TABLE 1. Impurities found in gallium.

Impurity	(A) As purchased	(B) After a single growth cycle	(C) After two growth cycles
Mg	TR	VW*	TR
Sn	W	TR-VW	VFTR
Cu	W	VW	TR
Pb	W	VW	TR
Hg	TR	FTR	VFTR
Cd	TR	FTR	
Si	TR	VW*	
Al		FTR*	
Mo	TR		
Ge	TR		
Zn	FTR		
Ca	TR		
Ag	VW		

* See discussion.
Interpretation of symbols: W, 0.1–0.01%; VW, 0.01–0.001%; TR, 0.001–0.0001%; FTR, 0.0001–less than 0.0001%; VFTR, less than 0.0001%; blank means not detected.

was used in preference to some other coolant, such as dry ice-alcohol, because it was easier to maintain the supply of coolant at the tip of the cold finger where there was no solid matter to block the free supply of liquid.

The first container for the gallium was made of Pyrex; but since there seemed to be a pickup of aluminum, magnesium, and silicon (see Table 1, column B) when this was used, a silica container was made and used for the second crop of crystals.

Gallium single crystals were grown with a minimum of effort by the Kyropoulos technique and samples were taken from the tops and tips of all crystals. The first crop of crystals showed a qualitative improvement by a factor of 10 in the concentration of copper, tin, lead, cadmium, and mercury but showed an increase in magnesium, aluminum, and silicon. A recheck on the latter three showed that the impurities were strongly concentrated as a surface film and that in the body of the crystal there had been only a slight pickup of magnesium and no pickup of silicon and aluminum (Table 1, column B). These first crystals were remelted, and a second crop of crystals was grown. Further purification by a second factor of 10 was obtained in the lead, tin, and mercury content and by the elimination of spectrographically detectable amounts of silicon, aluminum, molybdenum, cadmium, zinc, germanium, calcium, and silver, all of which had appeared in the initial qualitative-analysis (Table 1, column C).

Thus, after two cycles of crystal growth, there were only five elements remaining that could be detected spectrographically as impurities. Eight others had been reduced so that they were no longer detectable. All the impurities had been reduced by a minimum factor of 10 and some by a factor of more than 1000. The total impurity content of the second crop of crystals was not more than 0.005 percent and probably nearer 0.001 percent.

Since this purity was sufficient for our purposes, no additional regrowth was attempted. It is felt, however, that by continued growth further purification by at least one factor of 10 could be achieved.

References and Notes

1. The spectrographic work for this investigation was performed by Mr. S. Cress of this laboratory.
2. Throughout this paper, the qualitative spectrographic results have been considered to indicate the approximate range of impurity concentration within the upper and lower limits indicated in the legend for Table 1.
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Estimation of Blood Loss in Hookworm Infestation with Fe^{59}: Preliminary Report[1]

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A number of workers (*1, 2, 3*) have expressed the belief that in cases of hookworm infestation the anemia frequently observed could be caused solely by the blood lost from the intestines. In a review of the subject Lane (*4*) concludes that blood loss is the most probable reason for hookworm anemia. Wells (*2*) attached *A. caninum* to the intestinal mucosa of living dogs and from iron balance observations calculated the maximum loss of 0.8 ml blood per worm per day, but he allowed for a considerable variation in the blood consumption of individual worms. Hahn and Offutt (*5*) found a method of studying the blood loss in hookworm infestation of dogs using radioactive iron.

We have used a similar technic on three human patients, who, except for the hookworm ova found in their stools, were otherwise healthy. Our purpose was to determine whether the blood loss per worm per day was constant (improbable) and to what degree the blood loss was responsible for the comparatively low hemoglobin values of our patients.

Our method was briefly as follows. Approximately 40 μc of Fe^{59}, in acid solution containing less than 1 mg of iron, were sterilized, buffered, mixed with 20 ml of the patients' own heparinized plasma, and injected intravenously. Small samples of blood were taken at intervals of 2 or 3 days and the radioactive content measured. After about 8 days, this became approximately constant and the collection of stools was

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then started and continued for about 12 days. The stools were collected in periods of 3 days and lumped together for extraction of the iron and measurement of the activity. Finally, after the last 3-day period, the patient was dewormed and quantitative worm counts made on the stools for the next 5 days. Where possible, ova concentration methods were employed after the fifth day to check the efficiency of deworming.

The radioactivity of the blood was determined with an end-window Geiger counter after the sample had been ashed and electroplated on to copper disks, following essentially the method described by Vosburgh, Flexner, and Cowie (*6*). The stools were dried with concentrated HNO_3 on a water bath, ashed, extracted with HCl, and the activity of the liquid measured with a Veall type (*7*) liquid counter. A reference solution of Fe^{59} was also made from each batch used for intravenous injection. Then if

Cs = counts/min in the liquid counter of the stool ash extract,
Cb = average counts/min ml blood over the period during which the stools were collected,
Se = counts/min ml of the reference solution electroplated,
Sf = counts/min in the liquid counter of some of the same reference solution,
Vs = volume of stool ash extract in ml,

the blood loss is given by

$$(Cs \times Se \times Vs)/(Cb \times Sf).$$

Our results, as summarized in Table 1, appear to support Wells' conception of a considerable variation in the blood loss per worm. The daily loss of 10–20 ml (i.e., 5–10 mg iron) would seem to account for the hemoglobin values obtained. They are definitely low for Angola Africans, who have a high iron intake (*8*). The observed hookworm loads combined with a normal iron intake should, therefore, produce a marked anemia.

We feel for the present unable to explain the high blood loss of patient A, harboring such a small worm load, unless there are one or two profusely bleeding injuries left by the worms. To what degree blood loss is due to these bleeding injuries, or to the consumption of blood by the worm, remains to be determined in future experiments.

TABLE 1. Hemoglobin values; blood loss during subsequent 3-day periods; average blood loss per day; number of worms recovered from the stools after deworming; and blood loss per worm per day for the 3 patients examined, and one control.

Patient	Hb (g %)	Blood loss (ml)					Hookworms	Blood loss per worm per day (ml)
		1st period	2nd period	3rd period	4th period	Av/day		
M	10.7	33.5	26.8	25.1	28.7	9.5	368	0.026
A	13.1	48.6	30.8	47.9	—	14.1	63	.22
C	13.1	71.2	60.0	42.8	51.6	18.8	354	.053
Control	—	2.3	2.3	2.0	—	0.7	—	—

On the basis of our results, it is not necessary to argue that the anemia is caused by the effect of toxins (*9*). However, our results have been obtained on patients with light loads in the age group from 18 to 30 yr, and various factors such as adaptation and immunity might counteract the full effect of hookworm toxins and so affect our conclusions. There is, therefore, a need for further studies on younger patients and those with heavier hookworm loads.

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Virulence in *Pasteurella pestis*[1]

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Studies on the nature of *Pasteurella pestis* infection have suggested that at least two factors contribute to the virulence of this bacterium: (*a*) the envelope substance (Fraction I) produced by this organism (*1–3*), which has been shown to protect it from phagocytosis (*4*) and probably blocks the action of antibodies on the cell (*4*), by virtue of the freely soluble nature of this substance (*3*); and (*b*) toxin production (*1, 2, 5, 6*), which no doubt is responsible for death from this disease (*4*). Since avirulent *P. pestis* have been found which produce both envelope and toxin, the possibility was suggested that virulence in this organism may be based upon the quantity of these substances produced. In order to investigate this possibility, a comparison of several virulent and avirulent strains was undertaken.

In initial experiments, various virulent and avirulent strains were tested for their ability to produce toxin and envelope in a casein hydrolysate mineral glucose medium at 37° C (*3*). A few virulent and avirulent strains, however, did not grow in this medium at this temperature. In the case of those strains that did grow, cell yields were so diverse that accurate estimates of toxin and envelope production were impos-

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