found to have evidence of tuberculosis. One died from hypertension, and at autopsy an active lesion was found in the apex of the right lung.

An interesting case was that of a newborn, whose mother was a far advanced case, giving a negative Von Pirquet but a positive precipitin test. The blood of the newborn gave a heavy precipitin reaction.

Of the 90 sanatorium cases 85 were positive and 5 negative. The 5 negative cases were either "healed" or "arrested."

We have found that the acid fast actinomycetes, as A. *gypsoides* and A. *asteroides,* make a good antigens as the tubercle bacillus. On the other hand the non-acid fast actinomycetes fail to react with tubercular serums. This is in agreement with the findings of Henrici and Gardner[2] and Nelson and Renrici.[3]

The glycerine broth filtrate of the tubercle bacilli as of the actinomycetes fails to give this reaction.

171 (2131)

The determination of iodin in iodin metabolism.

By J. F. McCLENDON.

[*From the Laboratory of Physiologic Chemistry, University of Minnesota, Minneapolis, Minnesota.*]

The organic material for iodin analysis is dried, mixed with and covered with CaO to render it alkaline and reduce the rate of combustion, and burned in pure oxygen in a large combustion tube with the end narrowed and bent downward for about 50 cm. The first third of this narrowed portion is covered with a thin layer of asbestos fibers to protect it from a lead coil through which cold water runs. The middle third is water-jacketed. The lower third dips into an absorbing apparatus filled with NaOH solution. The greater the amount of CaO mixed with the un-

2 *Jour. Inf. Dis.*, 1921, xxviii, 237-248.

3 PROC. SOC. EXP. BIOL. AND MED., 1921, xix, 351-352.

known material, the greater amount of iodin remains in the ash. The alkaline solution from the absorption apparatus is evaporated to 10 c.c. after adding to it the rinsings of the combustion tube. The ash is ground with water or the solution from the absorption apparatus in a ball mill and the extract is evaporated to 10 c.c. The solution is acidified until $P_H = 1$ and placed in a 12 c.c. separatory funnel and 0.1 c.c. of 0.1 N arsenius acid added, and allowed to remain for one-half hour to reduce any iodate to iodide. Then 0.1 c.c. of 5 per cent. sodium nitrite is added to oxidize the iodide to iodin which is extracted with 1 c.c. of CCl_4. The partition coefficient of iodin between CCl_4 and the aqueous solution is about 86, but varies with conditions. The iodin in CCl_4 is run into a 1 c.c. glass stoppered bottle and centrifuged to remove water droplets. The iodin is determined colorimetrically with a Bausch and Lomb Biological colorimeter specially made with cups holding 1 c.c. at 2 cm. depth, against a standard solution of pure iodin in CCl_4 (1 mg. in 10 c.c). The amount remaining in the aqueous solution may be calculated, using an approximate partition coefficient, or it may be recovered by repeated extractions. CCl_4 is freed from reducing substances by oxidation with bromine in the sunlight for about a week; then the excess Br is removed by shaking with a dilute solution of KI and then titrating the water phase with sulphurous acid while shaking the container. It is then washed and filtered (and preferably dried) and distilled, rejecting the first and last portions of the distillate. Standard solutions of iodin in CCl_4 cannot be sealed by fusing the glass without decolorizing. Preserved in glass stoppered bottles it will turn yellow. The color may be regenerated by shaking with 1/10 its volume of water of $P_H=1$ containing nitrous acid, but some of the iodin is removed. This water may then be used to regenerate new portions of standard solution without appreciable loss.

By determining the iodin in the ash separately from that caught by the absorption apparatus, the amount of CaO that must be added to make the absorption apparatus unnecessary may be determined, but under such conditions the combustion is slow.

By this method it was determined that bacteria remove iodin from the medium.