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DETERMINATION OF IODIDES IN BLOOD

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For the determination of iodides in blood, Ba(OH)₂ and ZnSO₄ should be used for deproteinization of blood instead of trichloroacetic acid. There is no effect of time, temperature and pH on the stability of PdI₄ complex.

INTRODUCTION

Urinary and faecal excretion studies show that iodide requirement is between 100-200 ug per day under ordinary physiological circumstances (Wohl and Goodhart, 1970).

Iodides are excreted by the kidneys, liver, lungs, etc., therefore, urinary and blood iodides may give an indication about various diseases, i.e. thyroid disorder, pregnancy and of iodide poisoning. In some other diseases like stomach and gastrointestinal disorders, iodides are given as medicine, in such cases it is of great clinical importance to check iodide level in blood (Spector, Mitchell and Hamilton, 1945).

Many methods have been used to determine iodides in blood (Barbour, Pilkington and Sargent 1936. Barakat, Bassioni and Wakil 1972). But none is perfect. The best reagents which may be used for deproteinization of blood during iodide determination as well as the effect of time, temperature and pH on the stability of PdI₄ complex are given below :

MATERIALS AND METHODS

Reagents

Potassium Iodide : Stock solution was prepared by dissolving 200 mg of A.R. potassium iodide in 100 ml of distilled water.

Palladium Chloride : 0.1 per cent solution was prepared by adding 100 mg of A.R. Palladium chloride in 100 ml of dilute hydrochloric acid.

Carboxy Methyl Cellulose (CMC) : The solution was prepared by adding one gram of CMC to the 100 ml of distilled boiling water to make a concentration of 1.0 per cent.

Trichloroacetic Acid (TCA) : 20 per cent TCA solution was prepared by dissolving 20 mg of TCA in 100 ml of distilled water.

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Equipment

All measurements were made with a UNICAM SP600 spectrophotometer.

Collection of Blood: Blood was collected from the Lahore slaughterhouse in bottles containing 3 per cent sodium citrate and was refrigerated at 1.3 C.

Deproteinization of Blood with 20 per cent Trichloroacetic Acid: Procedure of Barakat, Wahab and Sadr (1955) was adopted by taking 5 ml blood sample and deproteinized by adding 20 per cent 5 ml trichloroacetic acid and the mixture was centrifuged. The supernatant was diluted to 100 ml with distilled water.

Deproteinization of Blood with $Ba(OH)_2-ZnSO_4$: Deproteinization of blood was done after Sarwar *et al.* (1975).

Calibration Curve of Iodide Concentration: 5 ml solution was pipetted from the stock solution of KI (0.2 per cent) into 100 ml measuring flask. The volume was made up to the mark with distilled water. Five 25 ml flasks were taken and pipetted accurately 1, 2, 3, 4 and 5 ml from the above KI solution. Then 2 ml of one per cent carboxymethyl cellulose and 1 ml of 0.1 per cent $PdCl_2$ solution was added, until a brownish yellow colour developed. These flasks were then filled with distilled water up to the mark. In another 25 ml measuring flask all the above mentioned reactants were added except KI solution. This solution was used as a blank. Aliquots were taken from these reaction mixtures and were read at 380 nm. A calibration curve of known concentration of iodides against absorbance was plotted (Fig. 1)

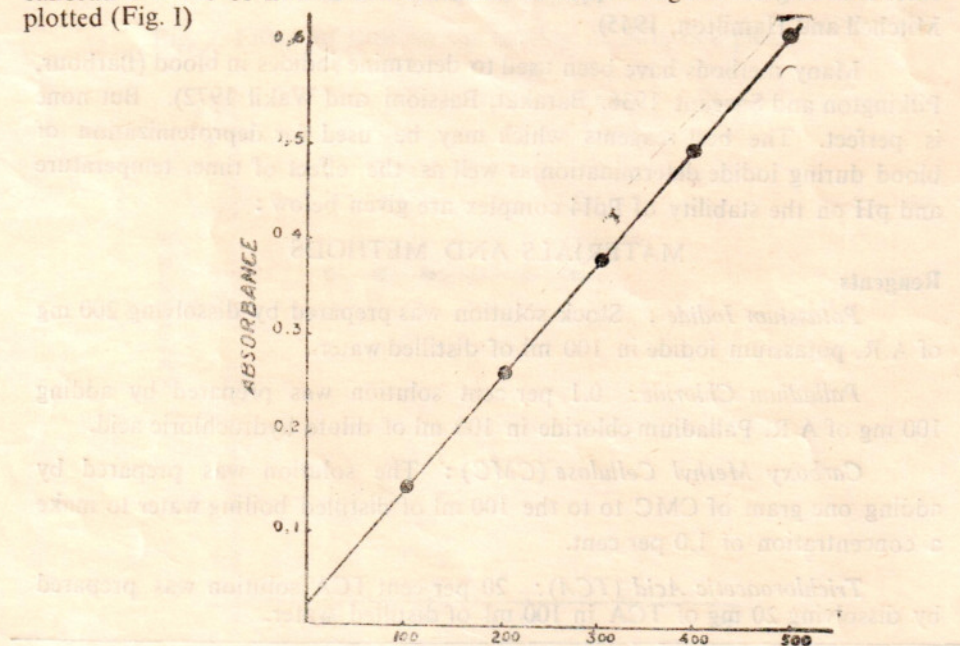


FIG. 1. AMOUNT OF IODIDES IN μg

Fig. 1. Calibration curve of Iodide concentration in distilled water.

PROCEDURE

After deproteinization of blood with trichloroacetic acid, iodide in blood was determined after Sarwar *et al.*

Three experiments on the blood samples deproteinized by $\text{Ba}(\text{OH})_2$ — ZnSO_4 solutions were conducted to see the effect of time, temperature and pH. The details of the procedure are as follows :

(a) **Effect of Temperature**

0.5 ml portion of blood containing 264 μg of RI was deproteinized with $\text{Ba}(\text{OH})_2$ — ZnSO_4 solutions as mentioned before. The filtrate was converted into 25 ml flask and the reaction was carried out by adding 2 ml of 1 per cent carboxymethyl cellulose and 1 ml of 0.1 per cent PdCl_2 until brownish yellow colour developed. The maximum absorbance was measured at 380 nm after heating the reaction mixture at 20-70 at 10°C interval.

Effect of Time

0.5 ml portion of the blood containing 237 μg of KI was deproteinized with $\text{Ba}(\text{OH})_2$ and ZnSO_4 solutions. After completion of the reaction, the absorbance of the resultant mixture was read at 380 nm. after 2, 4, 6, 8, 10 and 12 hours.

(c) **Effect of pH**

Seven portion of blood (each 0.5 ml) was taken separately each containing 190 μg of KI and was deproteinized with $\text{Ba}(\text{OH})_2$ and ZnSO_4 solutions as mentioned above. The filtrates were converted into 25 ml measuring flasks. Citrate buffer and carbonate/bicarbonate buffer solutions (Colowik and Kaplan, 1955) of pH 3, 4, 5, 6, 9.2, 10 and 10.7, respectively, were added separately in the flasks containing filtrates. The rest of the procedure for determining iodide was carried out as previously described.

RESULTS AND DISCUSSION

The results in Table 1 and 3 show that recovery of iodides in blood was better when deproteinization was carried out with $\text{Ba}(\text{OH})_2$ — ZnSO_4 instead of trichloroacetic acid. The reason is that iodides absorbed on protein precipitates. Although the recovery of iodides in trichloroacetic acid procedure is quite low but still it is better than the method described by (Mantozos and Malamos, 1968) which showed only 60 per cent recovery. So the conclusion is that $\text{Ba}(\text{OH})_2$ — ZnSO_4 reagents should be used for deproteinization during iodide determination in blood.

Fig. II, III and IV show that time, temperature and pH have no effect on the stability of PdI_4 complex, so this method can be used over a wide range of temperature and pH without any consideration of time.

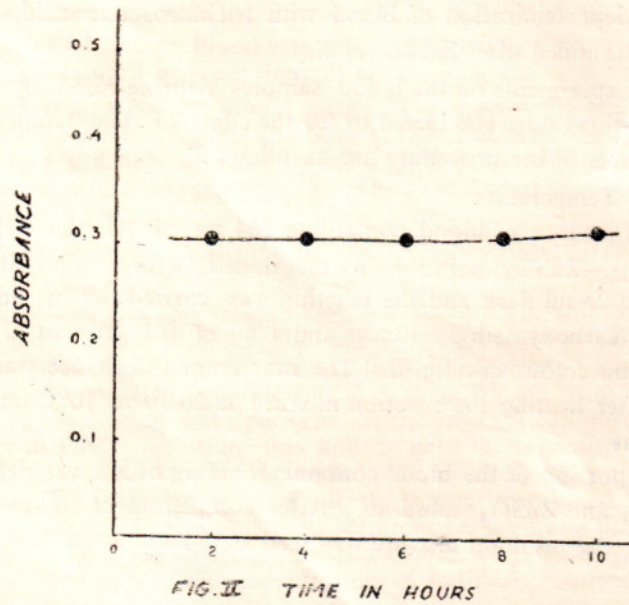


Fig. 2. Effect of time on the stability of PdI₄ complex.

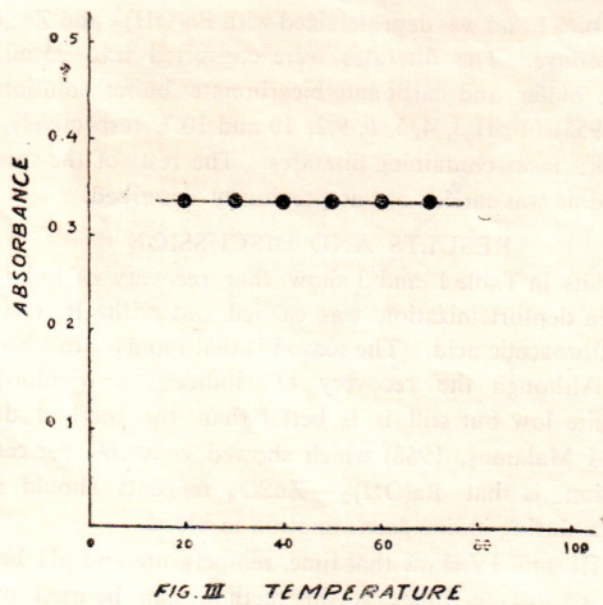


Fig. 3. Effect of temperature on the stability of PdI₄ complex.

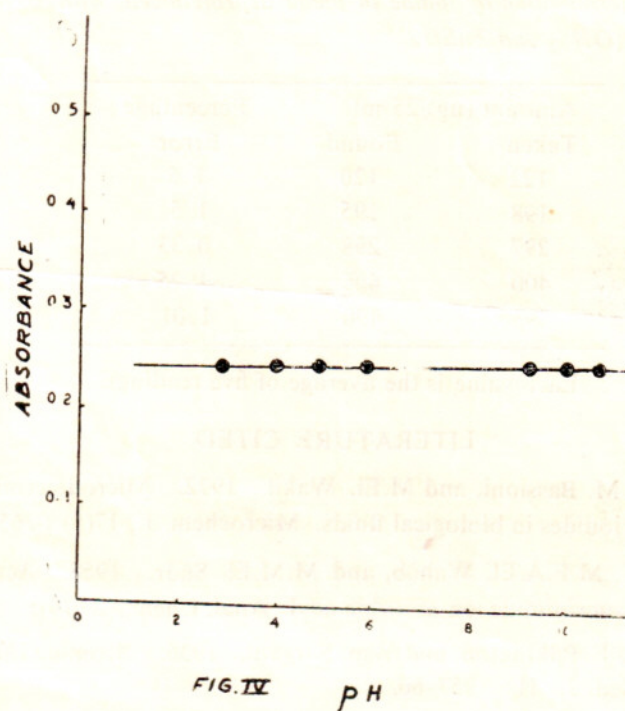


FIG. 4. Effect of pH on the stability of PdI₄ complex.

TABLE 1. Determination of iodides in blood deproteinized with trichloroacetic acid.

Amount (μ g)/25 ml		Percentage
Taken	Found	Error
80	61	23.75
102	77	24.64
150	112	25.67
188	142	24.80
230	173	24.80

Each value is the average of five readings.

TABLE 2. *Determination of iodide in blood deproteinized with a mixture of Ba(OH)₂ and ZnSO₄.*

Amount (ug)/25 ml		Percentage
Taken	Found	Error
122	120	1.6
198	195	1.5
297	298	0.33
400	405	1.25
495	490	1.01

Each value is the average of five readings.

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