ISSN 0377-4910

ICMR BULLETIN

Vol. 30, No. 2 February, 2000

IRON ABSORPTION AND ITS IMPLICATIONS ON STRATEGIES TO CONTROL IRON DEFICIENCY ANAEMIA

Iron deficiency anaemia is one of the most common nutritional disorders world-wide, especially in India and other developing countries. Young children and women in the reproductive age group are the most vulnerable to iron deficiency anaemia. Surveys in different parts of the country reveal that 87% of pregnant women suffer from anaemia and about 10% have severe anaemia (H < 80 g/l). Variations in the prevalence rates of anaemia are seen within the country with the lowest prevalence of 33% being reported from Andhra Pradesh to the highest of 98% in Rajasthan[1]. A recent study on the prevalence and etiology of nutritional anaemia in early childhood in an urban slum area of east Delhi indicated a high prevalence (76%) of anaemia and iron deficiency in 41% children[2]. Earlier studies from the National Institute of Nutrition (NIN), Hyderabad and other studies prior to 1985 showed an average anaemia prevalence rate of 68% in preschool children[3,4].

Consequences of Iron Deficiency Anaemia

Iron has diverse biological functions. It is this diversity that accounts for the wide-ranging impact of its deficiency. Iron deficiency commonly remains unrecognised. Severe anaemia is an important risk factor in pregnancy. Reports from India indicate that 16% of all maternal deaths are attributable to anaemia[5]. Maternal anaemia also contributes to an increase in perinatal mortality, low birth weight, and foetal wastage. Studies on low income pregnant women in India showed a three-fold greater incidence (34.5%) of premature deliveries in severely anaemic women compared to the normals. Maternal immune depression and increase in morbidity has also been reported among anaemic pregnant women[6].

In the past, several studies have shown that iron deficiency anaemia often leads to irreversible impairment of the child's learning ability and other behavioural abnormalities[7]. The neurochemical roles of iron are not fully understood but it is clear that low levels of iron can have a significant adverse impact on brain function. In adults iron deficiency results in impaired work performance and productivity.

Etiology of Iron Deficiency

Dietary factors play an important role in the development of iron deficiency. Although most habitually consumed diets in different regions of India contain adequate amounts of iron (26 mg)[8], absorption of iron from such diets is only 1-5%[9]. Other factors that contribute to anaemia are chronic blood loss due to hookworm infestation and malaria. In the vulnerable segments of the population the increased demand due to the physiological status aggravates the deficiency of iron.

Iron Requirement

Absorption of iron is influenced by the amount of iron in the body; it decreases if individuals are iron replete and increases if they are deficient. Normally men lose only 1 mg of iron every day (14 µg/kg/d), which is easily replenished through the diet. Such losses are proportionally less in women (0.7 to 0.8 mg/d). Menstrual bleeding causes an additional loss of 0.4 to 0.5 mg iron daily amounting to a total loss of 30 µg/kg/day. Women also lose iron to the placenta and the foetus, amounting to about 1.3 g of iron as the cost of normal delivery. Another critical period of increased iron requirements is early childhood and adolescence. During 6-24 months of age both physical growth and brain development occur at a rapid rate. In adoles cence there is a marked demand for iron to increase blood volume and muscle mass. For Indians, the recommended dietary allowances for iron computed are: adult men 28 mg/d, adult women 30 mg/d, pregnant women 38 mg/d, lactating women 30 mg/d, boys aged 13-15 yr 41 mg/d, girls 13-15 yr 28 mg/d, and children 7-9 yr 26 mg/d[10].

Sources of Iron

All the iron needed for the biological functions comes from diet. Although cereal-pulse based diets are regarded as good sources of iron, the non-heme iron present is relatively poorly absorbed. In contrast, iron from red meat (heme iron) is highly bioavailable. Vitamin C enhances the utilization of non-heme iron but substances like tannin from tea as well as fibre and phytates from plants inhibit it

Approaches to the Control of Iron Deficiency Anaemia

The National Nutritional Anaemia Prophylaxis Programme (NNAPP) was initiated in 1970 to control iron deficiency anaemia in the vulnerable groups through daily supplements of iron-folic acid tablets. The suggested prophylactic doses of iron and folic acid respectively were 60 mg and 500 µg for pregnant women and 20 mg and 100 µg for children per day for 100 days[11]. These tablets were distributed to the high risk groups by the local health workers.

An evaluation in 11 states during 1985-86 indicated very poor coverage and performance of the Programme[12]. There was no impact of the Programme on the prevalence of anaemia in pregnant women of more than 37 weeks of gestation. Hence, the dosage of iron in iron-folic acid tablets was increased from 60 to 100 mg in 1992[13].

Other sustainable approaches to control anaemia are food fortification and dietary diversification. Two different technologies of fortification of common salt were developed at the NIN, Hyderabad as a long-term strategy to control and prevent iron deficiency anaemia in the population. These are (i) iron fortified salt – common salt fortified with iron; and (ii) double fortified salt – common salt fortified with iron and iodine. In depth studies carried out with this strategy have clearly shown that fortified salts improve haemoglobin status[14].

Dietary diversification to improve absorption of iron by lowering inhibitor and increasing promoter concentrations has been suggested. This may need nutrition education and changes in dietary habits of the population.

Therapeutic Supplementation of Iron – Some Concerns

Absorption of iron is a highly regulated process, due to which a large fraction of the administered dose remains unabsorbed in the small intestine. This is particularly important in the context of daily supplementation of iron in prophylactic doses. Recent clinical studies have shown that intermittent supplementation of iron is as beneficial as daily supplementation[15,16]. This is believed to be due to the reduced iron absorption when the intestine is exposed continuously to high doses of iron. To overcome this effect it is suggested that large doses of iron may be supplemented every 5th day which is the turnover cycle of the intestinal absorptive cells. Added to this, some recent studies raise doubts about the relationship between haemoglobin levels and pregnancy outcome and even question the need for routine iron supplementation during pregnancy[17,18]. This has brought to focus the issue of exposing the intestine to large amounts of supplementary iron which may generate free radicals via the Fenton reaction leading to peroxidative damage of the tissue. It is suggested that excess iron in the GI tract increases free radical generation and alters the structure and function of the absorptive microvill[19].

Recent Studies at the NIN, Hyderabad

For the past 5 years studies have been carried out at the NIN, Hyderabad to answer some of the concerns regarding iron absorption and the behaviour of intestine to large supplements of iron.

Mechanism of intestinal absorption of iron in humans and rats

The duodenum and jejunum are the sites of maximal absorption of dietary iron. The means by which iron is

taken up by the intestine and transported to the blood remains incompletely understood. The iron transport protein transferrin and its receptor have been shown to be involved in the intracellular transport of iron in all tissues except the intestine. The specific role of iron storage protein ferritin has also not been established.

The NIN studies have demonstrated that the WKY strain of rat is a good animal model closer to humans to study iron absorption. Using this model the simultaneous identification of iron and the proteins involved during iron absorption was done. Iron deficient intestine was found to have a higher concentration of the transferrin receptor. Functionally this is expected to increase iron absorption through the following mechanism. During the intestinal transit of iron, luminal transferrin gets saturated with iron. Subsequently, binding of this transferrin to mucosal cell surface transferrin receptor occurs. In the intestinal cells iron was shown to get internalised as a complex of transferrin-transferrin receptor. Intestinal ferritin regulates the extent of such an uptake process. The possibility of such a system existing in the human intestine was demonstrated using endoscopic biopsy specimens. The receptor mediated uptake was found to occur in the iron deficient and control states but not in the iron excess state, explaining the theory of mucosal block as the mechanism of regulation of iron uptake by the intestine[20].

Iron induced oxidative damage

In view of the limited capacity of absorption, the possible deleterious effects of excess iron was tested. This was carried out in iron deficient WKY female rats supplemented (D^+) for 15 days with $FeSO_4$ equivalent to 8 mg of iron (a dose required to regenerate Hb from 80 to 140 g/l) and compared with iron deficient (D) and iron adequate (C) rats. The following were evaluated: levels of intestinal mucosal cell thiobarbituric acid reactive substances (TBARS), protein carbonyls and degraded DNA on electrophoresis as markers of lipid peroxidation, protein oxidation and DNA damage, respectively. Activities of endogenous antiperoxidative enzymes status were also measured. As markers of functional integrity, the activities of alkaline phosphatase and lys-ala-dipeptidyl aminopeptidase at the sites of iron absorption were measured. The mucosal cell turnover number was determined (^{3}H-thymidine incorporation into DNA). In addition, the concentrations of ferritin, transferrin in intestinal mucosa and ceruloplasmin level in serum were measured. The salient findings are summarised below.

Iron supplementation resulted in increased oxidative stress in the small intestine of iron deficient D^+ rats. It was observed that iron repletion resulted in significant increase in TBARS and formation of protein carbonyls (Fig.1) and degraded DNA on electrophoresis. The mucosal cell turnover number was lowered significantly (Fig.2). Activities of enzymes of intestinal functions were significantly lowered in D^+ compared to C. The higher catalase and lower glutathione peroxidase activities were found to be in the directions of enhanced oxidative stress in D^+. Iron deficiency and its correction had no effect on other antiperoxidative systems measured. Higher mucosal ferritin and lower serum ceruloplasmin ferroxidase activity impaired the mobilization of intestinal iron. This contributed to greater peroxidative stress in the intestine of iron supplemented rats[21]. Transmission electron microscopic studies of D^+ rat duodenum showed reduction in height and complete erosion of microvillus. These effects were shown to be mediated through iron induced hydroxyl radicals produced within the micro-environment of the GI tract by electron paramagnetic resonance spectroscopy[22].

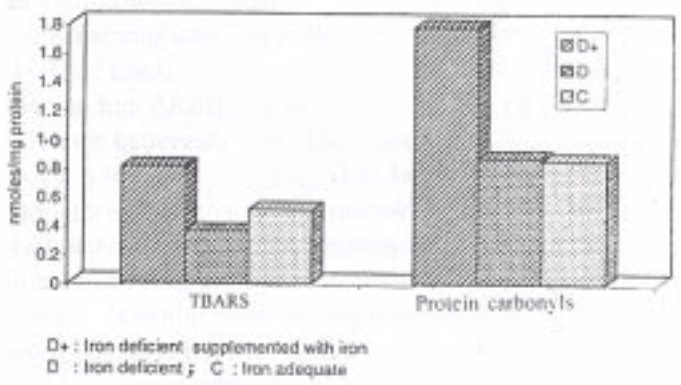

Fig.1. Effect of iron repletion on TBARS and protein carbonyl levels in the intestinal mucosa of iron deficient rats.

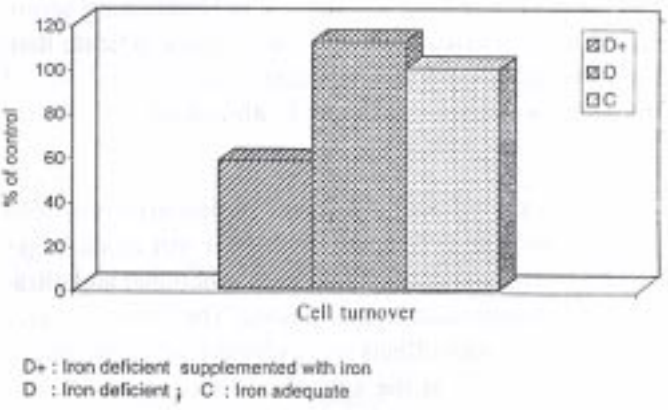

Fig.2. Intestinal mucosal cell turnover during iron repletion in iron deficient rats.

In contrast to the above findings, the effect of excess iron in the GI tract of rats fed with stock diet (natural ingredients) suggested that the GI tract is protected against iron mediated lipid peroxidation. This is attributed partly to the positive balance of antiperoxidative system operating in the GI tract and the antioxygenic principles of natural ingredients used in the stock diets of rats[23]. In view of the above findings, it was felt essential to devise strategies that could prevent oxidative damage to intestine during iron repletion.

Protective role of μ-tocopherol and ascorbic acid

Oral administration of vehicle (D) or 8 mg of iron (D^+) or in the presence of 40 mg of μ-tocopherol (D^++E) or 24 mg of ascorbic acid (D^++C) or a combination of both (D^++C+E) per day for 15 days was carried out in iron depleted rats, and the protective role studied. The results are summarised below.

The indicators of iron status were normalised on iron supplementation either with or without antioxidants. However, in D^++C group the haemoglobin regenerated was greater than D^++E group. The liver iron concentrations were higher in the D^++C group than the other supplemented groups (Table I). Groups which received vitamin E (D^++E and D^++C+E) had lower levels of TBARS and protein carbonyls in the GI tract (Table II). Degraded mucosal cell DNA was observed in D^+ and D^++C and to a lesser extent in D^++E groups. Normalisation of functional integrity was observed in all the groups treated with antioxidant(s). The alterations in endogenous antiperoxidative system in antioxidant supplemented groups were minimal. Serum ascorbate levels were significantly lowered in D^++C group, possibly due to the oxidation of this in the presence of iron (Table II). But in the presence of μ-tocopherol, serum ascorbate levels were significantly elevated suggesting the protection offered by μ-tocopherol in maintaining serum ascorbate level effectively. These findings indicate that, when iron deficient rats are repleted with high doses of iron along with vitamins C and E, almost all key adverse effects are reversed[22,24].

These studies have shown that repletion of iron deficient rats with iron promotes oxidative stress, damages the absorptive cells and brings about functional and ultrastructural derangements in the intestine. The causative factor responsible for such effects was hydroxyl radical produced by the excess iron at the site of iron absorption. Combined supplementation of μ-tocopherol and ascorbic acid protected the GI tract of iron deficient rats against iron mediated free radical damage during repletion.

Table I. Effect of supplementation of iron and antioxidants on haemoglobin, liver and serum iron levels[24]

Treatment	Hb (g/l)	Liver iron (μg/g tissue)	Serum iron (μg/100 ml)
Con	152^{a}±1.5 (9)	179^{a}±15.3 (6)	387^{a}±35.7 (5)
D	79^{b}±2.4 (9)	81^{b}±10.8 (6)	174^{b}±13.8 (5)
D^+	$147^{c,e}$±1.1 (9)	364^{c}± 7.9 (5)	449^{a}±16.1 (5)
D^++C	$151^{a,c,d}$±1.7 (9)	474^{d}±25.9 (7)	402^{a}±25.1 (6)
D^++E	146^{e}±0.8 (10)	378^{c}±41.9 (6)	257^{c}±25.6 (6)
D^++C+E	$148^{a,c,e}$±1.4 (9)	369^{c}±35.8 (6)	279^{c}±28.0

Superscript no. refers to the serial no. in the reference list

Values are Mean ± SEM (n)

Values with different superscript letters within a given column differ significantly by ANOVA ($P<0.05$)

Con: Control; D : Iron deficient; D^+ : Iron deficient supplemented with iron; D^+ + C : Same as D^+ plus vitamin C; D^++E : Same as D^+ plus vitamin E; D^++C+E : Same as D^+ plus vitamins C and E; Hb: haemoglobin.

Table II. Effect of antioxidants on lipid peroxidation, protein oxidation, serum ascorbate and μ-tocopherol levels during iron supplementation in iron depleted rats[24]

Treatment	TBARS (nmol MDA/ mg protein)	Protein carbonyls (nmol/mg protein)	Serum ascorbate (μmol/l)	Serum μ-tocopherol (μmol/l)
Con	0.82^{a}±0.09 (8)	$0.96^{a,e}$±0.11 (8)	$34^{a,c}$±6.4 (7)	$2.4^{a,c}$±0.41 (4)
D	0.56^{b}±0.07 (8)	$0.95^{a,b}$±0.09 (8)	$26^{a,b}$±3.5 (7)	2.9^{a}±0.40 (4)
D^+	1.63^{c}±0.09 (7)	2.06^{c}±0.21 (7)	17^{b}±2.1 (7)	4.9^{b}±0.37 (4)
D^++C	1.72^{c}±0.12 (7)	2.12^{c}±0.31 (7)	$26^{a,b}$±2.2 (8)	0.72^{c}±0.14 (4)
D^++E	1.21^{d}±0.06 (7)	1.02^{d}±0.15 (7)	$47^{c,d}$±6.3 (8)	6.1^{b}±0.82 (6)
D^++C+E	1.37^{d}±0.09 (7)	$1.16^{d,e}$±0.08 (7)	53^{d}±9.0 (7)	5.6^{b}±0.74 (4)

Superscript no. refers to the serial no. in the reference list

Values are Mean ± SEM (n)

Values with different superscript letters within a given column differ significantly by ANOVA ($P<0.05$).

Con: Control; D : Iron deficient; D^+ : Iron deficient supplemented with iron; D^+ + C : Same as D^+ plus vitamin C; D^++E : Same as D^+ plus vitamin C; D^++C+E : Same as D^+ plus vitamins C and E; TBARS: thiobarbituric acid reactive substances.

Conclusions

A critical element of the health care system is the health of women in the child bearing age and children under five. As per the1991 census, 56% of the population in India fall under this category. Anaemia, chronic undernutrition and complications during pregnancy and child birth are the orders of priority for tackling maternal health. In the case of children, the priorities are diarrhoeal diseases, anaemia, perinatal disorders and vitamin A deficiency. Effective antenatal care, prophylactic iron and folic acid supplements, food security, *etc* are some of the measures that would reduce these problems. NIN's research efforts have been directed towards reducing the iron deficiency in these high risk segments of the population.

As intestinal mucosa remains tuned to the body requirements of iron, the findings reported here have important implications with respect to the treatment of iron deficiency anaemia. The increased expression of transferrin receptor at the site of iron absorption in deficient conditions can functionally increase iron absorption. This is achievable only when saturating amounts of iron are present in the intestinal lumen. However, when therapeutic doses of iron are given the concentration of mucosal ferritin increases and blocks the further entry of iron into the mucosa. In this context, administration of iron intermittently rather than daily is a useful strategy.

The studies show that when iron deficient rats are repleted with a high dose of iron along with the antioxidant vitamins C and E, almost all the intestinal oxidative stress is reduced. But vitamin C alone with iron is not effective to reduce the oxidative stress. On the other hand, it enhances the adverse effects of large doses of iron. This is a very significant observation. Therefore, to reduce the oxidative stress in humans, it is suggested that daily administration of therapeutic doses of iron must be accompanied by supplementation with combination of ascorbic acid and μ-tocopherol.

A diet containing high amounts of these vitamins is recommended. A dietary intake of 3.5-5.0 mg of μ-tocopherol and 25-30 mg of ascorbic acid per 1000 Kcal appears to be a healthy goal for all family members. Vegetable oils, like soyabean, safflower and corn seed oils, nuts, wheat germ and whole grain are rich sources of vitamin E. Fresh fruits like *amla* (Indian gooseberry), guava, citrus fruits, banana and certain vegetables such as tomatoes are the main sources of ascorbic acid. Another approach could be fortification of suitable food vehicles like common salt and wheat flour with iron. Strategies involving biotechnological approaches of introducing genes that could accumulate iron (ferritin gene) or reduce the inhibitory effects of phytate (phytase gene) need to be explored to overcome iron deficiency anaemia. This would go a long way in improving the iron nutrition of vulnerable groups of our population.

References

1. Seshadri, S. Nutritional anaemia in south Asia. In: *Malnutrition in South Asia. A Regional Profile.* Ed.S.Gillespie, UNICEF Regional Office for South Asia, Kathmandu, Publication No.5, p75, 1997.
2. Gomber, S., Kumar, S., Rusia, U., Gupta, P., Agarwal, K.N. and Sharma, S. Prevalence and etiology of nutritional anaemia in early childhood in an urban slum. *Indian J Med Res 107:* 269, 1998.
3. Damodaran, M., Naidu, A.N. and Sarma, K.V.R. Anaemia and morbidity in rural preschool children. *Indian J Med Res 69:* 448, 1979.
4. *Studies on Preschool Children*. Technical Report Series, No18, Indian Council of Medical Research, New Delhi, 1984.
5. Prema, K., Neela Kumari, S. and Ramalakshmi, B.A. Anaemia and adverse obstetric outcome. *Nutr Rep Int 23:* 637, 1981.
6. Prema, K., Ramalakshmi, B.A., Madhavapeddi, R. and Babu, S. Immune status of anaemic pregnant women. *Br J Obstet Gynaecol 89:* 222, 1982.
7. Seshadri, S. and Gopaldas, T. Impact of iron supplementation on cognitive functions in preschool children and school aged children. The Indian experience. *Am J Clin Nutr 50(suppl):* 675, 1989.
8. *25 Years of National Nutrition Monitoring Bureau.* Natinal Institute of Nutrition (Indian Council of Medical Research), Hyderabad. p12, 1995.
9. Narasinga Rao, B.S. Studies on iron deficiency anaemia. *Indian J Med Res 58:* 58, 1978.
10. *Nutrient Requirements and Recommended Dietary Allowances for Indians* (*A Report of the Expert Group of the ICMR).* Indian Council of Medical Research, New Delhi, p68, 1990.
11. *Prophylaxis against Nutritional Anaemia among Mothers and Children*. Technical Information, MCH No. 1. Ministry of Health and Family Welfare, Government of India, New Delhi, p3, 1970.
12. *Evaluation of National Nutritional Anaemia Prophylaxis Programme*. (Report of a Task Force Study), Indian Council of Medical Research, New Delhi, 1989.
13. *Field Supplementation Trial in Pregnant Women with 60 mg, 120 mg and 180 mg of Iron with 500 μg of Folic Acid*. (Report of a Task Force Study), Indian Council of Medical Research, New Delhi, 1992.
14. Madhavan Nair, K., Brahmam, G.N.V., Ranganathan, S., Vijayaraghavan, K., Sivakumar, B. and Krishnaswamy, K. Impact evaluation of iron and iodine fortified salt. *Indian J Med Res 108:* 203,1998.
15. Kang, L.X., Xhao, J.L. and Viteri, F.E. Intermittent iron supple-

mentation in Chinese preschool children is efficient and safe. *Food Nutr Bull 16:* 139,1995.

16. Ridwan, E., Schultink, W., Dillon, D. and Gross, R. Effects of weekly iron supplementation on pregnant Indonesian women are similar to those of daily supplementation. *Am J Clin Nutr 63:* 884, 1996.
17. Steer, P., Alam, A.M., Wadsworth, J. and Welch, A. Relationship between maternal haemoglobin concentration and birth weight in different ethinic groups. *BMJ 310:* 489, 1995.
18. Zhou, L.M., Yang, W.W., Hua, J.Z., Deng, C.Q., Tao, X. and Stoltzfus, R.J. Relationship of haemoglobin measured at different times in pregnancy to preterm birth and low birth weight in Shanghai, China. *Am J Epidemiol 148*: 998, 1998.
19. Slivka, A., Kang, J. and Cohen, G. Hydroxyl radicals and the toxicity of oral iron. *Biochem Pharmacol 35:* 553, 1986.
20 Vasantha Lakshmi, K. *Interactions amongst Ferritin, Transferrin and other Iron Binding Proteins during Iron Absorption*. Ph.D Thesis. Osmania University, Hyderabad 1998.
21. Srigiridhar, K. and Madhavan Nair, K. Iron deficient intestine is more susceptible to peroxidative damage during iron supplementation in rats. *Free Rad Biol Med 25:* 660,1998.
22. Srigiridhar, K. *Effect of Excess Iron in the Gastrointestinal Tract of Rat*. Ph.D Thesis, Osmania University, Hyderabad 1998.
23. Srigiridhar, K. and Madhavan Nair, K. Protective effects of antioxidant enzymes and GSH *in vivo* on iron mediated lipid peroxidation in gastrointestinal tract of rat. *Indian J Biochem Biophys 34:* 402, 1997.
24. Srigiridhar, K. and Madhavan Nair, K. Supplementation with μ-tocopherol and ascorbic acid protects the gastrointestinal tract of iron deficient rats against iron induced oxidative damage during iron repletion. *Br J Nutr* (In Press), 2000.

ICMR PUBLICATIONS

	Price(Rs.)
Nutritive Value of Indian Foods (1985) by C. Gopalan, B.V. Rama Sastri and S.C. Balasubramanian, Revised and Updated (1989) by B.S. Narasinga Rao, Y.G. Deosthale and K.C. Pant (Reprinted 1999)	30.00
Growth and Physical Development of Indian Infants and Children (1972, Reprinted 1989)	10.00
Studies on Weaning and Supplementary Foods (1974, Reprinted 1996)	15.00
A Manual of Nutrition (Second Edition 1974, Reprinted 1995)	6.00
Low Cost Nutritious Supplements (Second Edition 1975, Reprinted 1996)	5.00
Menus for Low-Cost Balanced Diets and School-Lunch Programmes Suitable for North India (Second Edition 1977, Reprinted 1998)	6.00
Menus for Low-Cost Balanced Diets and School-Lunch Programmes Suitable for South India (Fourth Edition 1996)	6.00
Some Common Indian Recipes and their Nutritive Value (Fourth Edition 1977, Reprinted 1998) by Swaran Pasricha and L.M. Rebello	16.00
Nutrition for Mother and Child (Fourth Edition 1994, Reprinted 1996) by P.S. Venkatachalam and L.M. Rebello	11.00
Japanese Encephalitis in India (Revised Edition 1980)	5.00
Some Therapeutic Diets (Fifth Edition 1996) by Swaran Pasricha	7.00
Nutrient Requirements and Recommended Dietary Allowances for Indians (1990, Reprinted 1998)	16.00
Fruits (Second Edition 1996) by Indira Gopalan and M. Mohan Ram	20.00
Count What You Eat (1989, Reprinted 1997) by Swaran Pasricha	15.00
*The Anophelines of India (Revised Edition 1984) by T.Ramachandra Rao	150.00
Depressive Disease (1986) by A. Venkoba Rao	58.00
*Medicinal Plants of India Vol.2 (1987)	136.00
Diet and Diabetes (Second Edition 1993, Reprinted 1998) by T.C. Raghuram,Swaran Pasricha and R.D. Sharma	20.00

Dietary Tips for the Elderly (1992, Reprinted 1997) by Swaran Pasricha and B.V.S. Thimmayamma	5.00
Diet and Heart Disease(1994,Reprinted 1998) by Ghafoorunissa and Kamala Krishnaswamy	30.00
Dietary Guidelines for Indians – A Manual (1998)	23.00
Dietary Guidelines for Indians (1998)	10.00

* 25 per cent discount allowed to individuals

These publications are available on prepayment of cost by cheque, bank draft or postal order (bank and postal charges will be extra) in favour of the Director-General, Indian Council of Medical Research, New Delhi. Money orders are not acceptable. All correspondence in this regard should be addressed to the Chief, Division of Publication and Information, Indian Council of Medical Research, Post Box No.4911, Ansari Nagar, New Delhi-110029 (India).

Statement about ownership and other particulars of the ICMR Research Information Bulletin as required under Rule 8 of the Registration of Newspapers (Central) Rules 1956.

Place of Publication	:	Indian Council of Medical Research Ansari Nagar, New Delhi-110 029.
Periodicity of Publication	:	Monthly
Printer's Name	:	Shri J.N. Mathur
Nationality	:	Indian
Address	:	Press Manager Indian Council of Medical Research, Ansari Nagar, New Delhi-110 029.
Publisher's Name		
Nationality	:	Same as above
Address		
Editor's Name	:	Dr. N. Medappa
Nationality	:	Indian
Address	:	Indian Council of Medical Research, Ansari Nagar, New Delhi-110 029.

I, J.N. Mathur, hereby declare that the particulars given above are true to the best of my knowledge and belief.

Sd/- J.N. Mathur
Publisher

Printed and Published by Shri J.N. Mathur for the Indian Council of Medical Research, New Delhi
at the ICMR Offset Press, New Delhi-110 029
R.N. 21813/71