or none of its activity. The addition of blood to liver emulsion before incubation with atoxyl increased its activity, but liver emulsion washed *thoroughly* to free it of red blood corpuscles was inactive, probably because of the solubility of the transforming agent.

From the blood the active agent was apparently not extract-

[1] "Arbeiten aus dem kaiserlichen Gesundheitsamte," Berlin, 1911, xxxviii, 338.