THE EFFECT OF REPEATED BLOOD DONATION ON THE HAEMOGLOBIN, HAEMATOCRIT AND SERUM IRON VALUES OF REGULAR BLOOD DONORS

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It is generally accepted that regular donations of blood have no harmful effects on the health of the donor provided it is done at not too frequent intervals and that a haemoglobin estimation or screening test is carried out before each donation. Although these views are held by all blood bank workers there is still a widespread belief among the community that blood donation is injurious to the health. Every transfusion service has strict criteria for the acceptance of donors and no service would accept a donor who has obvious signs of ill health or whose haemoglobin level is below normal, Kwa *et al*, (1966). Yet it is difficult, almost impossible at times, to convince the prospective donor that the donation of blood is free from harmful ill effects.

From time to time it had been noticed that an occasional regular donor is found to be anaemic after a number of donations. The anaemia is usually due to iron deficiency associated with some hitherto unrecognised bleeding conditions like bleeding piles, peptic ulcer or menorrhagia. Less often it is due to an occult hemolytic state like thalassaemia minor. Often, however, no cause for the iron deficiency can be found when it must be attributed to the inadequate intake of iron in the diet to offset the effect of repeated donations.

Realising that the state of nutrition and the iron reserves of the population in Singapore may not be strictly comparable to donors from the more affluent countries like America, England and Australia, an investigation was initiated recently to study the effect of repeated donations of blood on the haemoglobin, haematocrit, mean corpuscular haemoglobin concentration, (MCHC) and serum iron values of regular blood donors. The aim of the study was to determine whether repeated donations at three to four monthly intervals exerted any harmful effects on the donor. Another object of the study was to establish the prevalence of anaemia among regular blood donors and to determine whether iron supplements were necessary for the regular donor. The possibility of any ethnic variations in the above indices was also looked into.

MATERIALS AND METHODS

Criteria for the Acceptance of Donors

In Singapore, donors are accepted if they are between the ages of 18 and 60. Volunteers must be free from major illnesses like tuberculosis, renal, or cardiovascular disease. A history of any bleeding disorder would disqualify the volunteer. Donors must weigh a minimum of 100 lb. for men and 95 lb. for women. A haemoglobin screening test by the copper sulphate technique, Phillips *et al*, (1945) is performed at each visit. Those found to have haemoglobin levels of less than 12.5 g. per 100 ml. were rejected as donors, Kwa *et al*, (1966).

In general, no other contra indications being present, a donor weighing less than 120 lb. may make a donation of between 250-300 ml. of blood and a donor weighing over 120 lbs. may make a maximum donation of up to 450 ml. The minimum interval between donations is three months, although donors are encouraged to make only three donations per year. None of the donors accepted into the study had taken iron supplements within two months prior to their donations.

PROCEDURE

For the first part of the study, male donors presenting themselves for donations were accepted without any selection except for the criteria mentioned above. The aim was to obtain data from approximately 50 donors within each donation group 1-5, 6-10, 11-15, 16-20. As the number of donors who had made 20 or more donations were limited, these donors were grouped into 21-30, 31-40 and 41 and above donation groups. As controls a larger number of donors who had not made any donation previously and presenting themselves for the first time were selected.

During the period of study any repeat donor found to have a haemoglobin level of less than 12.5 g. per 100 ml. would be rejected as donors but would be included in the survey and had

their haematological investigations performed to establish the degree and cause of anaemia.

From each donor, following their donation of blood, two samples of venous blood were collected from the proximal end of the segment tubing of the blood collection pack. The first sample was collected in EDTA for estimation of haemoglobin, haematocrit, and MCHC. The second specimen was collected in a special iron free container for estimation of the serum iron content. Estimation of haemoglobin was by the Cyanmethaemoglobin method using an EEL Haemoglobin Meter standardised against reagents obtained from Diagnostic Reagents Ltd., Thames, Oxon. Haematocrit was estimated by the Wintrobe tube technique centrifuged at 300 r.p.m. for 30 minutes. Serum iron estimations were performed by the method of Ramsay, (1954).

For the second part of the study, the haemoglobin and serum iron of a donor was followed over a total of five donations. Donors were serially allocated numbers. Those with odd numbers were provided with iron supplements in the form of Ferrous Sulphate, Gr. 3 to be taken twice daily for two weeks after each donation. Those with even numbers served as controls and were not provided with iron supplements. All donors were asked to come back for further donation after four months. Reminder calling up cards were routinely sent to all donors after three months.

RESULTS

Table I shows the distribution of donors according to ethnic and donation groups. A total of 663 donors were included in the study of whom 258 were Chinese, 207 were Malays, 88 were Indians and 110 were Other Races, comprising mainly Eurasians and Europeans. The 243 donors of different ethnic groups who had not made any donations previously served as the controls for comparison.

TABLE I

DISTRIBUTION OF DONORS ACCORDING TO ETHNIC AND DONATION GROUPS

No. of Previous Donations	Chinese	Malays	Indians	Others	Total
0	108	71	46	18	243
1— 5	20	48	6	8	82
6—10	24	29	6	13	72
11—15	24	18	11	11	64
16—20	16	13	6	12	47
21—30	43	27	9	28	107
31—40	18	1	2	15	36
41 and above	5	—	2	5	12
TOTAL	258	207	88	110	663

Haemoglobin Levels

Table II shows the mean haemoglobin and standard deviation of donors of different ethnic and donation groups. The mean haemoglobin of the control series was 15.3 g. per 100 ml. for Chinese, 15.2 g. per 100 ml. for Indians, 15.3 g. per 100 ml. for Malays and 14.7 g. per 100 ml. for Other Races. Within each ethnic group there were no significant differences in the mean haemoglobin values between donors of different donation groups. There was no evidence to suggest a tendency for the mean haemoglobin level to fall with repeated donations. There were no significant ethnic differences in the mean haemoglobin values of the control series or of the different donation groups.

The frequency distribution of the haemoglobin values of donors of all ethnic groups is shown in Fig. 1. There were no donors with haemoglobin levels below the 12.0 g. per 100 ml. level in the control series because donors were only accepted if their haemoglobin was above 12.5 g. per 100 ml.

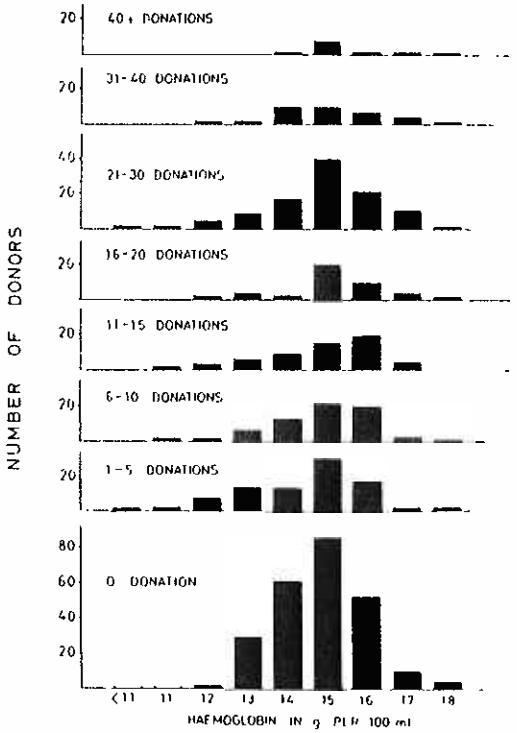

Fig. 1. Frequency distribution of haemoglobin values of donors (all races) of different donation groups.

TABLE II

MEAN HAEMOGLOBIN AND STANDARD DEVIATION IN DONORS OF DIFFERENT ETHNIC AND DONATION GROUPS

No. of Donations	Chinese		Indians		Malays		Others	
	Mean Hb.g./100ml.	S.D.	Mean Hb.g./100ml.	S.D.	Mean Hb.g./100ml.	S.D.	Mean Hb.g./100ml.	S.D.
0	15.3	1.0	15.2	0.7	15.3	1.2	14.7	1.1
1— 5	15.0	2.0	15.8	1.8	14.9	1.3	15.5	0.2
6—10	15.4	1.1	15.6	1.0	15.3	1.1	15.2	1.6
11—15	15.2	1.7	14.4	1.9	15.4	1.1	15.1	1.5
16—20	15.9	1.0	15.1	2.0	15.4	1.3	15.6	1.3
21—30	15.3	1.3	15.2	1.5	15.3	1.2	15.4	1.5
31—40	15.8	1.3	16.4	1.5	14.6	—	15.0	1.3
41 and above	15.6	1.0	15.6	0.2	—	—	16.1	1.1

TABLE III

MEAN HAEMATOCRIT AND STANDARD DEVIATION IN DONORS OF DIFFERENT ETHNIC AND DONATION GROUPS

No. of Donations	Chinese		Indians		Malays		Others	
	Mean PCV %	S.D.	Mean PCV. %	S.D.	Mean PCV. %	S.D.	Mean PCV. %	S.D.
0	44.8	3.5	45.0	3.6	45.2	2.5	44.3	3.4
1— 5	44.5	4.3	46.8	4.1	43.9	3.2	45.1	1.4
6—10	46.0	2.2	46.2	3.2	45.1	2.4	45.2	3.9
11—15	46.1	4.5	43.6	3.7	45.3	2.5	44.6	1.2
16—20	47.4	2.7	43.8	2.6	45.5	2.9	45.3	3.8
21—30	46.3	3.3	46.4	3.6	45.1	3.1	46.0	3.5
31—40	46.6	3.2	50.5	2.5	44.0	—	44.0	3.1
41 and above	45.6	2.8	45.5	0.5	—	—	45.6	1.7

Among the 420 repeat donors 13 (3%) had haemoglobins of less than 12.5 g. per 100 ml. Five donors were from the 21-30 donation group, four from the 11-15 donation group, two from the 1-5 donation group, and one each from the 6-10 and 31-40 donation group. There were no donors in the 40 and above donation group with haemoglobin levels of less than 12.5 g. per 100 ml.

Haematocrit

Table III shows the mean haematocrit and standard deviation of the donors. The mean haematocrit of the control series was 44.8% for Chinese, 45.0% for Indians, 45.2% for Malays and 44.3% for the Other Races. Mean haematocrit values remained relatively constant for the different donation groups with no evidence to suggest a fall with repeated donations. The frequency distribution of the haematocrit values of donors of all ethnic groups is shown in Fig. 2. Twenty, (3%) donors had haematocrits of less than 40%, 6 of whom belonged to the control group, five to the 1-5 donation group, three each to the 11-15 and 21-30 donation group and one each to the 6-10, 16-20, and 31-40 donation group (Fig. 2).

Mean Corpuscular Haemoglobin Concentration

MCHC and standard deviation is shown in Table IV. The mean MCHC of the control series was 33.0% for Chinese, 33.3% for Indians, 33.6% for Malays and 33.2% for Other Races. The mean MCHC for all ethnic groups remained within the normal range for all donation groups. There was no evidence of a trend to a fall in the mean MCHC with repeated donations. Taking a MCHC of 32% as the lower limit of normal, there were 70 (10%) donors with MCHC values below the lower limit, 22 of whom were donors from the control group (Fig. 3).

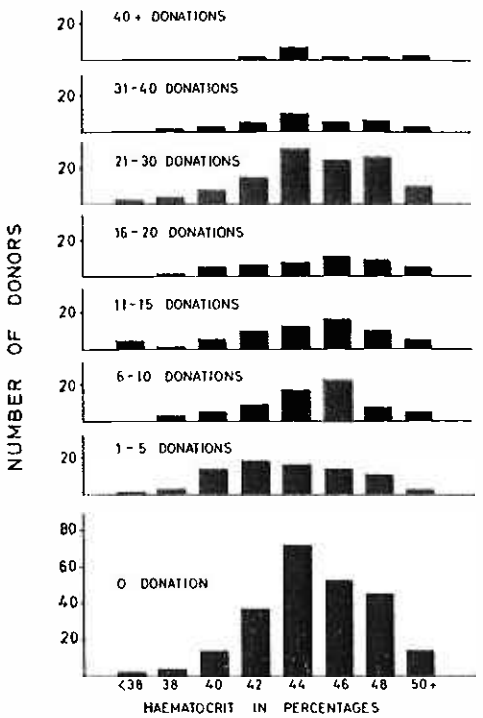

Fig. 2. Frequency distribution of haematocrit values of donors (all races) of different donation groups.

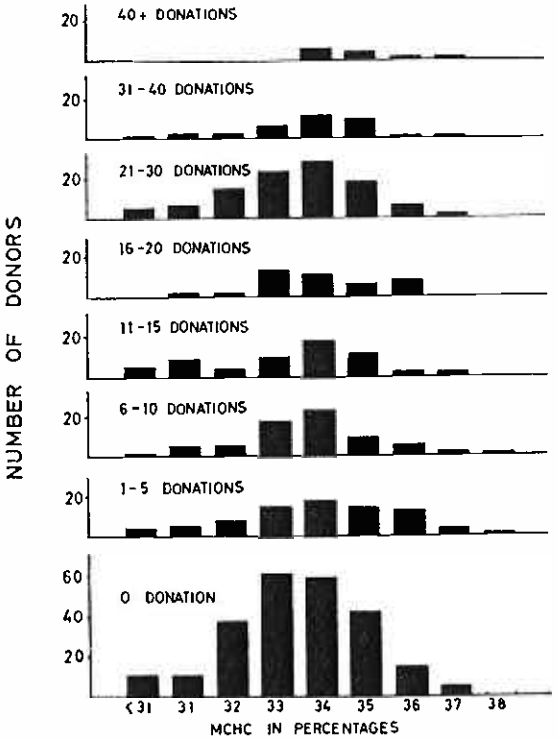

Fig. 3. Frequency distribution of MCHC of donors (all races) of different donation groups.

Serum Iron Levels

The mean serum iron values and the standard deviation is shown in Table V. The level for the control group of donors was 117.3 ug. per 100 ml. for Chinese, 106.4 ug. per 100 ml. for Malays, 105.3 ug. per 100 ml. for Indians and 103.1 ug. per 100 ml. for the Other Races. The mean serum iron tended to fluctuate with different donation groups. In the case of the Chinese, the mean serum iron level fell slightly from 117.3 ug. per 100 ml. initially to 110.8 ug. per 100 ml. for the 11-15 donation group after which values fluctuated. The mean serum iron for the 41 and above donation group was 103.6 ug. per 100 ml. In the case of Indian donors the mean serum iron fell from the initial value of 105.3 ug. per 100 ml. to 74.7 ug. per 100 ml. for the 11-15 donation group after which it fluctuated some what. The mean serum iron of the 41 and above donation group was 138.5 ug. per 100 ml. The mean serum iron for Malay donors fell from the initial value of 106.4 ug. per 100 ml. to 100.0 ug. per 100 ml. for the 6-10 donation group after which it showed a variable rise. In the case of Other Races, the serum iron fell from 103.1 ug.

TABLE IV

MEAN MCHC AND STANDARD DEVIATION IN DONORS OF DIFFERENT ETHNIC AND DONATION GROUPS

No. of Donations	Chinese		Indians		Malays		Others	
	Mean MCHC %	S.D.	Mean MCHC %	S.D.	Mean MCHC %	S.D.	Mean MCHC %	S.D.
0	33.0	1.8	33.3	1.8	33.6	1.6	33.2	1.5
1— 5	33.6	2.1	33.7	1.6	33.9	1.6	34.3	0.3
6—10	33.6	1.4	33.8	0.8	33.8	1.7	33.5	1.6
11—15	32.9	2.2	33.0	2.3	33.9	1.3	33.8	1.9
16—20	33.5	0.8	34.2	2.7	33.9	1.6	34.5	1.5
21—30	33.1	1.5	32.9	1.7	33.8	1.5	33.5	1.9
31—40	33.8	1.3	32.3	1.3	33.2	—	34.0	1.5
41 and above	34.2	0.6	34.2	0.7	—	—	35.2	0.3

TABLE V

MEAN SERUM IRON AND STANDARD DEVIATION IN DONORS OF DIFFERENT ETHNIC AND DONATION GROUPS

No. of Donations	Chinese		Indians		Malays		Others	
	Mean Serum Iron ug./100ml.	S.D.	Mean Serum Iron ug./100ml.	S.D.	Mean Serum Iron ug./100ml.	S.D.	Mean Serum Iron ug./100ml.	S.D.
0	117.3	39.6	105.3	30.2	106.4	29.4	103.1	26.9
1— 5	112.7	27.2	96.5	43.0	107.5	34.5	101.9	25.6
6—10	112.0	32.3	95.5	34.0	100.0	29.4	92.5	29.6
11—15	110.8	38.7	74.7	33.6	121.6	46.4	101.4	27.3
16—20	117.9	34.4	105.5	35.4	115.9	34.0	102.3	25.9
21—30	121.1	43.3	104.3	41.3	105.7	34.5	108.9	38.6
31—40	110.8	32.9	88.1	33.0	148.0	—	110.3	24.4
41 and above	103.6	21.1	138.5	14.4	—	—	132.2	49.9

per 100 ml. initially to 92.5 ug. per 100 ml., after which there was a variable rise.

Fig. 4 shows the frequency distribution of the serum iron values of the donors. For the control series it will be seen that there is an asymmetrical distribution with a larger proportion of donors with serum iron values below 100 mg. per 100 ml.

Thirty-six (6%) donors had serum iron values of less than 60 ug. per 100 ml., five of whom were from the control series of 243 donors, six from the 1-5 donation group, five from the 6-10 donation group, 9 each from the 11-15 and 21-30 donation groups, and two from the 31-40 donation group.

Effect of Iron Supplements on Haemoglobin and Serum Iron

Tables VI, VII show the mean haemoglobin and serum iron values of donors of all ethnic groups following successive donations of blood. Three hundred and thirty-one donors received iron supplements after each donation whilst 320 did not receive any.

It will be seen that the number of donors decreases with increasing number of donations. This drop out of donors occurred in spite of reminder calling cards being sent to all the donors. The drop out was most marked after the first donation and was found to be due mainly to donors who had no intention of being regular donors and therefore failed to come up for further donations.

From Table VII it will be seen that the mean haemoglobin fell very slightly from 15.4 g. per 100 ml. initially, to 15.1 g. per 100 ml. after the 5th donation for the group who were not given iron supplements. There was no significant change in the haemoglobin level with successive donations for the group who received iron supplements.

Mean serum iron levels for donors receiving iron supplements rose slightly from the initial value of 111.4 ug. per 100 ml. to 118.1 ug. per 100 ml. after the 4th donation. After the 5th donation there was a slight fall in the mean serum iron to 103.6 ug. per 100 ml. Mean serum iron values for the donors not receiving any iron supplements increased slightly from 111.1 ug. per 100 ml. initially to 123.5 ug. per 100 ml. after the 3rd donation but fell to 110.8 ug. per 100 ml. after the 5th donation.

Among donors receiving iron supplements three were disqualified from making further donations because of anaemia with haemoglobins of less than 12.5 g. per 100 ml. Among donors not receiving iron supplements there were six donors who became anaemic.

Among the group receiving iron supplements there were 14 who had serum iron values of less than 60 ug. per 100 ml. initially. Thereafter, the number of donors found to have serum iron values of less than 60 ug. per 100 ml. was 35 in all. The corresponding figure for donors who received no iron supplements was 17 donors initially and 33 in all after five donations.

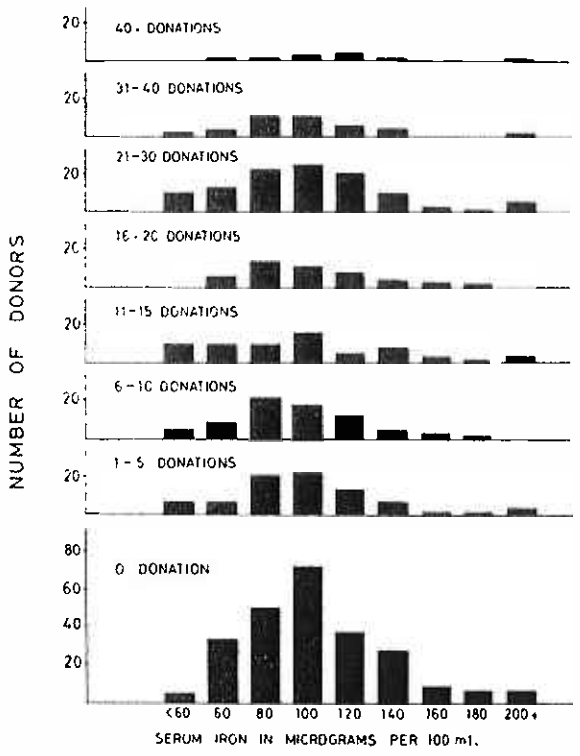

Fig. 4. Frequency distribution of serum iron values of donors (all races) of different donation groups.

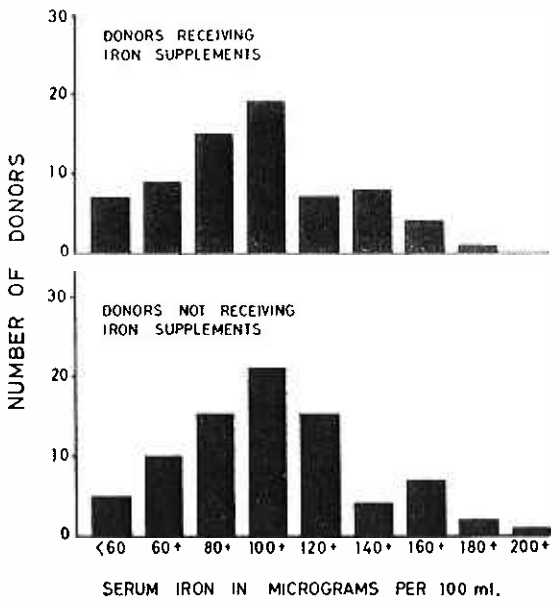

Fig. 5. Frequency distribution of serum iron values of donors after completing five blood donations.

TABLE VI

MEAN HAEMOGLOBIN AND SERUM IRON IN DONORS RECEIVING IRON SUPPLEMENTS

	No. of Donors	Mean Hb. g./100ml.	No. of Donors with Hb. Below 12.5g./100ml.	Mean Serum Iron ug./100ml.	No. of Donors with Serum Iron Below 60 ug./100ml.
INITIAL	331	15.2	—	111.4	14
After 1 donation	164	15.2	3	111.4	10
After 2 donations	143	15.1	—	119.1	5
After 3 donations	128	15.1	—	118.1	6
After 4 donations	117	15.1	—	118.1	8
After 5 donations	70	15.1	—	103.6	6

TABLE VII

MEAN HAEMOGLOBIN AND SERUM IN DONORS **NOT** RECEIVING IRON SUPPLEMENTS

	No. of Donors	Mean Hb. g./100ml.	No. of donors with Hb. Below 12.5g./100ml.	Mean Serum Iron ug./100ml.	No. of donors with Serum Iron Below 60 ug./100ml.
INITIAL	320	15.4	—	111.1	17
After 1 donation	175	15.4	1	118.8	9
After 2 donations	158	15.3	1	121.5	6
After 3 donations	145	15.1	2	123.5	6
After 4 donations	128	15.2	1	117.1	7
After 5 donations	80	15.1	1	110.8	5

The frequency distribution of the serum iron values of donors after completion of the five donations with and without iron supplements is shown in Fig. 5. There is no difference in the frequency distribution of the two groups.

Fig. 6 shows the distribution of donors according to whether there was a nett increase or decrease in the serum iron values after five donations. Among the donors receiving iron supplements 27 out of 70 showed an increase, the remaining 43 showing a decrease. Among donors not receiving any iron supplements 40 out of 80 showed an increase whilst the remaining 40 showed a decrease.

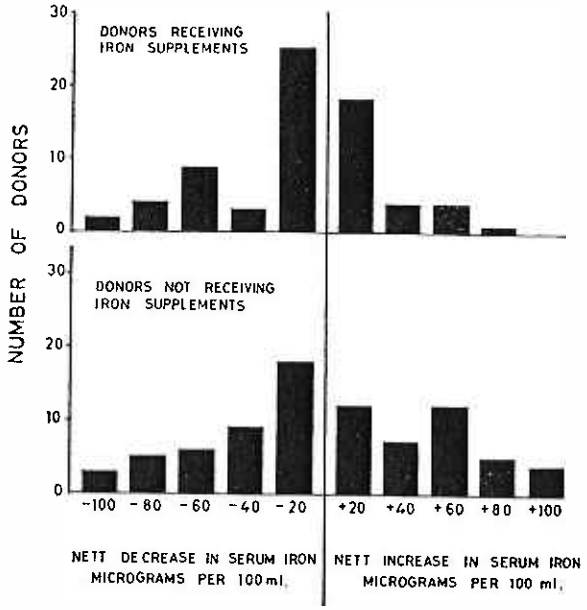

Fig. 6. Frequency distribution of donors after five blood donations.

DISCUSSION

Our results indicate that the mean haemoglobin, haematocrit and MCHC values of donors of all ethnic groups remained relatively constant after repeated donations of blood. There were no significant differences in the mean values of these indices among the different ethnic groups.

The findings suggest that in the majority, the haemopoietic system is capable of meeting the extra demands necessitated by the regular donation of blood. In approximately 3% of donors however, repeated donation of blood led to the development of mild degrees of anaemia necessitating their withdrawal from the donor panel.

It must be admitted that because of the large number of drop outs from the study, these figures may be an under estimate, as the donors investigated may not be entirely representative of the volunteers as a whole. It might even be argued that donors may have stopped donating blood because they felt unwell due to the development of anaemia, whereas those who continued were not anaemic. However, inquiry gave the impression that this was not so and that the majority of drop outs stopped donating because they were not prepared to continue as regular donors.

The finding of a number of individuals among the control group with low haematocrit and MCHC suggest that mild degrees of iron deficiency anaemia is not uncommon among the population. This is confirmed by the finding of serum iron of less than 60 mcg.% in 2% of donors with haemoglobin of over 12.5 mg. per 100 ml. presenting themselves for the first time. Approximately 7% of repeat donors were found to have low serum iron, the actual percentage ranges from 6% to 14% for the different donation groups. An investigation of these donors showed that there was usually a combination of factors like inadequate diet or occult blood loss which was responsible for the gradual depletion of iron stores.

Mean serum iron of the control group was highest for the Chinese, 117.3 mcg.% and lowest for the Other Races 103.1 mcg.% with approximately similar values for Malays, 106.4 mcg.% and for Indians, 105.3 mcg.%. These findings are lower than the generally accepted mean level of 126 mcg.% for normal adult males reported by Bothwell and Finch (1962). Recently, Lie-Ingo and de Witt (1964) studied serum iron levels in blood donors presenting themselves for the first time. They found mean serum iron of 123 mcg.% for Chinese, 112 mcg.% for Malays and 114 mcg.% for Indians. Our results would appear to be slightly lower than those found for the same ethnic groups in Malaya.

A comparison of the mean serum iron among donors of different donation groups showed that although there was a trend to a fall in the mean values with increasing number of donations initially, this trend was not sustained with increasing number of donations. The results on the whole are difficult to explain and may possibly be due to the small number of donors in the latter donation groups.

The provision of iron supplements to regular blood donors did not appear to have any significant effect on the mean haemoglobin or serum iron values.

Three donors receiving iron supplements became anaemic compared to six from the group not receiving iron supplements.

The mean serum iron remained relatively constant between 111 mcg.% and 119 mcg.% after the 1st, 2nd, 3rd and 4th donation. After the 5th donation, there was a slight fall in the mean serum iron to 103.6 mcg.% in the group receiving iron supplements. No such fall was observed in the group not receiving iron supplements. The percentage of repeat donors with serum iron of less than 60 mcg.% was approximately 5% for both groups of donors irrespective of whether they were receiving iron supplements or not.

Laurell (1947) investigating the serum iron concentration of regular blood donors some two months after their last donation found the haemoglobin and serum irons to be slightly subnormal. Hagberg *et al* (1958) also showed that donors who had given five to six units of blood in the previous year or two had diminished serum iron values. The importance of blood donation as a source of blood and iron loss was stressed by Moore (1948), whilst Finch *et al* (1950) demonstrated the importance of iron as a limiting factor in haemoglobin regeneration after regular withdrawal of blood at weekly intervals in healthy males with normal iron stores.

There is thus general agreement that those who donate blood regularly should be provided with extra iron supplements, Mollison (1961), Heisto and Foss (1958) and that if no supplemental iron is given the minimum interval between donations should be at least three months.

Our findings indicate a definite need to provide iron supplements to all regular blood donors to prevent the development of anaemia or latent iron deficiency. In view of the finding that supplemental iron therapy in the form of ferrous sulphate gr. 3 twice daily for two weeks after the donation did not have any significant effect on reducing the incidence of anaemia among repeat donors, a study is currently in progress to determine if extending the period of supplemental iron therapy to four weeks would lead to a reduction in the incidence of anaemia among regular donors.

SUMMARY

The effect of repeated donations of blood on the haemoglobin, haemotocrit, MCHC and serum iron of regular blood donors has been investigated. In the majority, mean haemoglobin, haematocrit, MCHC and serum remained relatively constant with no evidence to suggest a tendency to fall with repeated donations. In approximately 3% repeated donations resulted in the development of mild degrees of anaemia, whilst low serum iron values were demonstrated in approximately 7%.

Iron supplementation in the form of ferrous sulphate gr. 3 twice daily for two weeks did not exert any significant effect on the indices or in reducing the incidence of anaemia or the development of latent iron deficiency among regular donors.

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