# A Simplified Method for Fe<sup>55</sup> and Fe<sup>59</sup> Assay in Mixed Plasma Preparations<sup>1</sup>

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#### INTRODUCTION

Two of the radioactive isotopes of iron, Fe<sup>55</sup> and Fe<sup>59</sup>, have found extensive use in biological research. Many studies require the simultaneous employment of both isotopes, therefore a reliable and accurate method to determine each isotope is essential. It would also be helpful to use a method simpler than most in use at the present time.

Fe<sup>59</sup> has a half-life of 45 days and decays with emissions of both beta-rays and gamma-rays. These emissions consist of a beta-radiation of 1.560, 0.462, and 0.271 Mev. maximum energy as well as gamma quanta of 1.1 Mev. and 1.3 Mev. Fe<sup>55</sup> has a half-life of 2.94 years and disintegrates to manganese solely by K-capture. The manganese characteristic x-rays have an energy of 5.9 kev.

Peacock et al. (1) devised a method of determining Fe<sup>59</sup> and Fe<sup>55</sup> individually employing Geiger-Müller tubes of different characteristics after the samples were electroplated on copper planchets. Stewart and Rossi (2) reported a method of separately determining Fe<sup>59</sup> and Fe<sup>55</sup>. Their method was to divert the beta radiation of Fe<sup>59</sup> by means of a magnetic field set at right angles to the

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trajectory between the sample and a Geiger-Müller tube. The samples were electroplated onto copper planchets by the method of Hahn (3). This method has low efficiency.

Dern and Hart (4) described a method for the simultaneous quantitative measurement of  $Fe^{55}$  and  $Fe^{59}$  in mixed preparations by use of liquid scintillation counting techniques. Although the efficiency is quite good ( $Fe^{59}$ , 50% and  $Fe^{55}$ , 14%), the instrumentation is quite expensive and sample preparation is tedious and exacting.

### **METHOD**

The proposed method for preparing the samples for counting is a direct one. Heparinized blood samples are centrifuged. One milliliter samples of plasma

COMPARISON OF PROPOSED DIRECT PLANCHET METHOD VS ELECTROPLATING METHOD

100,000

100,000

1ron 55

1ron 59

1ron 59

TIME (Hours)

ELECTROPLATING METHOD

ELECTROPLATING METHOD

FIGURE 1.

COMPARISON OF PROPOSED DIRECT PLANCHET METHOD VS ELECTROPLATING METHOD

N.B. Iron<sup>55</sup> curve (upper curve) multipled x10 to avoid superimposition of curves on coordinates.

are pipetted directly onto aluminum planchets. These are dried and then counted in a gas flow detector with a Mylar window of less than 150 μg/cm<sup>2</sup> average density. (Nuclear Chicago model D-47). The sample is counted again with an aluminum filter in place. An aluminum absorber, with an average area density of 22.2 mg/cm<sup>2</sup>, was found to give the most satisfactory separation of Fe<sup>55</sup>

The amounts of Fe<sup>55</sup> and Fe<sup>59</sup> in a plasma sample can be determined by solving these two equations:

$$R = Fe^{55} + Fe^{59}$$
 (1)  
 $R' = \beta Fe^{55} + \alpha Fe^{59}$  (2)

where R and R' are the respective counting rates without and with the aluminum absorber in place. The ratio by which the Fe<sup>59</sup> counting rate is reduced by the absorber is  $\alpha$  Beta ( $\beta$ ) is the ratio by which the counting rate of Fe<sup>55</sup> is reduced by the absorber. Each of these values is determined by counting a sample containing each isotope alone.

The solution of the equations (1) and (2) are:

$$Fe^{55} = \frac{R' - \alpha R}{\beta - \alpha} \qquad (3)$$

$$Fe^{59} = \frac{R' - \beta R}{\alpha - \beta} \qquad (4)$$

TABLE I

Units*		Counting Rate; Absorber (cpm)		Computed Counting Rate (cpm) due to		Error in Determination**	
$Fe^{59}$	Fe <sup>55</sup>	Absent	In Place	Fe <sup>59</sup>	Fe <sup>65</sup>	Fe <sup>59</sup>	Fe <sup>55</sup>
1.0	0.0	12,249	2,246				
0.9	0.1	12,736	2,006	10,459	2,279	<b>−5</b> %	-10%
0.8	0.2	14,463	2,000	9,800	4,164	0 %	-18%
0.7	0.3	15,422	1,818	8,420	7,001	-2 %	- 8%
0.6	0.4	16,203	1,707	7,490	8,715	+2 %	-14%
0.5	0.5	20,000	1,645	5,836	13,971	-5 %	+10%
0.4	0.6	19,248	1,495	5,193	14,054	+6 %	- 8%
0.3	0.7	22,069	1,350	3,439	18,633	-6 %	+ 5%
0.2	0.8	23,063	1,194	2,390	20,676	-2 %	+ 2%
0.1	0.9	24,381	1,131	930	23,449	-24%	+ 3%
0.0	1.0	25,347	979		<u></u>		
					AVERAGE	-4 %	- 4%

<sup>\*</sup>Milliliters of diluted Standard Solution

<sup>\*\*</sup>Actual counting rate — computed counting rate × 100 computed counting rate

#### RESULTS

The efficiency (per cent of total disintegrations counted) of this system was determined to be 27.7 per cent for Fe<sup>59</sup> and 2.1 per cent for Fe<sup>55</sup>. This compares with 2.17 per cent for Fe<sup>59</sup> and 0.13 per cent for Fe<sup>55</sup> for electroplated samples measured by the method of Stewart and Rossi.

To date, data from ten experiments has been collected using this method, concurrently with the electroplating method. The correlation has been very good. One of these comparisons is illustrated in Figure 1. The proposed system counts about 2 per cent less Fe<sup>59</sup> radioactivity than electroplated samples counted in the same gas flow system. As expected, and possibly due to self-absorption, Fe<sup>55</sup> counts are about 25 per cent lower than electroplated samples.

The chief advantages of this system are its simplicity and saving of time. Using this method, isotope studies measuring gastrointestinal absorption of iron can be completed within one day while the electroplating method takes several days to complete. It may be seen from Table I that the efficiency and accuracy of the method in question compares favorably with that of electroplating. Large errors are noted when quantities of isotope are quite small.

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