CHAPTER 4

IRON METABOLISM

The need of the body for iron is self-evident by reason of the constitution of haemoglobin. In addition,iron, through its biocatalytic activities, participates in oxidation-reduction reactions in all living cells. Iron is present in every living cell of the body. Iron is,therefore, essential for life and it is extremely important to have some knowledge of its distribution, sources, requirements and metabolism if the function of iron in the treatment of iron deficiency anaemia is to be understood.

The metabolism of this vital compound, its anabolism and catabolism, its incorporation into the heme molecule as well as the complex factors which regulate absorption, excretion and the preservation of iron balance, have been more completely studied than any of the other factors essential for erythropoiesis.

DISTRIBUTION OF IRON

Iron is widely distributed in the animal's body, where it exists in both ionized and non-ionized forms. Iron is present in haemoglobin of red blood cell, in blood-forming organs, in bloo-destroying organs, in chromatin of all tissue cells and in oxidative enzyme systems.

TYPES OF IRON

The distinction between two types of iron viz. organic and inorganic is of considerable historical interest and deserves brief consideration. It is now universally agreed that inorganic iron is absorbed, assimilated, and incorporated into haemoglobin. Moreover, it is far superior to food iron in causing a rapid blood response in iron deficiency anaemias. The attempt is still made to promote organic iron preparations as superior in the treatment of anaemia.

(a) Inorganic Iron :

Inorganic iron includes all dissociable iron salts; whether the iron is combined with organic or inorganic anion is immaterial. Most of the available iron is inorganic. Many foods, formerly thought to contain organic iron, are now considered to contain ionizable iron.

(b) Organic Iron :

The iron in organic combination is nondissociable and

constitutes an integral part of the molecule. The examples of such organic combinations of iron are haemoglobin, cytochrome, certain nucleoproteins and foods containing heme compounds.

VALENCY OF IRON

Bivalent (Ferrous, Fe^{++}) and Trivalent (Ferric, Fe^{+++}) forms are existing. All forms are assimilated, but ferrous (bivalent) salts are more rapidly absorbed and utilised than ferric (trivalent) salts because the latter require reduction before they can be absorbed.

Abundent evidence is available from clinical studies and from radio-active iron experiments (Hahn et al.,1945; Moore, 1957) that ferrous iron is absorbed and utilised at least three times more than ferric iron.

ABSORPTION OF IRON

Normally the food contains 10-20 mg. of iron. About 10 per cent of the dietary iron, i.e. 1-2 mg. is absorbed. But in deficiency states, the absorption may be increased even upto 60 per cent.[24] Thus, in states of iron deficiency, characterised by low serum iron levels and, usually, high iron-binding capacities, the potential iron absorption is greatly augmented.

Recently, with the use of radio-active iron, it has been shown that patients with iron deficiency, contarary

to what might be expected, do not absorbs food iron any more than do normal persons.[20] Although iron deficient patients cannot absorb more iron from the food than healthy persons, they fortunately absorbs large amounts of inorganic iron [20] — hence the importance of ferrous sulphate in iron therapy and the failure of substances rich in iron like spinach and meat.

Continued administration of iron preparations to persons who do not have iron deficiency, usually lead in a short space of time to virtual non-absorption of the metal. If this was not so, the relatively common practice of adminstering iron over prolonged periods to patients with chronic anaemias not associated with haemorrhage would be expected to result in accumulation and excessive deposition of iron in tissues, finally leading to what is known as haemochromatosis. It is true that cases of haemochromatosis have been recorded but its frequency must be extremely rare.

Food iron is in a state of organic combination, and requires conversion to simpler compounds before absorption can take place. Since the beginning of the current century, oral administration of inorganic iron has been shown to result in fairly good absorption as evidenced by animal experiments.

Iron is absorbed only in the ionic form and ferrous iron is much more efficiently absorbed than ferric

iron. The stomach has an important action on the iron ingested with the food. First, it converts the colloidal ferric compounds of food stuffs ~~it~~ into simpler 'monomolecularly dispersed' ferric ions. Secondly, in the acidic medium of stomach, certain substances in the food help to reduce the ferric ion to the ferrous form. Thus, it is clear that the acidic gastric juices not only converts non-ionized iron to ionic form but also favours the reduction of ferric iron to the more readily absorbable ferrous iron.

Certain substances like ascorbic acid, cysteine and the SH groups of proteins in food reduce ferric salts at and below pH 5, but not at pH 7. This is of great importance because it has been demonstrated that iron can be absorbed only in the ferrous form.

Absorption of iron occurs mainly from stomach and upper part of small intestine. No iron is absorbed from the colon. Though iron can be absorbed all along the intestinal tract, very little is absorbed normally except in the duodenal region. The region just below the pyloric sphincter is, therefore, the most effective in absorption of ferrous iron. Nevertheless, if large doses of iron are given, more of the intestinal tract, perhaps even the large intestine and stomach may absorb significant amounts of iron.

As the iron reaches further down in the gut, the digestive juices of pancreas, bile and the intestinal mucosa tend

to raise the PH to 6-7. The ferrous iron at the higher pH is liable to autoxidation to ferric hydroxide. Thus,in the lower portion of the gut, the absorption of iron is rendered difficult.

After absorption, iron is taken to the tissues for cellular respiration or to the storage organs like liver, spleen, etc. from where it is doled out as needed to the bone marrow for haemopoiesis.

MECHANISM OF IRON ABSORPTION

Iron is absorbed intracellularly, indicating a vital activity of the mucosal cells. According to the current theory, there is a specific protein in the receptor cells of the intestinal mucosa. This specific protein is known as 'Apoferritin,' and it serves as an iron acceptor and carrier. Apoferritin is a colourless, iron-free protein having a molecular weight of 460,000. Apoferritin combines with iron (ferric hydroxide) to form 'Ferritin.'

Ferritin is a brown coloured, iron-containing protein. It is a compound of the protein apoferritin with a basic ferric hydroxide phosphate of complex structure $[(FeOOH_8)(FeOPO_3H_2)]$. It is found abundantly in the liver, spleen and bone marrow as well as in the cells of the intestinal mucosa where iron absorption is preceding. The ferritin molecule contains about 23 per cent iron, i.e. about 2,000

iron atoms.[15] This iron can be displaced to yield the iron-free protein namely, apoferritin.

The presence of iron in the receptor cells of the intestinal mucosa stimulates these cells to produce within a few hours this specific protein to which the iron gets attached. The amount of iron absorbed, therefore, depends upon the presence in the mucosal cells of apoferritin which acts as an iron acceptor to form ferritin. This, in turn, yields its iron moiety to the plasma which transfers it to the stores where it is once again stored as ferritin. Thus, the iron acceptor compound is said to be concerned with the transport of iron across the mucosal barrier and into the blood stream.

REGULATION OF IRON ABSORPTION

Since the body cannot regulate its iron content by excreting excess amounts, control of absorption of iron is exceedingly important.

For many years, it was assumed that the gut does exert precise control over the amount of iron absorbed, the hypothetical regulatory device having been termed as "Mucosal Block." In this mechanism, the mucosa of small intestine plays an active part in the regulation of iron absorption. The mucosal block depends upon alternate saturation and desaturation, of an acceptor mechanism in the mucosal cell,

desaturation taking a matter of days but saturation taking only a matter of hours.[31]

According to above hypothesis of regulation of iron absorption, the mucosal cell must provide a one-way transfer of ferrous iron from gut into the cells. The ferrous iron moves into the cell and in the presence of certain enzymes and oxygen, it is oxidised to ferric iron which combines with apoferritin to form ferritin. At the blood stream end of the cell, the ferric iron of ferritin is reduced to ferrous iron by the reducing system of the cell. This ferrous iron then enters the blood strear, gets autoxidised once again to form ferric iron. The ferric iron thus formed then combines with an iron-binding plasma protein β_1 globulin which is known as 'transferrin' or 'Siderophilin,' in presence of $C\theta_2$ to form Iron-transferrin. The plasma iron is deposited and stored in the tissue - depots as ferritin once again (Figure 1) .

When larger amounts of iron enter the mucosal cell, some of the ferric iron may be trapped in the form of ferritin. With still larger amounts, there may be an accumulation of ferric iron in the front part of the cells so that the pores of the apoferritin get completely clogged. In these circumstances there may be some delay before the reducing mechanism at the rear part of the cell can clear the ferric iron and allow unclogging of the ferritin, resulting in subsequent

delay in further absorption to take place.

The mucosal block operates when the mucosal cells become saturated with ferrous iron which in turn is said to be in equilibrium with the ferritin stored in the mucosa and with the ferric ions of the blood plasma. More iron is further absorbed, if the ferritin in the mucosa gets depleted or if the plasma iron falls. Many investigators have finally come to question the theory of "Intestinal intelligence."

It has been claimed that there is a specific iron-free protein in the intestinal mucosa. This specific protein is known as apoferritin. Granik supposed that the apoferritin molecule may be a part of the membrane of the mucosal cell.[31] It is instructive to note that in experimental iron deficiency anaemias, apoferritin is not detected in the cells of the intestinal mucosa, but it appears quickly when iron feeding is instituted. This leads to the attractive theory that apoferritin is specifically formed in response to the stimulus supplied by the presence of ionized iron in the gut.[130] Apoferritin combines with ferrous iron to form ferritin. It is suggested that apoferritin is constantly being formed and broken down. Its degradation only stops when it combines with iron to form ferritin. The presence of ferritin prevents further absorption of iron, until the ferritin can give up its iron to the blood stream in form of plasma iron.

Recently, Granick's hypothesis is criticised and there is a tendency to regard the ferritin as a convenient

form of non-toxic storage of iron in the mucosal cells rather than as a regulator of iron absorption. It seems an over simplification to state that the mucosal cells play the major role in control of iron absorption; rather multiple factors are concerned, some tending to increase the absorption, others tending to decrease the absorption, and mucosal cell itself playing a part among these factors.

Another mechanism of regulation of iron absorption is supposed to be connected with the so called 'redox' level of the cell. Here, the ferric hydroxide is reduced to ferrous iron by the reducing system of the cell. This ferrous iron then diffuses into the blood stream. In anaemic conditions, such reduction is favoured by the lower oxygen tension in the mucosal cell. This facilitates more absorption of iron. Thus, the mucosal cell responds, by absorbing more iron, to the low oxygen supply brought about by low haemoglobin level in the blood. Therefore, it might be postulated that it is the low oxygen supply which is the factor indirectly increasing the iron absorption, thus satisfying the increased demand by the body in iron deficiency anaemia.

Certain facts are exceedingly difficult to explain on the basis of such theories. For example, normal or increased iron absorption occurs frequently in the presence of abnormally high levels of body iron or in the absence of anaemia. Conversely, large doses of parenteral iron have little

effect in 'blocking' the iron absorption.

Investigators are currently restudying the problem of iron absorption using newer techniques, but at present no single theory accounts for the known facts. Conceivably, iron absorption is under dual control of two factors[104] viz. (a) the size of the iron stores, and (b) the bone marrow activity or the degree of anaemia.

Various studies continue to indicate, that iron absorption can be influenced by various factors. Some workers observed that iron absorption was increased above the normal in iron deficient subjects. Other investigators noted that iron absorption was accelerated when erythropoiesis was active, and depressed when iron stores were large. Nevertheless, if there is a regulatory mechanism, its nature is obscure and its operation imperfect.

The iron absorption may be said to depend mainly on:[100]

(a) Bodily needs of iron - the absorption of iron is strictly controlled by the needs of the body.

(b) Conversion and maintenance of iron in an absorbable (ferrous) form.

(c) Permiability of the absorbing cells to iron.

However, the actual mechanism of iron absorption still remains obscure and it seems likely that further advances will be made only at a cellular level.[31]

FACTORS AFFECTING IRON ABSORPTION

The factors affecting iron absorption may be described under four headings viz. (I) Factors within the gut, (II)Iron stores and erythropoiesis, (III) Mucosal factors and (IV) Miscellaneous factors.

I. Factors Within the Gut :

(1) Diet : The presence in the diet of large amounts of phosphate, calcium, phytate and phytic acid may inhibit iron absorption by forming insoluble, non-absorbable iron compounds. Milk, cereals and pulses diminish the iron absorption.On the other hand, diet low in phosphate and phytate encourage iron absorption. Moreover, the presence in the diet of a number of reducing substances like ascorbic acid and sulpha hydryl compounds, which are set free during digestion, increase the iron absorption by reducing the food iron to readily absorbable ferrous form. When the total iron intake is kept constant, the amount of iron absorbed bears a direct relationship with the bulk of the meal.

(2) Drugs : The regular use of drugs like antacids may impair iron absorption. Thus, peptic ulcer patients respond poorly to iron therapy if they are receiving antacids. Their anaemia responds to the same iron therapy when alkali ingestion is stopped. Conventional antacids like mucin, calcium carbonate, aluminium hydroxide and magnesium hydroxide

all decrease the iron absorption. The adverse effect is exerted mainly through an increase in pH of gastro-intestinal contents. Kelley, Logan and Christ (1955) reported a case of sprue in which cortisone appeared to improve iron absorption.[12] Adjuvants like copper and cobalt are supposed to augment the iron absorption.

(3) Vitamins : Vitamin C (ascorbic acid) by virtue of its reducing property increases iron absorption. The addition of this reducing substance, which converts ferric food iron into more easily assimilable ferrous form, is not of assistance when ferrous preparations are given. The allegation that Vitamin B complex aids in iron absorption and utilisation has been proved fallacious.[12]

(4) Reaction of Medium : Iron salts are poorly disassociated in a medium with a pH greater than 5. The pH of the gastric contents prevent the formation of insoluble and non-dissociable complexes which form between iron and other food constituents at a pH above 5. The relatively low pH of the duodenal contents similarly favours absorption by preventing oxidation to the ferric form. At an acid pH reductants like ascorbic acid, SH group of proteins present in food convert ferric iron to readily absorbable ferrous iron. On the other hand, at an alkaline pH, iron combines with amino acids, nucleic acids and phosphorus to form insoluble, non-absorbable iron complexes.

(5) Site of Iron Absorption : The largest amount of iron is absorbed from the duodenum, where the conditions are most favourable; smaller amounts are absorbed from stomach and jejunum; little amount is absorbed from the ileum and none from the colon.

(6) Gastric Acidity : Gastric HCl plays an important role in iron absorption. It acts in multiple ways. It reduces ferric to ferrous iron, increases ionization of ferrous iron, and retards the formation of insoluble iron complexes.[26] Patients with iron deficiency anaemia frequently have achlorhydria, which interferes with iron absorption, especially when therapy with ferric salts is employed. Experimentally, however, subjects with achlorhydria neither show any reduced absorption of food iron nor do they show enhanced absorption after addition of HCl, (Moore 1955).

(7) Gastric Secretions : There is evidence to suggest that there may be other, yet undefined, local secretary factors which may influence the iron absorption. Patients with partial gastrectomy show poor absorption of food iron and not of free iron salts.[31] This could mean that some factor in normal gastric secretion, other than HCl, is necessary for normal absorption of food iron.

(8) Panereatic Secretions : Experimental evidence suggests that pancreatic secretions may play some part in iron absorption. For example, Siderosis may be produced in cats by the ligation

of pancreatic ducts.[31]

(9) Bile : The influence of normally secreted bile on intestinal absorption of iron is disputed. However, the experimental evidence suggests that bile is not necessary for iron absorption unless there is an excess of fat in the diet, in which case absorption is diminished, probably because of the formation of insoluble iron soaps.[51]

II. Iron Stores and Erythropoiesis :

Apart from local factors inside the gut, recent experimental work of Bothwell (1958) suggested that the state of iron stores and the rate of erythropoiesis are of major importance in control of iron absorption.

Experimental increase of iron stores or depression of erythropoiesis decreases the iron absorption. On the other hand, reduction of iron stores, or stimulation of erythropoiesis (by haemorrhage or by inducing haemolytic anaemia) increases the iron absorption.[31] Blood transfusion stimulates the bone marrow which afterwards responds better to oral iron therapy.[127] The effect of two opposing factors viz. increased iron stores plus increased erythropoiesis, may nullify each other, resulting in relatively normal iron absorption. How the effect of stimuli is conveyed to the mucosal cells is quite obscure. Nevertheless, the clinical experience, in general, confirms these experimental findings.

III. Mucosal Factors:

The contribution of mucosal block in the regulation of iron absorption has already been described.

In certain clinical conditions the normal regulation of iron absorption is broken down. In coeliac disease and idiopathic steatorrhea there is failure to absorb iron as a part of general malabsorption syndrome. This occurs even in presence of severe iron deficiency. On the other hand, excessive iron absorption occurs in haemochromatosis. Here the mucosal cell continues to absorb iron because of some familial and constitutional defect as well as due to disturbed oxidative metabolism of the mucosal cell. An inborn error of metabolism in the mucosal cells has been postulated in haemochromatosis leading to undue absorption of iron, overcoming the normally functioning mucosal block.

IV. Miscellaneous Factors :

(1) Need of Iron : The most important factor affecting iron absorption is the need of the body for further supply of iron. When the need exists, factors like chemical nature of food iron and gastric acidity are unimportant. Physiological conditions like pragency and lactation increase the demands of iron, hence greater iron absorption during these periods. The iron absorption is also increased in patients suffering from iron deficiency anaemia than in a normal person.

(2) Type of Iron : Ferrous iron is readily absorbed than ferric iron. Experimental evidences have shown that ferrous iron is absorbed and utilised at least three times as effective as ferric iron. The poor utilisation of ferric iron is partly due to the fact that they readily form insoluble compounds with various substances in the gastro-intestinal tract. Moreover, in contrast to food iron, medicinal iron is absorbed particularly well in states of iron deficiency.[26]

(3) Dosage : Proportionately more iron is absorbed from smaller doses than from larger doses. The absorption at high dosage level may be as low as 1-2 per cent.[26] At low dosage levels, a much higher percentage of the total amount given is utilised for haemoglobin formation than at higher dosage levels. Further more, the ingestion of large doses of iron in infants can cause severe rickets because of interference with assimilation of phosphorus.

(4) Age : Increasing age diminishes the iron absorption.[127]

(5) Infection : Presence of infection considerably diminishes the iron absorption from gastro-intestinal tract.[82]

DISTRIBUTION AND STORAGE OF IRON

Iron absorbed from the gastro-intestinal tract is transported through the blood to various storehouses from which it is mobilized for haemoglobin formation.(Figure 2)

The distribution and storage of iron normally depend on two factors: (a) iron intake by the individual and (b) iron demand by the bone marrow. Although iron is present in all tissue cells, its concentration differs greatly in different organs of the body. The various iron compartments of the body are as follows :

1. Haemoglobin Iron in Erythrocytes :

The largest fraction of the body iron is present in erythrocytes in the form of haemoglobin performing the essential function of oxygen carriage. The iron in haemoglobin is present in the ferrous form. Normally, 2.65 gms. of iron is present in haemoglobin constituting about 55 to 60 per cent of the total iron content of the body. However, this value varies considerably.

2. Non-haemoglobin Iron in Erythrocytes:

A small amount of free iron (unassociated with haemoglobin) is present in erythrocytes. The amount exceeds that present in plasma and is uninfluenced by various conditions like iron deficiency or iron excess which affect other fractions of the body iron. The function of this compartment of body iron is unknown.

3. Serum Iron :

Serum iron represents the non-haemoglobin iron compartment of the blood. In a normal adult, it amounts to 4 mg. constituting approximately 0.1 per cent of the total

body iron. This iron is in ferric form and is in combination with β_1 globulin.

4. Muscle-haemoglobin Iron :

This fraction of the body iron is present in striated muscles in the form of myoglobin. It is a true non-circulating haemoglobin. The amount of muscle haemoglobin hardly decreases even in severe anaemias and increases with age and exercise. The iron in muscle haemoglobin constitutes about 7 per cent of the total body iron and probably performs the function of carrying the oxygen from blood to striated muscle.

5. Parenchymal Iron :

This fraction of the body iron is present in all tissues of the body, but especially in skeletal muscle. It includes the iron of cytochrome, catalase, and peroxidase. The amount of iron in this form varies from 1 to 3 mg. per 100 gm. of fresh muscle tissue, constituting about 16 per cent of the total body iron. The function of parenchymal iron is not known.

Neither myoglobin iron nor parenchymal iron is utilised for the regeneration of blood haemoglobin regardless of the severity of anaemia. Both of these fractions together constitute 23 per cent of the total body iron. It is clear, therefore, that nearly 1 gram of tissue iron is unavailable for haematopoiesis.

6. Visceral Storage Iron :

Storage iron is the most important fraction of the body iron. Storage iron is stored in certain organs rich in reticuloendothelial cells, especially the liver, spleen and bone marrow, and occasionally the kidney. The total amount of storage iron is probably equivalent to the iron content of 250 grams of haemoglobin, constituting about 20 per cent of the total body iron. Storage iron represents a reserve derived from the diet and from the continuous physiological destruction of red blood cells. Storage iron, in contrast to parenchymal iron, is available for use by the bone marrow. It increases in hemosiderosis, after repeated blood transfusions and after excessive doses of intravenous iron.

Storage iron is utilised for haemoglobin synthesis only as needed. Excessive intake of iron in absence of increased demand merely augments storage in the viscera. When the stores are exhausted, body growth and haemoglobin formation suffer. Liver efficiently conserves iron released during the physiological destruction of erythrocytes, and this iron supplies a major reserve for the formation of new haemoglobin. The spleen is alsc important as a storehouse for iron and ranks second to the liver.

Iron is stored in body in several forms, particularly as hemosiderin and ferritin. The iron content of hemosiderin is almost double that of ferritin. Nevertheless, wk

ferritin, which is also concerned with the regulation of iron absorption in the gastro-intestinal mucosa, is normally more important than hemosiderin as a source of iron for haemoglobin synthesis. Hemosiderin is the storage form of iron which is visible under optical microscope, whereas ferritin is visible under electron microscope.[15]

The various iron compartments of the body differ in that not all the iron in the storage depots is equally accessible for haemoglobin synthesis. Approximately 10 to 15 per cent of the total iron reserve (3 to 4 per cent of the total body iron) constitutes what is called "Labile iron pool." The liver contains the larger portion of the labile iron, whereas the spleen contains more of the stable iron reserves.

The approximate values for the larger fractions of iron distribution in human are given below :-

TABLE NO. I

Approximate Iron Content of various Iron compartments

Iron compartment	Iron content	
	Grams	Per cent of Total
I. IRON-PORPHYRIN(HEME) COMPOUNDS:		
1. Blood haemoglobin iron	2.45	57
2. Muscle haemoglobin iron	0.30	7
3. Parenchymal iron (Heme enzymes like cytochrome, Peroxidase and catalase).	0.69	16
II. NON-IRON-PORPHYRIN COMPOUNDS :		
4. Visceral storage iron (Ferritin, Hemosiderin etc. in liver, spleen and bone marrow) plus serum iron (Transferrin).	0.64	15
5. Other tissue iron	0.22	5
III. T O T A L ...	4.30	100

EXCRETION OF IRON

Iron is excreted through various channels, like faeces, urine, bile, skin, menses and milk. Small and insignificant amounts are eliminated in faeces, urine and bile, totalling approximately 1 mg. daily in normal human adults. Moreover, Hussain et al. (1960) have shown that skin represents an important excretory channel through which considerable amount of iron can be lost.

1. Faecal Excretion :

The faecal excretion of iron is mainly determined by the unabsorbed food iron. The faecal excretion of iron derived from normal haemoglobin breakdown varies from 0.2 to 0.9 mg. per day in normal individuals and only one fifth as high in patients with iron deficiency anaemia.

Experimental studies of normal and iron-deficient subjects with radio-active iron have shown that the normal excretion of iron in faeces is about 0.33 - 0.52 mg. daily, and that the excretion in iron deficiency is as little as 0.03 - 0.06 mg. daily.[130]

The faecal excretion of iron does not increase even after intravenous injection of large doses of iron. McCance and Widdowson showed that in a case of haemolytic anaemia the faecal output was almost identical with the

dietary intake and contained no supplement derived from the haemolysed cells.

2. Urinary Excretion :

Excretion of iron in the urine is also limited and insignificant. The urinary excretion of iron in human subjects on a normal diet is 0.08 - 1.6 mg. daily, with an average of 0.4 mg. The amount is constant for a particular individual and is higher in females than in males. Barer and Fowler found that the urinary excretion of iron in males is 0.4 mg. daily whereas that in the females is 0.5 mg. daily.[130] The urinary excretion of iron is not increased by oral administration of large doses of the metal.[26] However, about 1-2 per cent of an intravenously administered dose of iron is detectable in the urine, probably derived from the non-transferrin-bound diffusible fraction of serum iron. Barer and Fowler (1949) have shown that the urinary excretion of iron is increased by administration of an acid salt (e.g. ammonium chloride) and markedly decreased by an alkaline salt. Some workers, however, have regarded the urinary iron as not a true excretion but merely as derived from cell debris from the urinary tract.

3. Biliary Excretion :

Iron excretion in bile is at a low, constant rate. In principle, it has been concluded that about 3 per cent of the iron derived from the breakdown of effete corpuscles

is excreted in bile. Acute intravascular haemolysis increases the amount in proportion to the extent of haemolytic process and can even increase the biliary excretion of iron tenfold. In contrast, the biliary excretion is not increased by intravenous or oral administration of large doses of iron. It is, therefore, logical to deduce that the biliary excretion of iron is entirely associated with pigment excretion and that the iron is bound to the pigment.

4. Dermal Excretion :

The dermal loss of iron can occur through insensible perspiration as well as through active sweating. A proper appreciation of the excretory role of the skin is important particularly in tropical climates in which sweating occurs normally.

Active sweating is accompanied by desquamation of the superficial layers of epidermal cells and hence sweat always contains epithelial debris. Thus, the major portion of loss of iron is primarily associated with desquamation probably of cell lining the sweat glands.

Different investigators have found different figures for iron excretion in sweat, the discrepancy between the results being due to the different techniques employed for iron estimation. In general, it has been suggested that as much as 15 mg. of iron can be lost per day under conditions

of maximal sweating.[64] Although the amount of iron in 'cell free' sweat is considerably less than that of 'cell rich' sweat, it also might assume great significance in the tropical hot humid climate where people may sweat profusely to the extent of 2 to 11 litres per day (Kuno, 1934).

The loss of iron in insensible perspiration is considered to be insignificant and negligible. Iron deposition in growing hairs and nails is another means by which some of the metal is 'lost.'[51,136]

5. Menstrual Loss :

The normal adult excretes about 1.2 mg. of iron per day, and normal adult female additionally loses iron in the menstruation at an average rate of 1 mg. per day over the whole menstrual cycle.[130] It is because of this reason that the iron requirement of females is double than that of males. In pregnancy, no iron is lost in the menses, but on an average 2.7 mg. per day is supplied to the foetus.[45]

6. Lactation Loss :

About 1.5 mg. of iron is excreted in the milk daily during the lactation period.[130]

IRON BALANCE

Maintenance of a positive iron balance is readily accomplished with a low rate of absorption when the only

requirement is replacement of that lost by excretion. On the other hand, the demand for iron may be greatly increased as a result of growth, mensturation, pregnancy, blood donations and pathological bleeding. A useful figure for the estimation of iron lost by bleeding is based upon the fact that a given amount of blood expressed in milli-litres contains approximately one-half that number of milligrams iron. This co-relation is virtually exact in the case of blood with a haemoglobin value of 15 gm. per 100 ml. Such blood contains 50 mg. of iron per 100 ml. The amount of iron lost by a single blood donation of 500 ml. is approximately 250 mg.[44]

The amount of iron necessary to replace the daily physiological loss of Haemoglobin can be calculated theoretically. The life span of the human erythrocyte averages 120 days. A normal person, therefore, destroys 1/120th of his circulating red cells per day. An average man has a blood volume of 6 litres containing 15 gm. haemoglobin per 100 ml. and each gram of haemoglobin contains 3.34 mg. of iron. From these facts it may be simply calculated that the daily destruction of red cells liberates about 26 mg. of iron. Probably 95 per cent of the iron released from erythrocytes is reused and therefore only a small amount of iron absorbed from the diet is adequate to maintain the iron balance. Iron balance is said to be maintained by the daily

utilisation of 0.5 - 1.0 mg. of dietary iron in adult males and 1.0 - 1.5 mg. in adult females; the higher value in women is the result of iron loss by manstruation.

Hahn estimated that the biliary excretion of iron is about 3 per cent of the amount of iron released from erythrocytes, namely 0.8 mg. In females, who have a smaller blood volume, the amount if is about 0.6 mg. Accepting Barer and Fowler's figure for urinary excretion as 0.4 to 0.5 mg. according to sex, then by these two routes the body loses 1.2 mg. daily in case of males and 1.1 mg. daily in case of females.

Moreover, females suffer a menstrual loss which has been estimated as 10-40 mg. at each period, giving an average of about 1 mg.per day over the whole menstrual cycle.Females, therefore, lose almost twice as much iron as males. Pregnancy almost doubles this figure. Lactation adds not more than 1.5 mg. to the daily loss in bile and urine, but if menstruation is re-established during lactation the total iron loss becomes almost as great as in pregnancy.[130]

Thus, in order to maintain the iron balance, the supply of iron must compensate for the small daily excretion of 1.2 mg. a day in adult males, for the additional menstrual loss in females, for the added burden of pregnancy or lactation with or without menstrual loss, and for the adequate feeling of the continuously increasing storage space in the

growing child.

IRON REQUIREMENTS

Dietary iron requirements are difficult to determine because dietary iron is only partially absorbed and not all the absorbed iron is utilised for haemoglobin synthesis.

Infant's initial iron requirement is 300-500 mg. and this amount is provided by the mother,[31] and thereafter iron is derived from the diet. In pregnancy, on an average 2.7 mg. daily is to be provided for the foetus.[45] It is estimated that each pregnancy demands approximately 400 mg. of iron for the foetus. During the period of growth from infancy to adolescence, large amounts of iron are needed since expansion as well as excretion is to be covered, and for the first 20 years of life an additional 0.3 — 0.6 mg. daily must be provided.[45] When the boy reaches adult life his iron requirement drops sharply. From puberty onwards, females have the additional requirements for menstruation and pregnancy, amounting to 0.5 to 1.0 mg. per day.[31] Thus, the daily utilisation of iron in normal adult male is approximately 1-1.5 mg., in normal adult female 2 mg., in pregnancy 4 mg. and during lactation 2.5 - 3 mg.

Gubler[45] cites the National Research Council's estimate that allowing for only 10% absorption of iron from the food, the probable iron requirements are as follows :

1.	Infants	...	1.0 mg. per kg. per day
2.	Children	...	0.6 mg. per kg. per day
3.	Adult male	...	10.0 mg. per day
4.	Adult female	...	12.0 mg. per day
5.	Pregnant woman		15-20mg. per day
6.	Lactating woman		15-18mg. per day

The average diet makes this amount available but the margin in case of women is very small, so that the occurrence of pregnancy or even a moderately heavy menstrual loss can sooner or later reduce the iron reserves and precipitate an anaemic state.

Since the absorption of iron is an unknown factor and the needs of the body are variable, it is highly desirable to have iron intake considerably higher than the minimal needs. The diet of a healthy person should, therefore, contain 12-15 mg., preferably 20 mg. of iron per day.

Recently, Apte and Venkatachalam5 have suggested that the safe level of iron intake on a normal Indian diet which is based on cereals and pulses lies somewhere between 17 to 21 mg. per day.

In order to maintain health with adequate haemopoiesis, these varying iron requirements must be made available in the diet or otherwise, must be rendered absorbable by adequate digestive function and must be absorbed by a healthy mucosa.

SOURCES OF IRON

The main sources of iron in Indian diets are cereals, pulses, vegetables, condiments and spices. Cereals (2 to 8 mg. per cent) and pulses (2 to 10 mg. per cent) alone contribute the major portion of iron in every day diet.[63] Fortunately, leafy vegetables, condiments and spices also contain relatively high concentrations of iron. The importance of foodstuffs as sources of iron depends not only upon the concentration of iron in each ~~individual item~~ but also upon the amounts of individual items in the diet.

The actual iron content[60] of important foodstuffs in order of importance is as follows :-

I. CEREALS :

1.	Bajra	3.1	mg. per ounce
2.	Wheat, flour, whole (atta) ...	2.0	,,
3.	Rice, raw, home - pounded ...	0.8	,,
4.	Rice, raw, milled ...	0.3	,,

II. PULSES :

5.	Bengal gram (chana) ...	2.8	,,
6.	Black gram (Adad)	2.8	,,
7.	Red gram without husk (Tur dal)	2.5	,,
8.	Green gram (mag)	2.4	,,
9.	Peas, dried	1.3	,,
10.	Beans, dried	0.6	,,

III. LEAFY VEGETABLES :

11.	Amaranth, tender (Tandaljo) ...	6.1	mg. per ounce
12.	Fenugreek (Methi)	4.8	,,
13.	Coriander (Kothmir) ...	2.8	,,
14.	Spinach (Palakh)	1.4	,,

IV. ROOT VEGETABLES :

15.	Carrot	0.4	,,

V. OTHER VEGETABLES :

16.	Bitter gourd (Karela) ...	2.7	,,
17.	Onion stalks	2.1	,,
18.	Drumstick (Saragvo) ...	2.0	,,
19.	Cluster beans (Govar) ...	1.6	,,

VI. FRUITS :

20.	Dates	3.0	,,
21.	Mangò, green	1.3	,,
22.	Figs	0.3	,,

VII. CONDIMENTS AND SPICES :

23.	Cumin seeds (Jiru)... ...	8.8	,,
24.	Asfoetida (Hing)	6.3	,,
25.	Turmeric (Haldhar) ...	5.3	,,
26.	Coriander seeds (Dhana) ...	5.1	,,

27.	Mustard (Rai) ...	...	5.1 mg.per ounce
28.	Pepper, dry ...	...	4.8 ,,
29.	Fenugreek seeds (Methi)	...	4.0 ,,
30.	Tamarind, pulp ...	...	3.1 ,,

VIII. MILK AND MILK PREPARATIONS :

31.	Skimmed milk powder	...	0.4 ,,
32.	Butter-milk ...	...	0.2 ,,
33.	Curd ...	...	0.1 ,,
34.	Milk, cow's ...	...	0.1 ,,
35.	Milk, buffalo's ...	...	0.1 ,,
36.	Milk, goat's ...	...	0.1 ,,

IX. FLESH FOODS :

37.	Crab, muscle ...	...	6.0 ,,
38.	Liver, sheep ...	...	1.8 ,,
39.	Egg, duck ...	...	0.9 ,,
40.	Egg, hen ...	...	0.6 ,,
41.	Fish, big ;..	...	0.6 ,,
42.	Fish, small ...	...	0.3 ,,

X. MISCELLANEOUS :

43.	Pāppads (Pāpad) ...	...	4.9 ,,
44.	Juggery (Gol) ...	...	3.2 ,,

Singh et al.[109] studied the availability of iron in some common Indian pulses and concluded that the availability of iron was greater in germinated pulses than in ungerminated ones. Among green vegetables, tender amaranth, fenugreek, coriander, bitter gourd and drum stick are the rich sources of iron. Among condiments and spices, cumin seeds, asfoetida, turmeric, coriander seeds, mustard, dry pepper and fenugreek seeds contain large quantities of iron. Among miscellaneous foods, pappadas and juggery contain good amount of iron. Condiments and spices are used for flavouring foods and some of them contribute the appreciable amounts of essential nutrients even in small quantities in which they are used. A balanced use of all these foodstuffs must therefore be emphasised.