ASSOCIATION OF DENTAL CARIES WITH IRON DEFICIENCY ANAEMIA

DISSERTATION SUBMITTED FOR THE DEGREE OF

M.D BRANCH VII

(PAEDIATRIC MEDICINE)

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D.R M.G.R MEDICAL UNIVERSITY

CHENNAI, TAMILNADU

CERTIFICATE FROM THE DEAN

This is to certify that the dissertation entitled "**ASSOCIATION OF DENTAL CARIES WITH IRON DEFICIENCY ANAEMIA**" is the bonafide work of **Dr.MEKHA PREM ,**in partial fulfilment of the university regulations of the Tamil Nadu Dr. M.G.R Medical University, Chennai, for M.D Degree Branch VII – PAEDIATRIC MEDICINE examination to be held in May 2019.

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DECLARATION

I**, DR.MEKHA PREM,** solemnly declare that the dissertation titled **"ASSOCIATION OF DENTAL CARIES WITH IRON DEFICIENCY ANAEMIA"** has been conducted by me at the Institute of Child Health and Research centre , Madurai under the guidance and supervision of my unit Chief **PROF.DR.M.BALASUBRAMANIAN, M.D., D.C.H.**

This is submitted in part of fulfillment of the award of the degree of M.D. (Pediatrics) for the May 2019 examination to be held under the Tamil NaduDr. M.G.R Medical University, Chennai. This has not been submitted previously by me for any Degree or Diploma from any other University.

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PLAGIARISM CERTIFICATE

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Research Topic	:	Association of dental caries with Iron deficiency Anaemia
Ethical Committee as on	:	08.04.2019

The Ethics Committee, Madurai Medical College has decided to inform that your Research proposal is accepted.

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INTRODUCTION

Dental caries is a multifactorial chronic non communicable disease caused by cariogenic bacterium Streptococcus mutans.Poor oral health has a significant impact on general health and impairs the quality of life of the individual.The pain, problems with chewing and eating caused by dental caries also cause restriction of daily activities thereby causing millions of school and work hours to be lost each year throughout the world.[1]

In 1983 oral health was declared as part of the Strategy for Health For All and in 1989, World Health Organization endorsed the promotion of oral health as an important part of Health for All by the year 2000.. In addition, World Health Day in 1994 was entirely dedicated to oral health which reflects the importance given to oral health.[2]

The *Global Burden of Disease Study 2016* estimated that oral diseases affected at least 3.58 billion people worldwide, with caries of the permanent teeth being the most prevalent of all conditions assessed. Globally, it is found that 2.4

billion people are suffering from caries of permanent teeth and 486 million children are suffering from caries of primary teeth.[3]

In 2011, the total number of children belonging to the age-group 0-6 years were reported as 158.79 million.[4]Around 50% of them, that is, around 79.4 million are affected by ECC in India.In a meta analysis by Chandrashekar et al 2018, five to six year old males had more dental caries experience than females. Also the meta analysis reported prevalence of dental caries to be high in North India.This could be due to difference in socioeconomic factors. The Global Oral Health Data Bank reports a mean DMFT of 2.97 among 12-year-old for the South East Asian countries. Although India reports a comparatively better mean DMFT of 1.95, it is still higher compared to developed countries in the African, European and Eastern Mediterranean and Western Pacific regions of World Health Organization who report a mean DMFT of 1.06, 1.64, 1.81 and 1.05 respectively. This is attributed to the absence of effective oral health intervention services in India.[5]According to the meta analysis ,the SiC is 3.36 for 5 years which is significantly higher for the age. This denotes that nearly four teeth are affected by dental caries.

Anaemia, which is defined as a low blood haemoglobin concentration, is shown to be a public health problem affecting low-, middle- and high-income countries and has significant impacts on social and economic development.[6] Anaemia resulting from iron deficiency affects cognitive and motor development, and, when it occurs in pregnancy

may result in low birth weight and also increases maternal and perinatal mortality.[7][8] In developing regions, maternal and neonatal mortality were responsible for 3.0 million deaths in 2013 and are said to be important contributors to total global mortality.[9][10]. The 2011 estimates suggest anaemia affects around 800 million children and women. About 42% of anaemia in children would be amenable to supplementation of iron. (The Global Prevalence of Anaemia in 2011).

Despite both dental caries and iron deficiency anemia being public health problems in epidemic proportions, little evidence exists to show an association between both.It would be helpful to the public health authorities to frame health policies merging both oral health and iron deficiency anemia if an association really exists between the two.Therefore, we are at need of studies which finds an association between the two major global problem.Our study aims to determine whether dental caries is associated with iron deficiency anemia.

AIM OF THE STUDY:

The aim of the study is to determine an association between Dental caries and iron deficiency anemia.

REVIEW OF LITERATURE

2013 Schroth et al

This is a study conducted in Canadafrom October 2009 to august 2011.In this study, healthy children <72 months of age with S-ECC were recruited .Age matched caries free group is also recruited .Their serum samples were analysed for serum ferritin, hemoglobin and MCV.children with S-ECC had significantly lower mean haemoglobin levels than controls (109.8 ± 8.7 vs. 121.7 ± 7.6, p<.001)This study concluded that children with S-ECC were more likely to have less hemoglobin,MCV and serum ferritin than caries free chidren.[11]

2017 Buche et al

This is a study conducted in Brazil where 186 healthy children aged 11 to 14 years were examined and divided into caries group and control group.Saliva was collected from each individual by spitting method.Salivary pH was measured with a portable meter. Salivary iron was also measured using colorimetric method.The body iron stores is reflected by salivary iron.Statistically significant differences in salivary iron levelswere found between children with DMFT≥1 and childrenwith DMFT=0 (w=5088, p<0.0001).Thus this study concluded that salivary iron plays an important role in oral health.[12]

Babu and Bhanushali et al 2018

This is a study done in Karnataka,India. The study group included 120 children, hospitalized for uncomplicated medical problems. Blood reports were evaluated to determine serum iron and ferritin levels. Dental caries experience was assessed using deft index. Out of 120 children, 38 children showed low serum iron levels of which 31 children had dental caries and nine out of 15 children in the high serum iron level group showed dental cariesBased on the results, it was concluded that there is an inverse association between serum iron levels and dental caries.[13]

Bansal et al 2016

In this study,on comparison of percentage of children with IDA in S-ECC and control group, it was found that children with S-ECC were more likely to have IDA odds ratio (95% confidence interval): 10.77 (2.0, 104.9)(P = 0.001). In addition to this, S-ECC children were significantly more likely to have low Hb,MCV, and PCV levels (P < 0.001) which imply that S-ECC may be a risk marker for the development

of anemia.This study shows a relation between dental caries and iron deficiency anemia.[14]

`

Abdallah et al 2016

This cross sectional study was done at Saudi Arabia. A total of 160 children with dental caries were divided into 2 groups: nonanaemic and anaemic groups. The prevalence of caries was measured using the dmft index and was compared between the two groups.Children with lower mean hemoglobin levels had higher DMFT scores. This cross sectional study revealed that anaemic children are more prone to dental caries in comparison to healthy children.[15]

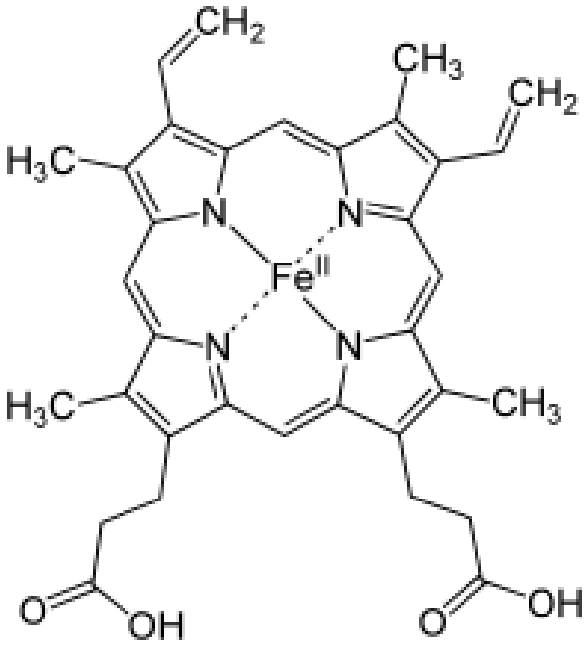
Koppal et al 2013

Sixty children of age 2 to 6 years in whom blood investigations are advised by pediatricians are selected for the study and are divided into early childhood caries(ECC) and control groups according to the def index. After obtaining the informed consent from parent, blood investigations are carried out in these children for the estimation of iron status. All the values depicting the iron status are found

to be decreased in the clinical trial group (ECC group) and they are statistically significant.This study concludes that iron deficiency is observed definitely in children having ECC.[16]

PHYSIOLOGIC CHEMISTRY OF IRON

. Iron lacks the glitter of gold and the sparkle of silver but outshines both in biologic importance.This metal is necessary for the function of many enzymes including catalases, aconitases, ribonucleotide reductase, peroxidases, and cytochromes, The key to the biologic utility of iron is its ability to exist in either of two stable oxidation states: Fe2+ (ferrous) or Fe3+ (ferric). This property permits iron to act as a redox catalyst by reversibly donating or accepting electrons. An excellent example is the electron transport chain of oxidative phosphorylation, in which adenosine triphosphate (ATP) is generated from glucose by the orderly transfer of electrons through a variety of iron-containing mitochondrial cytochromes.



PROTEINS THAT CONTAIN IRON

HEME PROTEINS
- Hemoglobin
- Myoglobin
- Cytochrome *a*, *b*, *c*
- Cytochrome P-450
- Tryptophan 1,2-dioxygenase
- Catalase
- Myeloperoxidase

IRON-DEPENDENT ENZYMES
- Aldehyde oxidase
- Reduced nicotinamide adenine dinucleotide dehydrogenase
- Tyrosine hydroxylase
- Succinate dehydrogenase
- Prolyl hydroxylase
- Tryptophan hydrolase
- Xanthine oxidase
- Ribonucleotide reductase
- Aconitase
- Phosphoenolpyruvate carboxykinase

From Griffin IJ, Abrams SA: Iron and breastfeeding. Pediatr Clin North Am *48:401–413, 2001.*
*Partial list.

When dissolved in aqueous solution, ferrous iron has the capacity to rapidly oxidizeinto ferric iron, which is insoluble at physiologic pH. To achieve stable solubility under physiologic conditions, iron complexes with iron-binding agents termed chelators.Transferrin is a chelator available in human plasma.

Iron also complexes with many small molecules.Heme is an example of iron-protoporphyrin IX complex. A best example of heme protein is hemoglobin in which a globin histidine residue donates the fifth electron and the sixth comes from molecular oxygen.[17] This property helps in the transport of oxygen by hemoglobin to the entire body.

Iron is also necessary for physical growth, neurological development, cellular functioning, and for the synthesis of some hormones.Normal cellular processes like respiration generate reactive oxygen species like superoxide and hydrogen peroxide.Iron in free form may help in converting reactive oxygen species into free radicals via Fenton reaction. [18].So, iron has to be sequestered with some proteins to abrogate this free radical formation and the consequent lipid peroxidation of cell membrane.Thus ferritin plays a tremendous role in sequestering iron tightly within cell. The expression of ferritin is enhanced by oxidative stress as a protective mechanism.[19]

ACQUISITION AND DISTRIBUTION OF IRON

An adult has an average of 4 to 5 grams of iron in his/her body. Around 0.5 to1 gram of iron is lost each day through the sloughing of cells from skin and

mucosal surfaces.[20] Since liver and kidney lacks the physiologic capability to excrete iron, absorption becomes the only way of regulating body iron stores.[21]

INTESTINAL IRON ABSORPTION

Iron is available in the diet in both heme and non heme iron. Heme iron is predominantly non vegetarian from myoglobin and haemoglobin of animals. Non heme iron is vegetarian in source. Acidity of stomach, ascorbic acid and citrate helps in the absorption of iron. Phytates, tannins inhibit its absorption.[22]Heme iron is not influenced by any external factors since it is tightly sequestered inside a protoporphyrin ring.Iron is absorbed mainly by the mature enterocytes of the proximal part of duodenum.[23]

Dietary iron in ferric (Fe3+) form is reduced to ferrous form (Fe2+) with the help of ferric reductase,an enzyme present in the brush border of the enterocyte.This enzyme is thought to be duodenal cytochrome *b*, a heme protein that is homologous to *b*561cytochromes..[24]Ferrous iron is transported across the apical membrane of enterocyte with the help of a transporter namely DMT1[(Divalent Metal Transporter 1) also known as Nramp2, DCT1, and

SLC11A2][25][26][27].This iron is carried across the basolateral membrane via transporter FPN1(ferroportin also known as SLC40A1, MTP1, and IREG1)[28][29].If iron is not needed by the and body immediately, it will be stored within iron storage protein ferritin within the enterocyte.

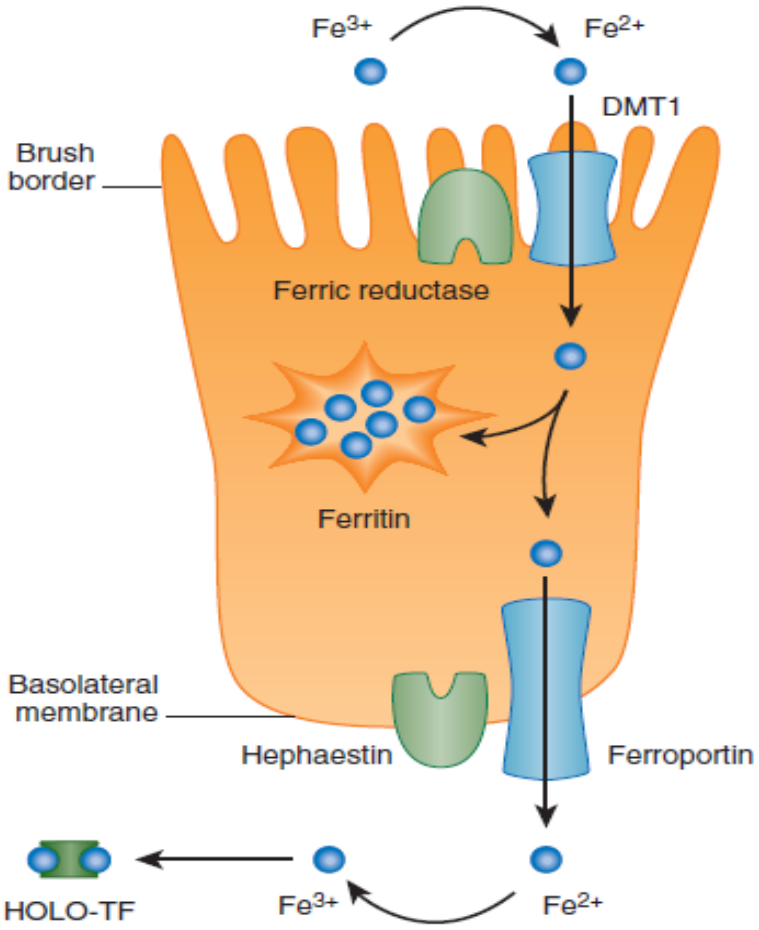


Figure 11-2 Duodenal iron transfer. The microstructure of the iron absorption apparatus is depicted in this cartoon. Iron is taken up by enterocytes lining the duodenal villi. These absorptive cells start out as undifferentiated precursors in the intestinal crypts. Crypt cells appear to be programmed for an iron absorption "set point" that is determined in response to iron needs. As the cells differentiate, they migrate up the villi and begin to express iron transporter proteins. According to current models, nonheme iron uptake occurs in mature enterocytes through the enzymatic reduction of iron, transmembrane import into the cell by DMT1, transmembrane export from the cell by ferroportin, and enzymatic oxidation by hephaestin before loading onto apotransferrin to produce diferric transferrin (HOLO-TF). *(Modified from Andrews NC: Iron homeostasis: insights from genetics and animal models.* Nat Rev Genet *1:208–217, 2000.)*

The mechanism of absorption of heme iron is not definitely known.It is presumed that heme iron binds intact to brush border membrane of the enterocyte.Once inside the enterocyte, it is acted upon by heme oxygenases and

then this iron is carried across the basolateral membrane via FPN1. The export of iron through the basolateral membrane is

greatly enhanced by the presence of hephaestin which is a copper dependent iron oxidase.[30][31]The role of hephaestin is to convert newly transported Fe2+ to Fe3+.

TRANSPORT OF IRON AND DELIVERY OF IRON TO TISSUES

Iron that is absorbed via enterocyte reaches the circulation and is carried in the circulation bound to transferrin. Transferrin is the most important physiologic iron supplier to cells.[32] It is a 80 kd glycoprotein that has homologous N-terminal and C-terminal iron binding domains.[33].Transferrin binds ferric iron avidly with a dissociation constant of approximately 10 to 22 mmol/L.[34] Most of the time, only 30% of iron binding sites on plasma transferrin pools are being occupied, meaning a transferrin saturation of only 30%. This property paves the way for buffering capacity against the appearance of potentially toxic non-transferrin-bound-iron.[35] Transferrin saturation is a useful index of iron supply to the bone marrow. Transferrrin saturation of less than 16% is linked to reduced production of red

blood cells(RBCs)..The sum of all the iron binding sites present in transferrin is the Total Iron Binding Capacity (TIBC) of plasma.Hence, on a molar basis, TIBC equals to twice the transferrin concentration since one molecule of transferrin could bind approximately 2 atoms of iron.

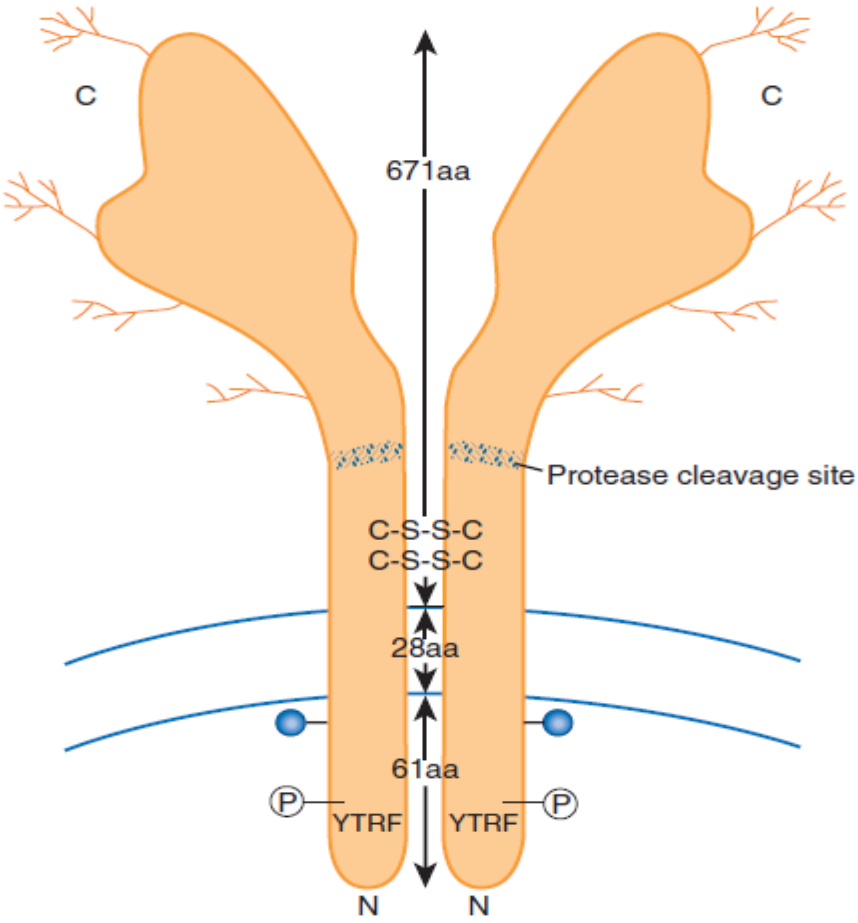


Figure 11-3 Structure of the dimeric transferrin receptor. The N-terminals of both subunits are inside the cell, and the C-terminals are outside. The 61–amino acid *(aa)* intracellular domain has three structural features that appear to play a role in endocytosis: a tyrosine-threonine-arginine-phenylalanine *(YTRF)* amino acid motif, a phosphorylated serine residue *(encircled P)*, and a covalently linked molecule of palmitic acid *(solid circle)*. The transmembrane domain is 28 amino acids long. The extracellular domain has 671 amino acids, including disulfide linkages *(C-S-S-C)*, as well as four glycosylation sites *(branched lines)*. A potential protease cleavage site is located between amino acids 100 and 101.

Diferric transferrin binds to transferrin receptor 1(Tfr) on the plasma membrane of cells. Transferrin-Tfr1 complex is internalized through the clathrin mediated endocytosis.[36] The endosome is acidified and due to this low pH, conformational change in transferrin occurs. The Fe3+ bound to transferrin is released by the action of a family of reductases [6-transmembrane epithelial antigen of the prostate family of reductases (6-transmembrane epithelial antigen of the prostate 3 in the case ofimmature erythroid cells)]. This iron is transported across the endosomal membrane via transporter DMT1 into the cytoplasm of the cell.[37][38] If iron is needed by the body, it is taken to the mitochondria.If it is not needed,it is stored in the form of ferritin. This iron may find its way out of the cell through FPN1 which acts as as a safety pop off valve in cases of iron overload conditions. The efficiency of iron export via basolateral membrane is enhanced by hephaestin homolog ceruloplasmin in most body cells unlike enterocytes.Iron is reduced to ferrous form by STEAP3.[39] The iron released is shuttled to mitochondria by mitoferrin 1 (MFRN1) for synthesis of heme.[40] The excess iron is stored in the form of ferritin within the cell. The apotransferrin-transferrin receptor complex recycles to the cell surface, where neutral pH promotes the release of apotransferrin into serum for further reuse later.[41][42][43]

The endocytic transferrin cycle is depicted in the figure below

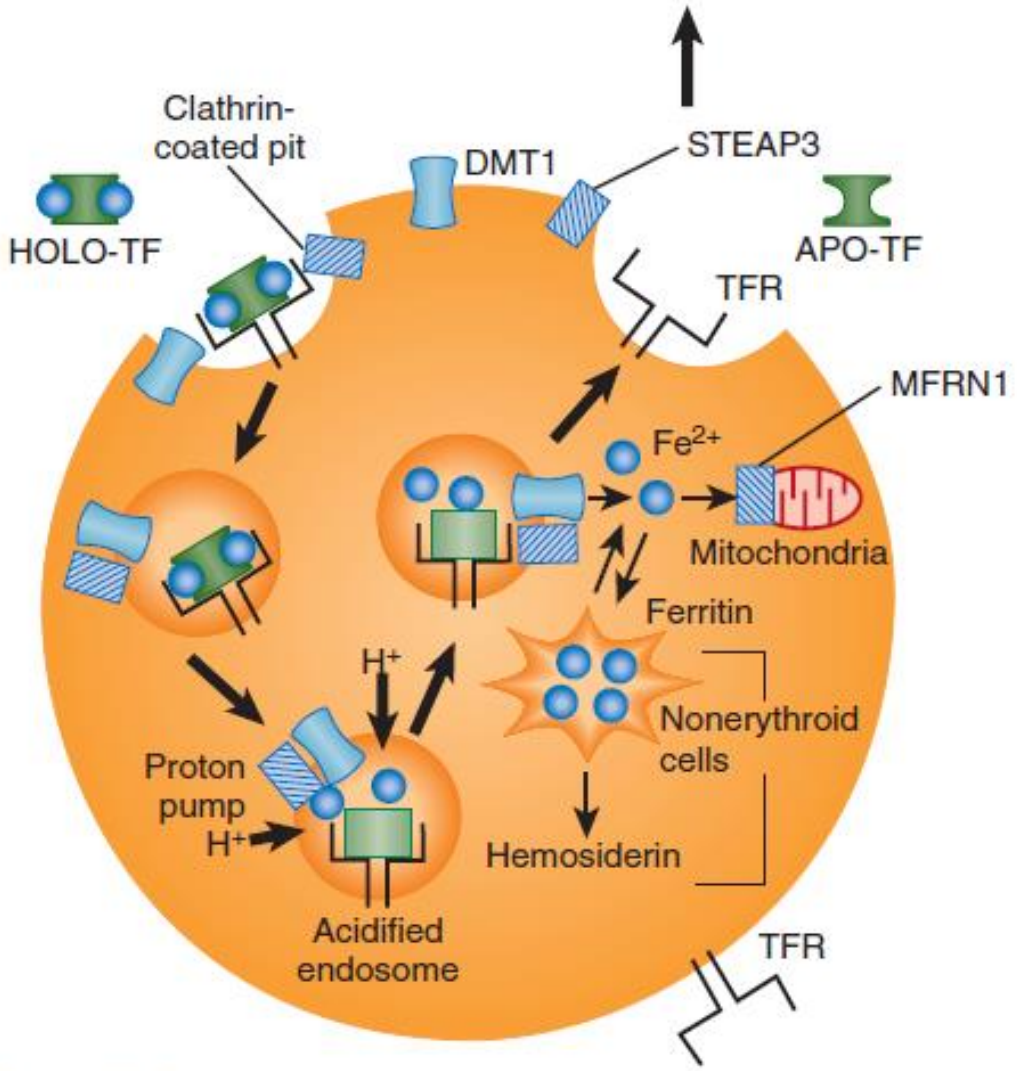


Figure 11-4 The endocytotic transferrin cycle. Apotransferrin *(APO-TF)* binds two atoms of iron per molecule to form diferric transferrin *(HOLO-TF)*. Diferric transferrin binds to the transferrin receptor *(TFR)* on the cell surface. The complex is internalized by invagination of clathrin-coated pits to form specific endosomes. The endosomes import protons, thus lowering the pH within the organelle and decreasing the affinity of transferrin for iron. Liberated iron is translocated through the endosome membrane to the cytoplasm by DMT1 after iron has been reduced from the ferric to the ferrous form by STEAP3. The iron released is shuttled to the mitochondria by mitoferrin 1 (*MFRN1*) for synthesis of heme and to ferritin for storage. The apotransferrin-transferrin receptor complex recycles to the cell surface, where neutral pH promotes release of apotransferrin into serum for reuse. Details are given in the text. *(Modified from Andrews NC: Iron homeostasis: insights from genetics and animal models.* Nat Rev Genet *1:208–217, 2000.)*

Transferrin is not the only source of cellular iron. Cells make use of non transferrin bound iron also. Nonhematopoietic tissues (particularly the liver, endocrine organs, kidneys, and heart) mainly take up NTBI. It has been shown recently that Zrt/Irt-like protein 14 is an important NTBI transporter.[44] It is surprising that even ferritin delivers its iron to cells.[45]

In pathological situations like hemolysis, heme and hemoglobin released from RBCs bind to hemopexin and haptoglobin respectively.These complexes are taken up by cells and used in the salvage of iron.[46] Iron uptake by various cells operate through various pathways. For example, liver makes use of different pathways for iron uptake whereas immature erythroid cells make use of transferrin-Tfr1 mediated endocytosis alone.[47]

INTRACELLULAR IRON TRAFFICKING AND STORAGE

Free iron is toxic in nature.Hence it binds to chaperones which helps iron move inside the cell.[48] Recently, poly(rC)-binding proteins are identified as intracellular iron chaperones.[49] Ferritin is considered as a major intracellular storage form of iron. Ferritin is a large protein made up of 24 subunits arranged in the form of a spherical shell with a large cavity. Iron can enter inside and come

outside of the shell via the pores in the ferritin molecule. One single molecule of ferritin can hold <=4500 atoms of iron. When large concentrations of iron-laden ferritin accumulate within the cell, the ferritin molecules aggregate, and fuse with lysosomes.As a result of this process,ferritin gets degraded and mixture ofFe3+ cores and peptides are formed which is known as hemosiderin. Iron is mobilized from both ferritin and hemosiderin when the body is requiring iron to perform its functions. Ferritin is also secreted by the cell in small amounts which reflects the intracellular concentrations of iron.Thus ferritin concentrations serve as an accurate indicator of body iron stores.[50]

REGULATION OF CELLULAR IRON HOMEOSTASIS

Iron homeostasis in the cell is regulated in such a way as to maximize the iron supply when the cell is deficient in iron and to restrict iron supply when the cell has excess of iron. One such example is the iron-dependent binding of iron regulatory proteins(IRPs) IRP1 and IRP2 to stem-loop structures [iron-responsive elements (IREs)]present in the untranslated regions (UTRs) of the messenger RNAs (mRNAs) encoding various iron-related proteins.The IRPs exist in the mRNA binding conformation when cellular iron content is low.IRPs bind to the 5’ untranslated region of ferritin mRNA and blocks its translation producing little or

no ferritin preventing storage of iron when it is needed by the body. At the same time,IRPs bind to the 3' UTR of TfR1 mRNA and protects the message for translation and hence more transferrin receptors are (produced.These transferrin receptors get expressed on the cell membrane and help in the entry of iron into cell. When cell is iron replete, the opposite occurs. That is ferritin gets translated and more ferritin is produced which helps in the storage of iron. At the same time, TfR1 is degraded thus limiting cellular iron uptake and promoting storage.[51]

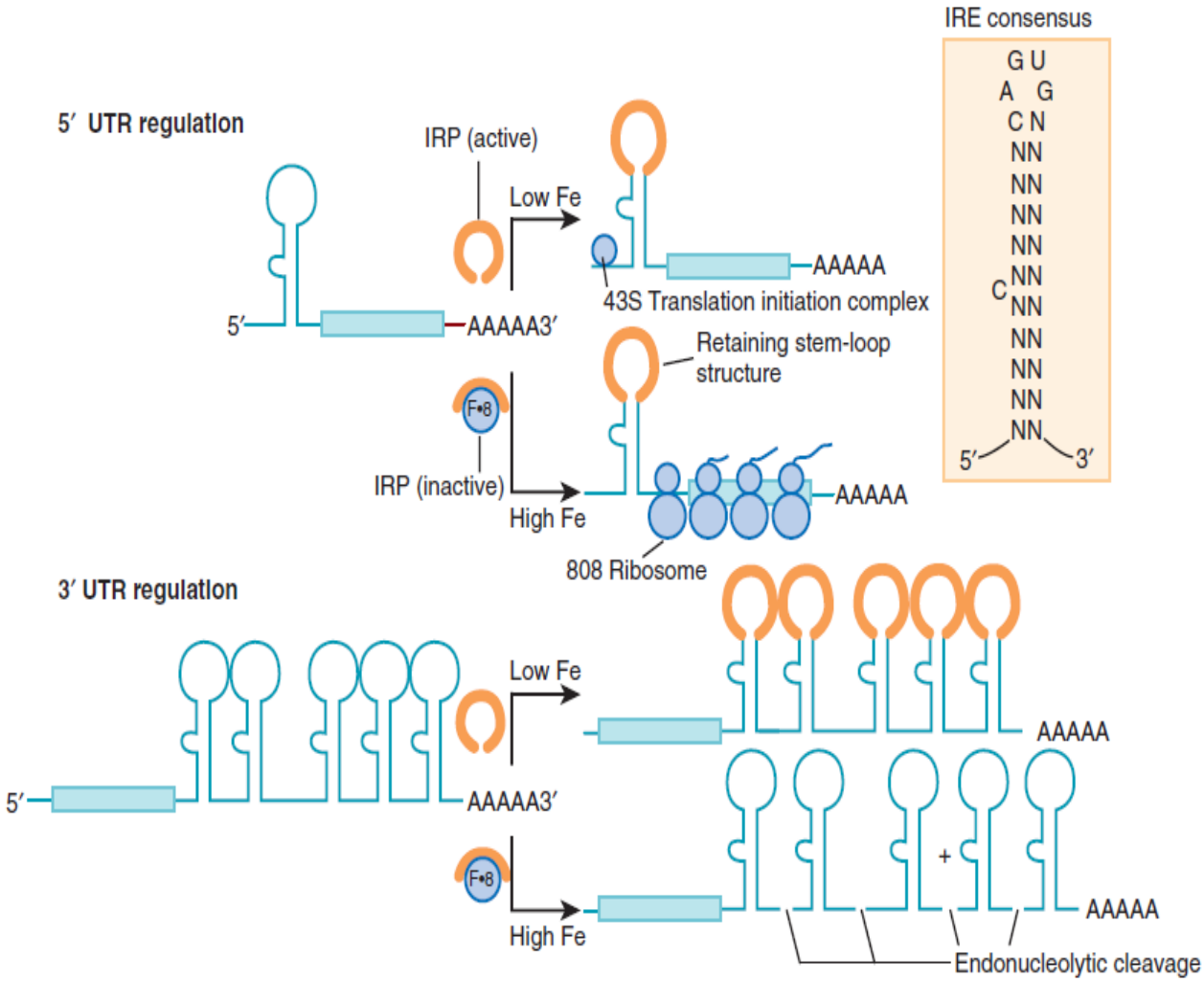


Figure 11-5 Iron response element *(IRE)*/iron regulatory proteins *(IRP)* regulation. Two mechanisms of action of IRPs are shown. An IRP molecule binds to an IRE stem-loop structure located in the untranslated regions *(UTRs)* of messenger RNA sequences. A consensus IRE structure is shown in the *inset*. Under low-iron or heme conditions, IRPs bind avidly to RNA. With abundant iron, no binding occurs. Binding of IRP to IRE elements in the 5′ UTR blocks translation. In contrast, binding of IRPs to 3′ UTR IRE elements protects messenger RNAs from degradation.

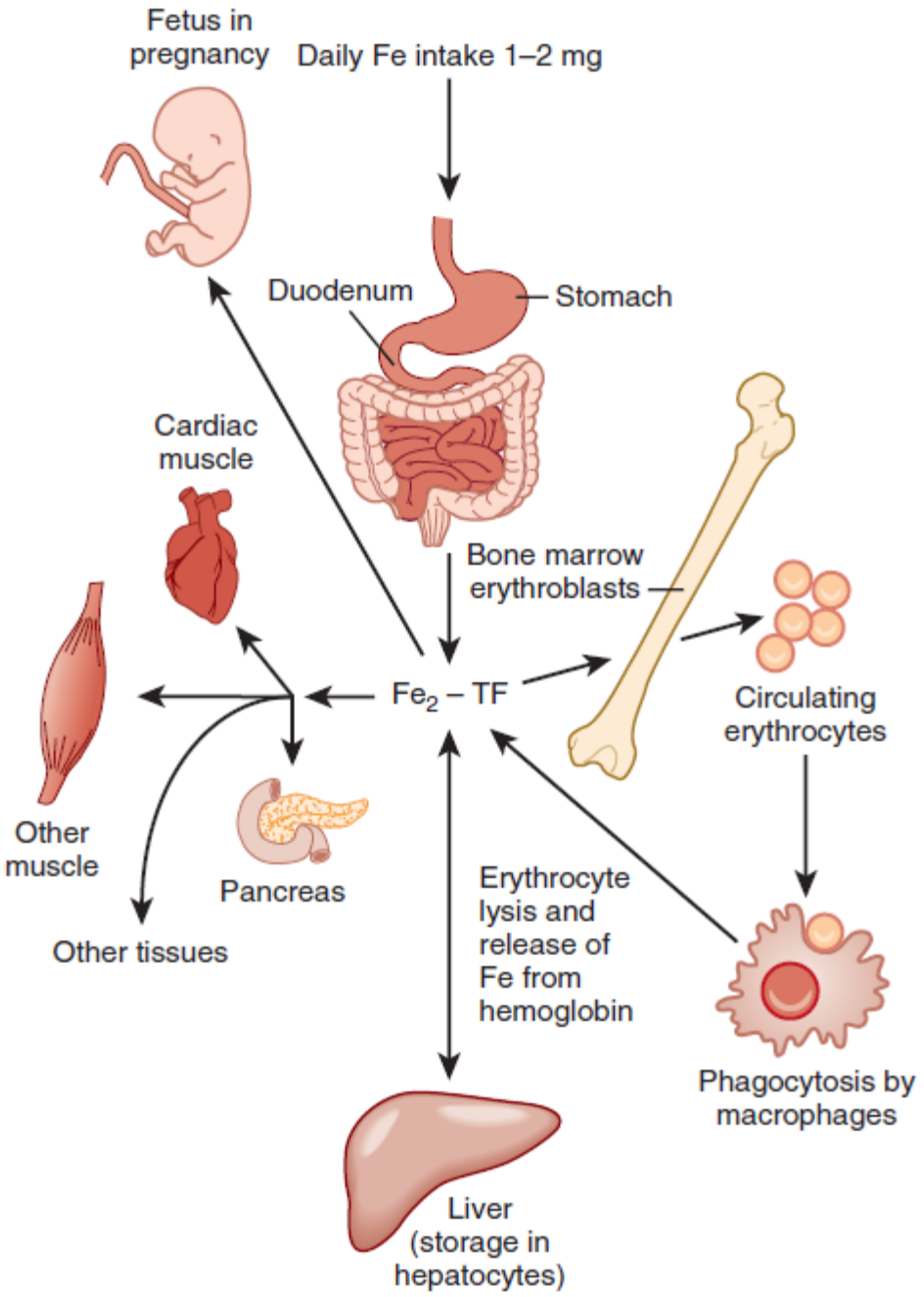


Figure 11-1 The body's iron economy. Although the average adult has 4 to 5 g of body iron, only 1 mg of dietary iron enters and leaves the iron economy on an average day. Dietary iron enters through the duodenum and becomes bound to plasma transferrin for delivery to tissues. The erythron is the largest site of iron use, but all cells require the metal. Storage iron is found primarily in the liver. Reticuloendothelial macrophages carry out iron recycling. Iron is lost from the body with bleeding and with exfoliation of skin and mucosal cells. *(Modified from Andrews NC: Iron homeostasis: insights from genetics and animal models.* Nat Rev Genet *1:208–217, 2000.)*

SYSTEMIC IRON HOMEOSTASIS

The changes in body iron demand are communicated to the liver,which responds by modulating the expression of hepcidin. Hepcidin is a 25–amino acid peptide produced in the liver from a larger precursor (reviewed by Nemeth and Ganz) [52]Hepcidin binds to the iron export protein FPN1.Hence FPN1 is rightly called the hepcidin receptor.This complex is ubiquitinated, internalized, and degraded thus stopping the export of iron from cells to circulation.Hepcidin levels are high when body is iron replete thus reducing the iron supply to circulation.Hepcidin levels are low when body is iron deficient thus providing the iron to circulation.[53] Major iron exporting cells are macrophages, hepatocytes and intestinal enterocytes. Increased iron stores and inflammation are the factors that enhance hepcidin expression, whereas reduced iron stores and hypoxia play a role in lowering expression.[54][55][56]Only the inflammatory cytokine interleukin-6 has been surely shown to be involved in the regulation of hepcidin expression by physiologic changes occurring outside the liver.[57][58][59]

Various organs have varying iron requirements.Erythroid marrow needs highest iron for production of young RBCs. Cells with highest proliferation rate like intestinal crypts also need more iron. Thus when iron supply is limited, the organs with high iron requirement are well protected at the expense of others.[60]

IRON DEFICIENCY ANEMIA

Iron deficiency is the most common nutritional deficiency in theworld.30% of the global population has iron deficiency anaemia.[61] A full term newborn infant has about 0.5 grams of iron in comparison with adults (5 grams of iron).This puts the baby in a situation wherein it has to absorb 0.8 mg of iron daily during the first 15 years of life.A small extra amount is also needed to compensate for the iron lost during shedding of cells.As a result, it is necessary to absorb 1 mg daily during childhood to maintain positive iron balance.Since <10% of dietary iron is usually absorbed, a daily iron requirement of 8 to 10 mg is usually required.[62]

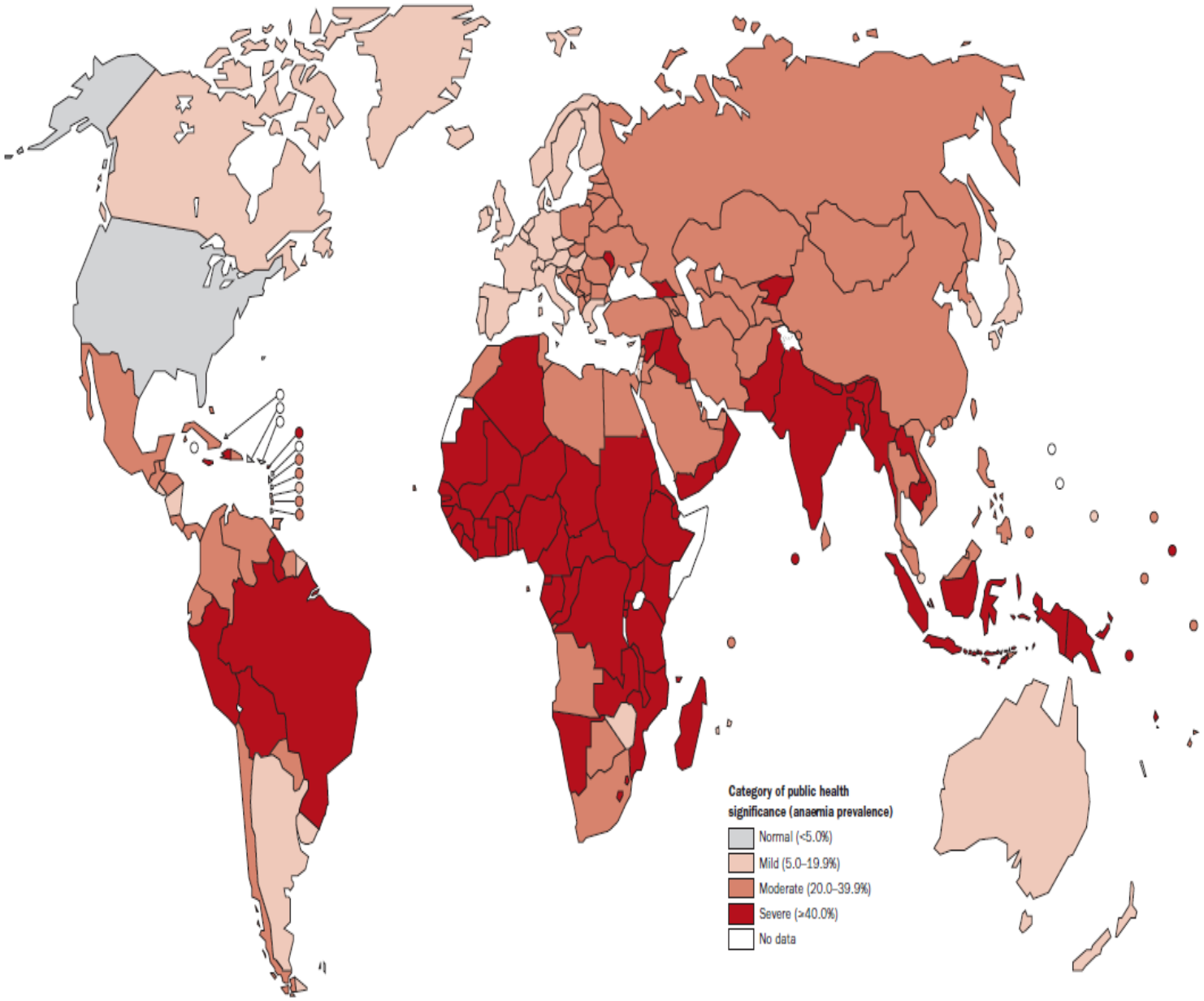
Table 2.3 ***Classification of anaemia as a problem of public health significance***

Prevalence of anaemia (%)	Category of public health significance
≤4.9	No public health problem
5.0–19.9	Mild public health problem
20.0–39.9	Moderate public health problem
≥40.0	Severe public health problem

Source: Iron deficiency anaemia: assessment, prevention, and control. A guide for programme managers. Geneva, World Health Organization, 2001 (WHO/NHD/01.3).

PRE SCHOOL AGE CHILDREN:

PROBLEM STATEMENT OF ANEMIA



ETIOLOGY OF IRON DEFICIENCY ANAEMIA

Most of the iron in neonates is in circulating haemoglobin.When the high hemoglobin concentration in a newborn falls during the first 2 to 3 months of age due to lysis of red blood cells, the iron from hemoglobin is recycled. These iron stores will be sufficient for the infant for erythopoiesis during the first 6 to 9 months.Iron stores are depleted faster in a baby born low birth weight or a baby who has suffered perinatal blood loss. Delayed cord clamping(1 to 3min) improves iron status and protects against iron deficiency anemia whereas early cord clamping(<30 seconds) places the baby at risk for iron deficiency anemia.[63]

.

Iron deficiency anaemia is also caused by insufficient dietary intake.It is mainly caused by consumption of cows milk in the first year of life.Cows milk contains caseinophosphopeptide which interferes with the absorption of iron.Its iron content and bioavailabilty is also low.[64]Cows milk can produce occult blood loss in stools due to allergic colitis.[65] This can be prevented by prolonged breast feeding, delaying the introduction of cows milk till 1 year and limiting intake of cows milk to 24 ounces/day thereafter.[66][67]

Iron deficiency anaemia is also caused lesion of gastrointestinal tract with occult or frank bleeding such as peptic ulcer,Meckels diverticulum, polyp and Inflammatory

Bowel Disease.[68]Infections with hookworm,Trichuris trichura, Plasmodium and Helicobacter pylori underlie causes of iron deficiency anaemia in developing countries.[69][70]Celiac disease and giardiasis also contribute to iron deficiency anaemia by interfering with iron absorption.[71]

CLINICAL MANIFESTATIONS OF IRON DEFICIENCY ANAEMIA

Most chidren with iron deficiency are asymptomatic and are identified through recommended lab screening at 1 year of age.Pallor which is the important sign of iron deficiency is visible only when the hemoglobin drops to 7 to 8 g/dl.Pallor is noted in the nail beds,palmar creases or conjunctiva. In children with mild to moderate iron deficiency anaemia (hemoglobin levels of 6 to 10 g/dl),compensatory mechanisms such as increase in 2,3-diphosphoglycerate and a shift in oxygen dissociation curve to the right operates well in hand masking symptoms of anaemia apart from mild irritability.When the hemoglobin drops to <5 g/dl, symptoms like irritability,lethargy and systolic flow murmurs pop out. As

the hemoglobin continues to drop ,tachycardia and high output cardiac failure occurs.[72]

Both iron deficiency and iron deficiency anaemia is associated with impaired neurocognitive function. For example, when Bayley Scales of Infant Development was used for testing , nonanemic iron-deficient infants had lower developmental scores

compared to iron-sufficient infants . Hence minimizing its incidence is essential to curb poor neurocognitive outcome. Any markers to predict the early stage of iron deficiency would be crucial in managing this problem.[73][74]

The mechanism by which iron deficiency impairs neurologic function is not fully elucidated.It is thought that iron deficiency could impair neurotransmitter mechanisms. It has been shown to decrease the expression of dopamine receptors in the rat brain according to one study.[75] It is shown that iron deficiency may also interfere with myelination and alters myelin proteins and lipids in oligodendro-cytes.[76]

PHASES OF DEVELOPMENT OF IRON DEFICIENCY

Prelatent iron deficiency : tissue stores are depleted reflected by low serum ferritin

Latent iron deficiency : reticuloendothelial macrophage iron stores are depleted reflected by decreased serum iron and increase in TIBC

Frank iron deficiency :erythrocyte microcytosis and hypochromia ;

Iron deficiency progresses from depletion of iron stores (mild iron deficiency), to iron-deficiency erythropoiesis (erythrocyte production), and finally to iron deficiency anemia (IDA). With iron-deficiency erythropoiesis (marginal iron deficiency), iron stores are depleted and transferrin saturation starts declining, but hemoglobin levels stay within the normal range. IDA is characterized by low hemoglobin concentrations, and low hematocrit (the proportion of red blood cells in blood by volume) and low mean corpuscular volume (a measure of erythrocyte size) .[77][78]

The American Academy of Pediatrics and U.S. Centers for Disease Control and Prevention recommend the use of Hemoglobin as a screening technique for children at risk of iron deficiency. Since iron deficiency that has not reached the stage of anaemia is associated with neurocognitive impairment, we are at need of

other indicators apart from Hemoglobin. Transferrin saturation less than 10% is considered the gold standard for diagnosing iron deficiency anaemia. Other parameters like serum iron, serum ferritin, TIBC ,MCV,RDW are also used to diagnose the same.

APPENDIX 30 Serum Ferritin, Iron, Total Iron-Binding Capacity, and Transferrin Saturation in Children and Adolescents

Age	Male Subjects		Female Subjects	
Ferritin	ng/mL	µg/L	ng/mL	µg/L
1-30 d*	6-400	6-400	6-515	6-515
1-6 m*	6-410	6-410	6-340	6-340
7-12 m*	6-80	6-80	6-45	6-45
1-5 y†‡	6-24	6-24	6-24	6-24
6-9 y†‡	10-55	10-55	10-55	10-55
10-14 y†	23-70	23-70	6-40	6-40
14-19 y†	23-70	23-70	6-40	6-40
Iron	µg/dL	µmol/L	µg/dL	µmol/L
1-5 y†‡	22-136	4-25	22-136	4-25
6-9 y†‡	39-136	7-25	39-136	7-25
10-14 y†	28-134	5-24	45-145	8-26
14-19 y†	34-162	6-29	28-184	5-33

TABLE 11-1 Common Clinical Laboratory Tests to Assess Iron Status*

Test	What It Indicates	Result in Iron Deficiency	Result in Iron Overload	Confounding Conditions
Iron	Iron recycling/usage, stores (indirect)	↓	↑-↑↑	↓ in inflammation
Total iron-binding concentration (TIBC)	Serum/plasma transferrin	↑	Normal	↓ in inflammation
Transferrin saturation	% plasma iron-binding sites on transferrin that are occupied	↓-↓↓	↑-↑↑	↓ in inflammation
Free or zinc protoporphyrin	Functional mitochondrial iron status	↑	Normal	↑ in reticulocytosis
Soluble transferrin receptor (sTfR)	Erythropoietic mass	↑	Normal	Ineffective erythropoiesis
Ferritin	Iron stores (indirect)	↓-↓↓	↑-↑↑	↑ in inflammation
Bone marrow iron	Macrophage iron stores/retention, red blood cell iron uptake	↓	Normal to ↑	↑ in inflammation
Liver iron	Iron stores	↓-↓↓	↑-↑↑	Secondarily increased in cirrhosis
Hepcidin†	Hepatocyte iron	↓-↓↓	↑-↑↑	↑ in inflammation

*All measures are in serum or plasma unless otherwise indicated.
†Hepcidin is not yet routinely available as a clinical test.

Table 447-1 Normal Mean and Lower Limits of Normal for Hemoglobin, Hematocrit, and Mean Corpuscular Volume

	HEMOGLOBIN (g/dL)		HEMATOCRIT (%)		MEAN CORPUSCULAR VOLUME (μM^3)	
Age (yr)	Mean	Lower Limit	Mean	Lower Limit	Mean	Lower Limit
0.5-1.9	12.5	11.0	37	33	77	70
2-4	12.5	11.0	38	34	79	73
5-7	13.0	11.5	39	35	81	75
8-11	13.5	12.0	40	36	83	76
12-14 female	13.5	12.0	41	36	85	78
12-14 male	14.0	12.5	43	37	84	77
15-17 female	14.0	12.0	41	36	87	79
15-17 male	15.0	13.0	46	38	86	78
18-49 female	14.0	12.0	42	37	90	80
18-49 male	16.0	14.0	47	40	90	80

From Brugnara C, Oski FJ, Nathan DG: Nathan and Oski's hematology of infancy and childhood, *ed 7, Philadelphia, 2009, WB Saunders, p. 456.*

LABORATORY FINDINGS

A sequence of biochemical and hematological events occur in progressive iron deficiency anaemia.Initially,tissue iron stores are depleted reflected by decrease in serum ferritin.But serum ferritin is an acute phase reactant elevated in inflammation.Hence inflammation has to be ruled out using CRP.[79]

Then, serum iron levels decrease, TIBC (Total Iron Binding Capacity) increase and Transferrin saturation falls below normal.Transferrin saturation is 100% sensitive and specific for diagnosis of iron deficiency anaemia.(Nathan and Oski)

Since the iron stores start decreasing, iron is not available to bind with protoporphyrin to form heme.Hence, free erythrocyte protoporphyrins accumulate and the production of hemoglobin is hampered.This is the point where iron deficiency progresses to iron deficiency anaemia. The absolute level of red blood cell Zinc Proto Porphyrin (ZPP) or, more specifically, the ratio of ZPP to iron protoporphyrin IX(i.e., heme), is increased in iron deficiency.[80]

sTfR is a truncated form of the transferrin receptor which is cleaved from reticulocytes and erythroid cells and circulates in plasma bound to transferrin. sTfR is increased in iron deficiency thereby helping to distinguish iron deficiency anemia from the anemia of inflammation.[81][82]

The cellular hemoglobin of the reticulocyte (CHr) is another test available only in selected laboratories .CHr has been evaluated prospectively as a means of routinely screening infants and toddlers in some studies.[83]

With less hemoglobin in each cell, the cell become microcytic and varied in size contributing to decrease in MCV(Mean Corpuscular Volume) and increase in RDW(Red Cell Distribution Width). Red blood cell count also decreases.White

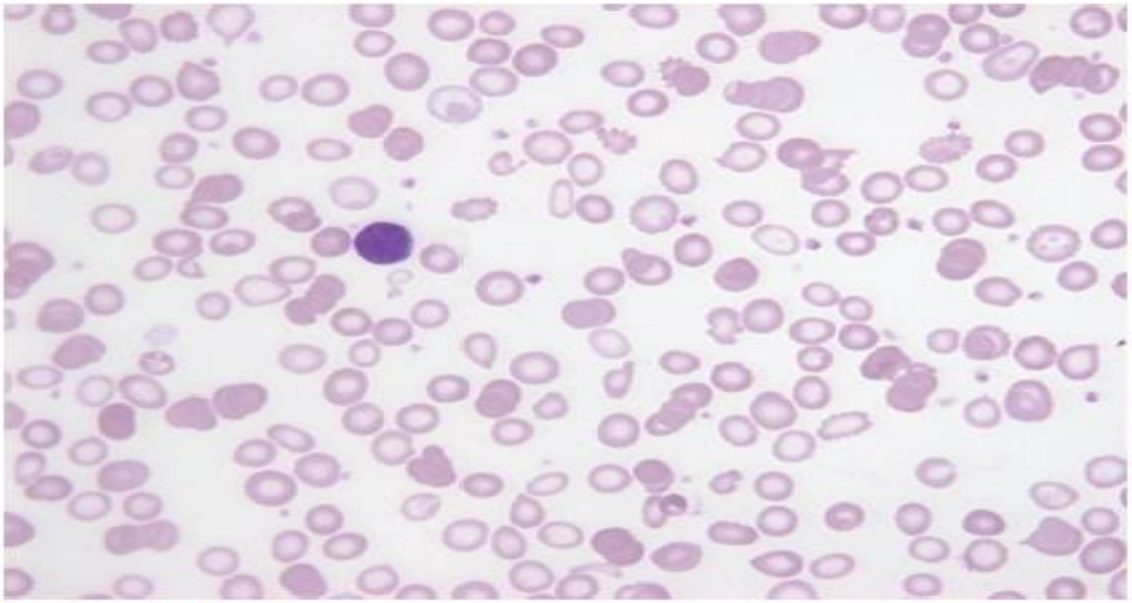


Figure 11-7 Peripheral blood smear in iron deficiency. Note the small, pale (microcytic, hypochromic) red blood cells with variable sizes and shapes (anisopoikilocytosis). Occasional target cells with central hemoglobin pooling as well as several somewhat elongated hypochromic microcytes (pencil cells) are present.

blood cell count is normal and platelet count is elevated(due to homology of erythropoeitin with thrombopoietin).Elongated cells called pencil cells are said to be characteristic of iron deficiency.[80][81][82]

An increase in hemoglobin to >1 g/dl after 1 month of iron therapy is a practical means of diagnosing iron deficiency anaemia.

RECOMMENDED DIETARY ALLOWANCES (RDA) FOR IRON

Age	Male	Female	Pregnancy	Lactation
Birth to 6 months	0.27 mg*	0.27 mg*		
7–12 months	11 mg	11 mg		
1–3 years	7 mg	7 mg		
4–8 years	10 mg	10 mg		
9–13 years	8 mg	8 mg		
14–18 years	11 mg	15 mg	27 mg	10 mg
19–50 years	8 mg	18 mg	27 mg	9 mg
51+ years	8 mg	8 mg		

Source:

Institute of Medicine. Food and Nutrition Board. Dietary Reference Intakes for Vitamin A, Vitamin K, Arsenic, Boron, Chromium, Copper, Iodine, Iron, Manganese, Molybdenum, Nickel, Silicon, Vanadium, and Zinc : a Report of the Panel on Micronutrients. Washington, DC: National Academy Press; 2001

SOURCES OF IRON

Heme iron is rich in lean meat and seafood . Nuts, beans, vegetables, and fortified grain products are rich in non heme iron. Breast milk contains little iron in highly bioavailable form that may not be sufficient to meet the needs of infants older than 4 to 6 months .WHO/FAO (2006) provided recommendations to add iron compounds to specific foods, including cereal products, condiments, milk, and cocoa products.. Infant formulas are being fortified with 12 mg iron per liter .[84]

RESPONSES TO IRON THERAPY IN IRON DEFICIENCY ANEMIA

TIME AFTER IRON ADMINISTRATION	RESPONSE
12 to 24 hr	Replacement of intracellular iron enzymes;subjective improvement;decreased irritability;increased appetite
36 to 48 hr	Initial bone marrow response;erythroid hyperplasia
48 to 72 hr	Reticulocytosis, peaking at 5 to 7 days
4 to 30 days	Increase in hemoglobin level
1 to 3 months	Repletion of stores

TREATMENT OF IRON DEFICIENCY ANEMIA

The regular response of iron deficiency anemia to adequate amounts of iron in iron deficiency anemia is a critical diagnostic and therapeutic characteristic.A daily total dose of 3 to 6 mg/kg of elemental iron in three divided doses is adequate

to treat iron deficiency anemia.[85]The maximum dose is 150 to 200 mg of elemental iron daily.Ferrous sulphate is 20% of elemental iron by weight and is usually given between meals along with juices.Oral iron is fast,effective,economical and safe..Iron is available in various formulations.The amount of elemental iron varies in each of these formulations.For example, ferrous fumarate contains 33% elemental iron by weight, ferrous sulfate contains 20% and ferrous gluconate contains 12% elemental iron.[86] Calcium interferes with the absorption of iron and so both these supplements cannot be taken at the same time of the day.

Parenteral iron is preferred only in cases of children with malabsorption or poor compliance of oral iron or when rapid replacement of iron stores is needed or along with erythropoietin therapy during dialysis.It is available in USA in four forms: iron dextran,iron gluconate,iron sucrose and iron oxide coated with polyglucose sorbitol carboxymethylether.Iron dextran is the FDA approved parenteral formulation of iron in children.[87]

A simple formula to determine the replacement dose of iron dextran:

Dose(ml) = 0.0442 x (desired Hb-observed Hb) x lean body weight +(0.26 x lean body weight)

The CDC recommends that infants under 12 months of age who are not exclusively or primarily breastfed be fed iron-fortified infant formula .Breastfed infants born as preterm or as a low birthweight baby should receive 2-4 mg/kg/day of iron drops (to a maximum of 15 mg/day) between ages 1-12 months. Breastfed infants who do not receive adequate iron (less than 1 mg/kg/day) from supplementary foods by age 6 months should receive 1 mg/kg/day of iron drops.[88]

The American Academy of Pediatrics makes daily supplementation of 1 mg/kg iron drops for exclusively breastfed full-term infants from age 4 months until the infants begin eating iron-containing complementary foods, as a recommendation. Standard infant formulas containing 10 to 12 mg/L iron are supposed to meet the iron needs of infants during the first year of life. The AAP further recommends 2 mg/kg/day iron supplementation for preterm infants aged 1 to 12 months who are fed breast milk.[89]

The WHO strongly recommends universal supplementation with 2 mg/kg/day of iron in children aged 6 to 23 months whose diet is deficient in iron or

who live in regions (such as developing countries) with more than 40% prevalence of anemia.[90]

Iron therapy may increase the virulence of malaria and potentiate certain gram negative infections.Yersinia is an organism that may flourish in iron overloaded states. Dietary counseling is needed in case of iron deficiency anemia.Consumption of cows milk should be limited to <24 ounce/day.[91]

The WHO therefore recommends 6-month supplementation cycles in malaria endemic regions as follows: children aged 24 to 59 months should receive 25 mg iron and those aged 5 to 12 years should receive 45 mg every week for 3 months period, followed by 3 months period of no iron supplementation.. The WHO recommends providing these supplements in malaria-endemic areas in addition to measures to prevent, diagnose, and treat malaria.[92]

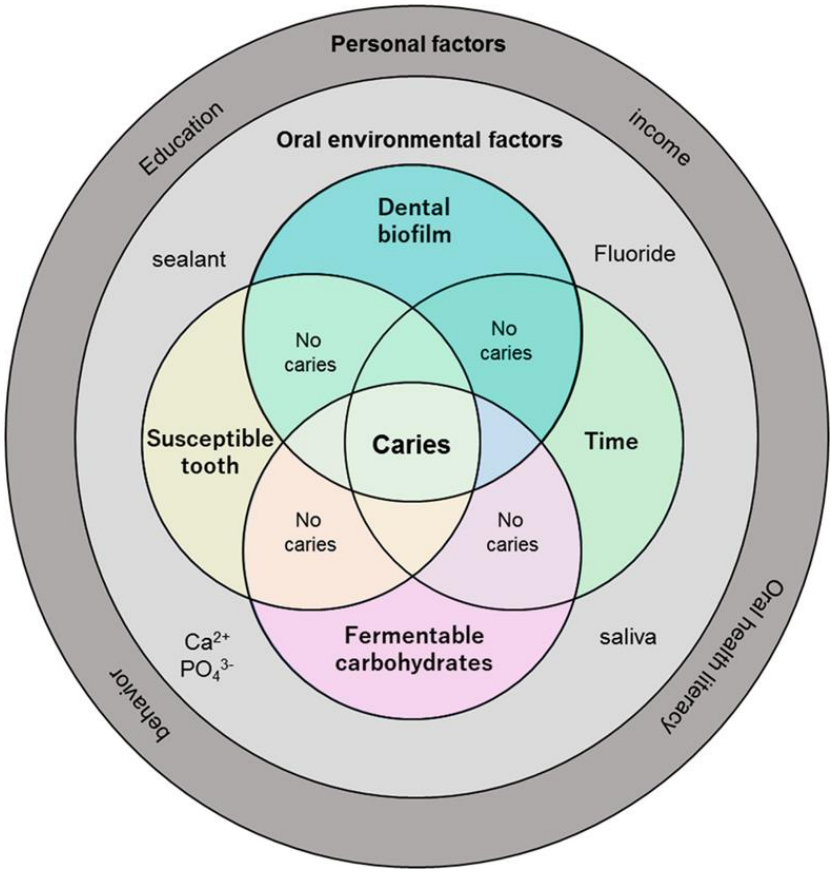
In cases of children with mild iron deficiency anemia, blood count is repeated 4 weeks after starting iron treatment.Hemoglobin increases to 1 g/dl at this time.In children with severe iron deficiency anemia, appearance of reticulocytosis is looked for within 48 to 96 hours of starting treatment.Hemoglobin in such individuals rises by 0.1 to 0.4 g/dl/day.Iron medications are continued for 2 to 3 months after the blood count becomes normal to replenish the iron stores.Blood transfusions are not routinely indicated in iron

deficiency anemia except in cases of imminent heart failure or evidence of substantial blood loss.[93]

DENTAL CARIES

Dental caries is a multifactorial microbial disease caused by cariogenic bacterium, Streptococcus mutans, Lactobacillus and Actinomyces species.When diet rich in sugars (sucrose) is ingested, dietary sugars get fermented resulting in acid production.(lactic acid) .This acid causes demineralization of enamel followed by the dentine. [94]

There are several models to explain dental caries risk factors.One such is multifactorial model shown below:



Caries, a Proteolytic process

Proteolytic enzymes liberated by cariogenic bacteria

↓

destruction of the organic matrix

↓

detachment of inorganic crystals from one another

↓

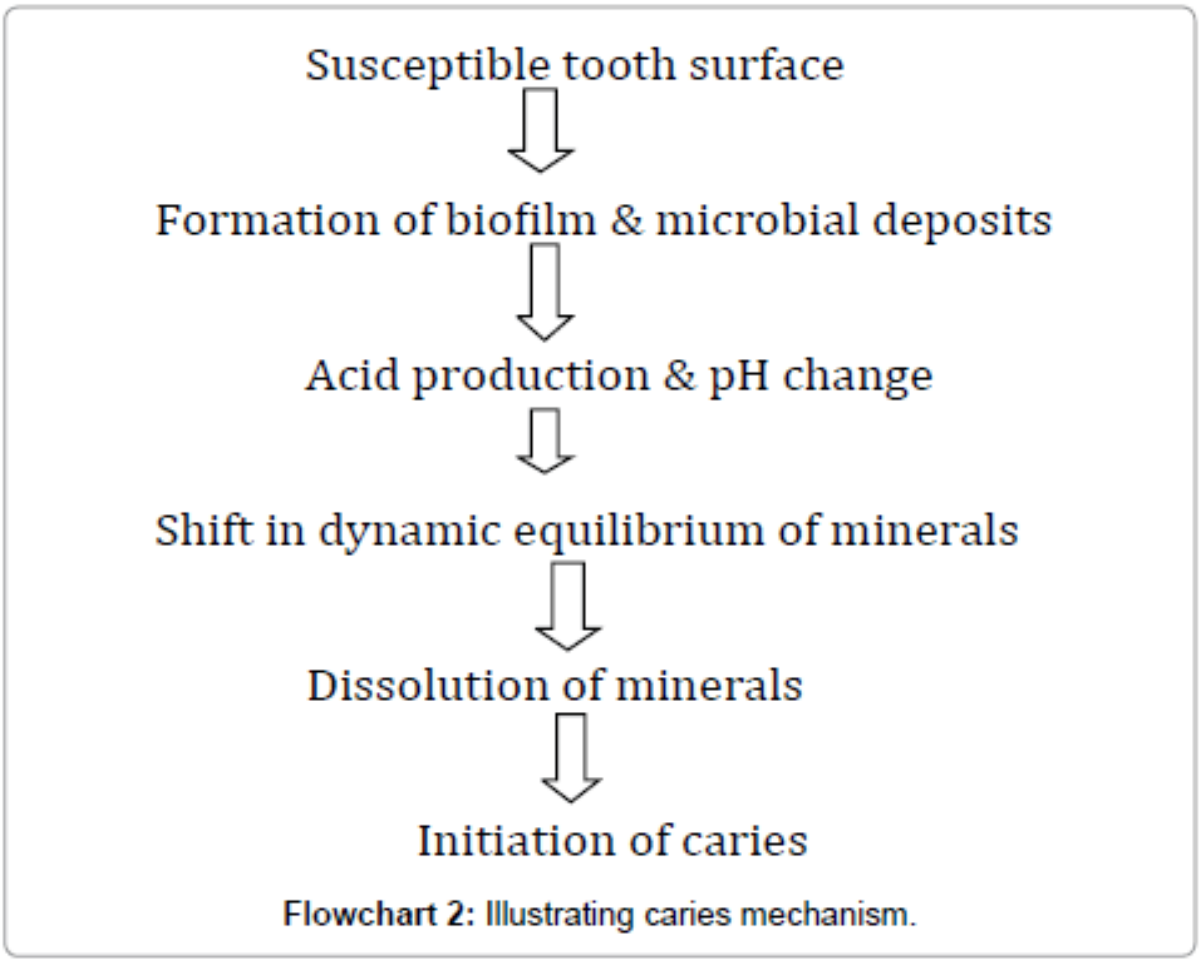
collapse of whole structure

↓

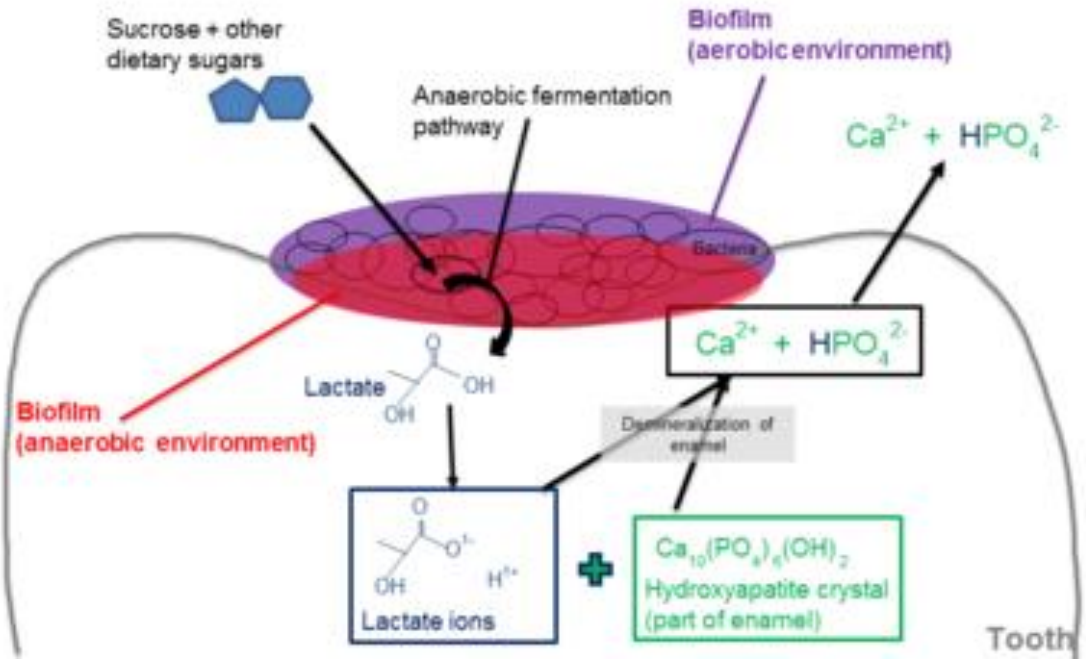
CAVITATION.

Gottlieb (1994) and Gottlieb, Diamond and Applebaum (1946)

_____ Proteolytic theory



Flowchart 2: Illustrating caries mechanism.



The plaque that is not exposed to any fermentable carbohydrate has a resting ph between 6 and 7.[95]This remains stable for an individual . Resting plaque contains high quantities of acetate compared with lactate (acidic) with glutamate and proline being the predominant amino acids. Ammonia, a pH neutralizer, is also seen. [96]

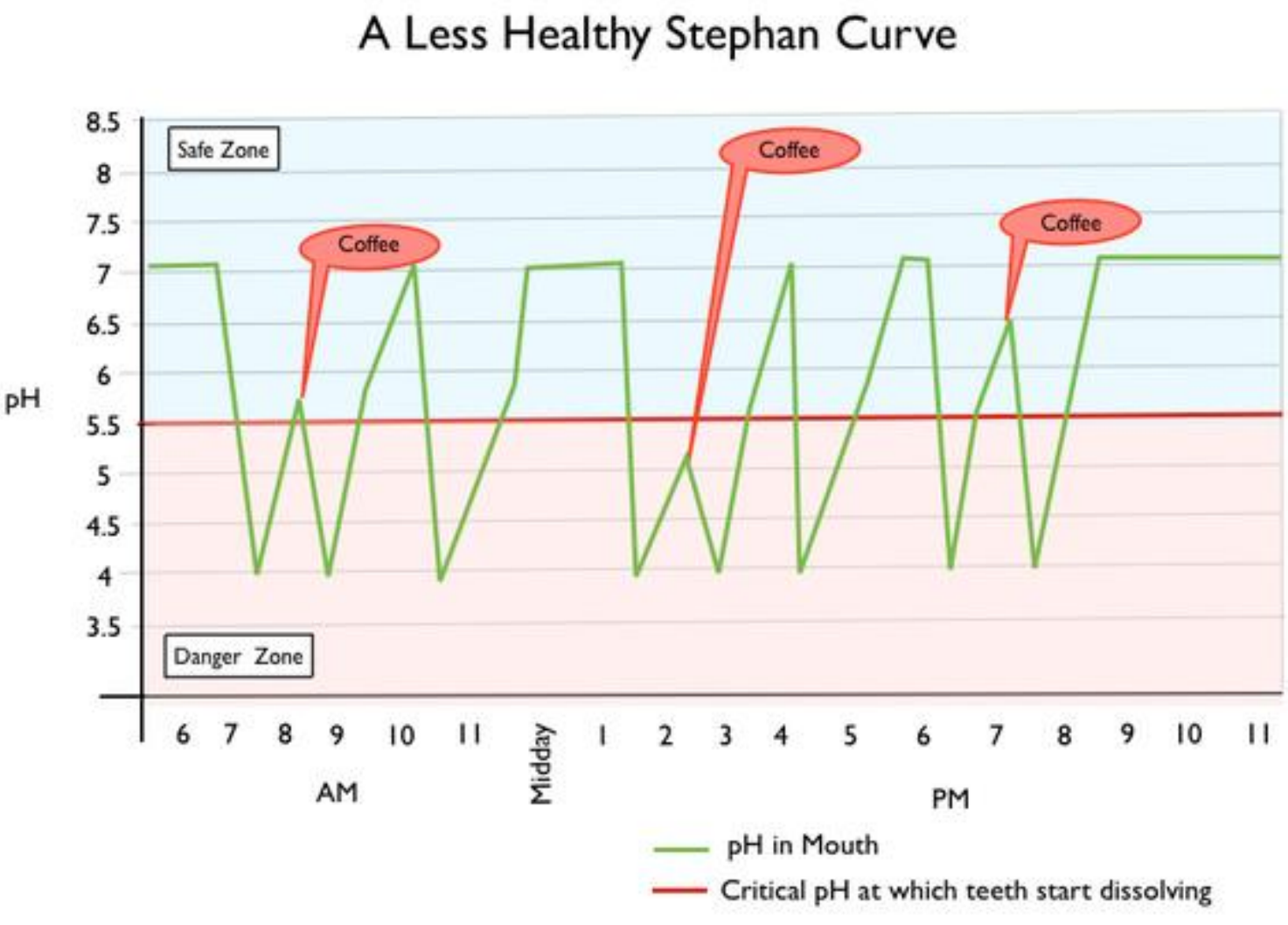
When the resting plaque is exposed to fermentable carbohydrates, pH drops rapidly due to the loss of acetic acid and propionic acid and the production of lactic acid. The rate at which the pH drops depends on several factors, one being the

composition of dental plaque.If more acidogenic bacteria is seen residing in the dental plaque, pH would drop quickly.[97]The other factor is dietary composition where sucrose would be metabolized fast resulting in acid production compared to starch.(larger molecule takes time to diffuse into the plaque).Yet another factor that affects the rate of pH decrease is said to be the buffering capacity of unstimulated saliva.[98] The rate at which plaque pH decreases is also influenced by the density of plaque wherein less dense plaque are easily penetrated by buffering saliva and oxygen causing slower pH decreases than very dense plaque.[99]

The gradual recovery of the plaque pH to normal depends upon the effective buffering capacity of the saliva.. It is also influenced by base production in plaque. For example, ammonia from the deamination of amino acids and breakdown of urea in saliva contribute to the pH rise. The rise in pH is also influenced by the genus *Veillonella* that use lactate as a substrate, metabolizing it to less acidic products like propionic acid. [100]

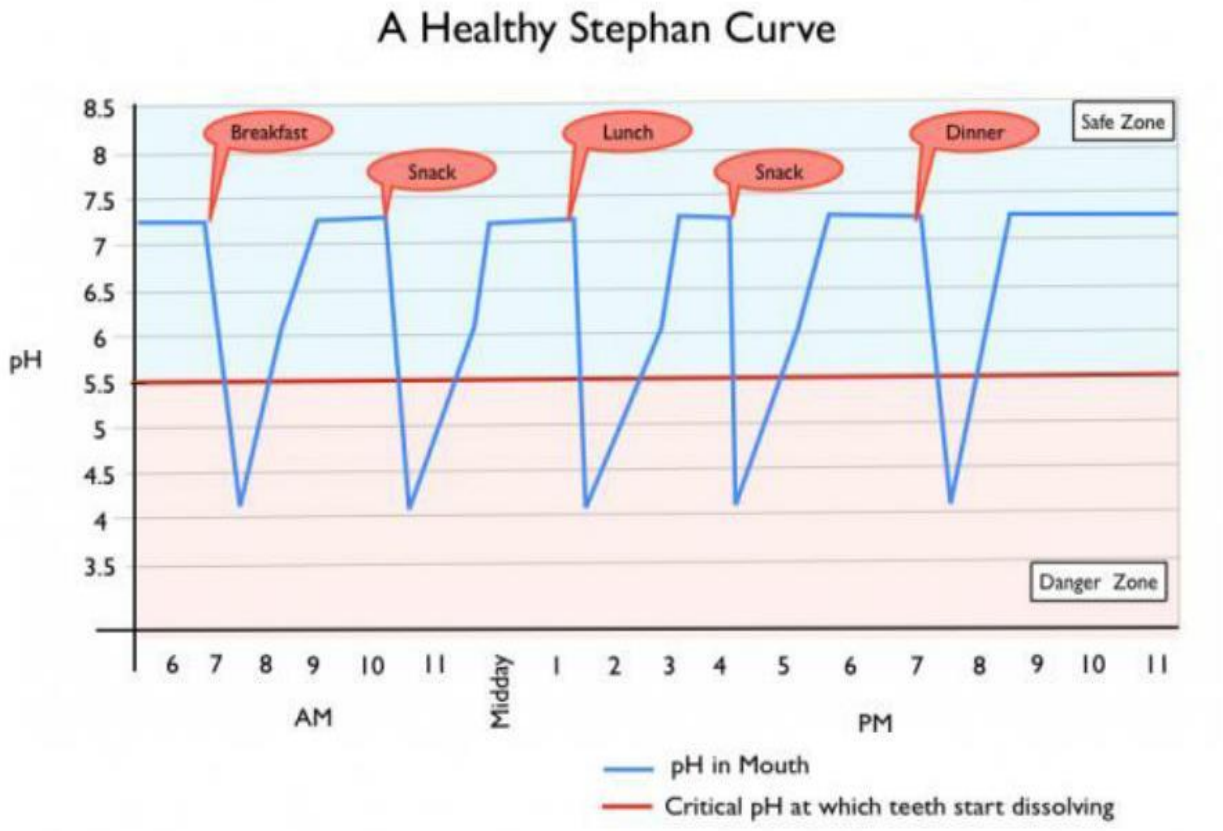
The critical pH is the pH at which saliva and plaque fluid stop getting itself saturated with calcium and phosphate, hence permitting the hydroxyapatite in the enamel of the tooth to dissolve. This is set at 5.5 for enamel. [101]

Tooth is constantly bathed by saliva whose factors help in remineralising the enamel lost during demineralization. When the episodes of demineralization exceeds the remineralisation as explained in Stephans curve, the lesions start progressing to caries.[102]



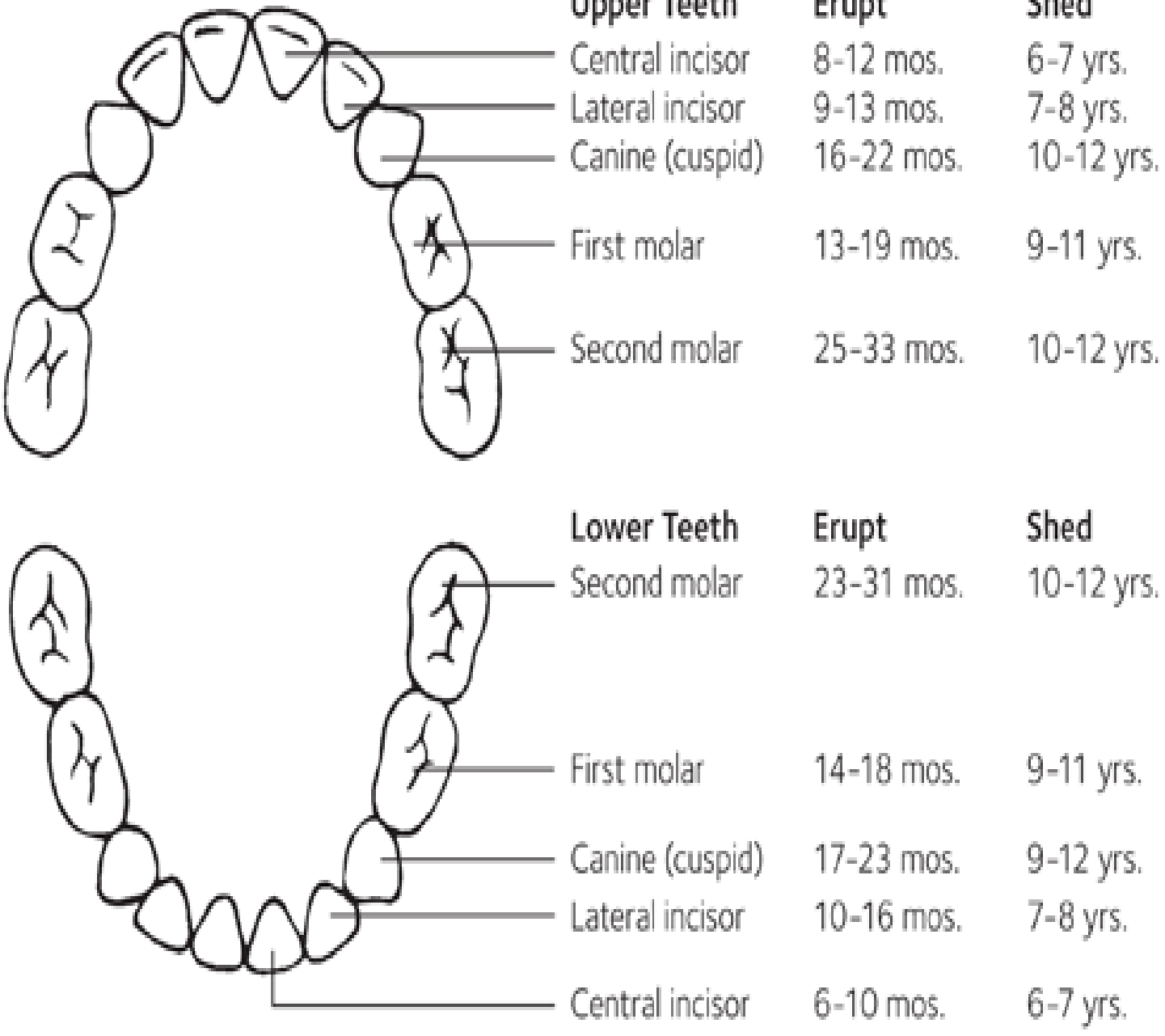
PRINCIPLE OF STEPHEN CURVE

- **Relationship of food intake with PH level with respect to time**



SOURCE: Adapted from: Stephan RM, Miller BF. A quantitative method for evaluating physical and chemical agents which modify production of acids in bacterial plaques on human teeth. J Dent Res. 1943;22;45-51.

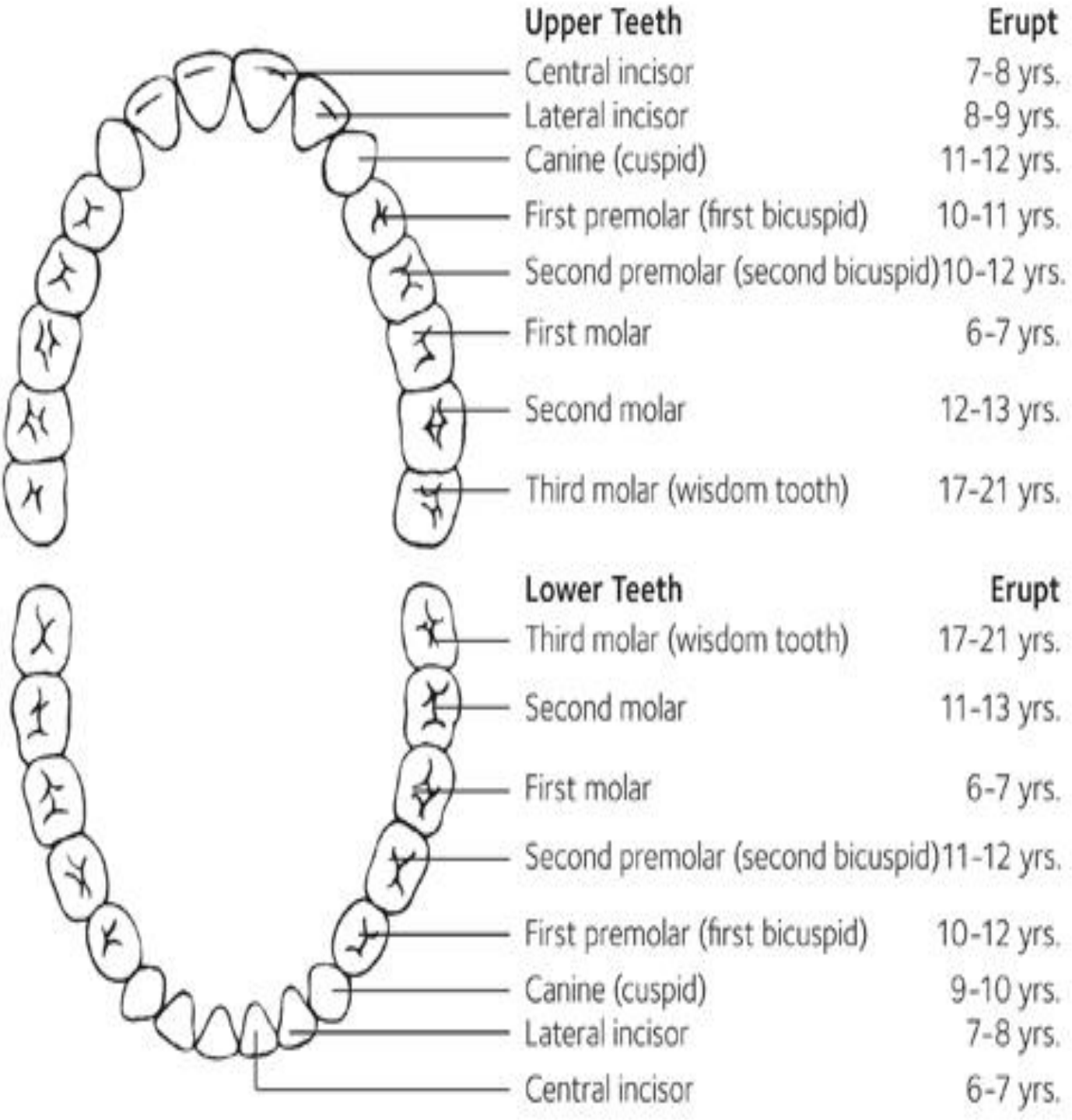
PRIMARY DENTITION



Upper Teeth	Erupt	Shed
Central incisor	8-12 mos.	6-7 yrs.
Lateral incisor	9-13 mos.	7-8 yrs.
Canine (cuspid)	16-22 mos.	10-12 yrs.
First molar	13-19 mos.	9-11 yrs.
Second molar	25-33 mos.	10-12 yrs.

Lower Teeth	Erupt	Shed
Second molar	23-31 mos.	10-12 yrs.
First molar	14-18 mos.	9-11 yrs.
Canine (cuspid)	17-23 mos.	9-12 yrs.
Lateral incisor	10-16 mos.	7-8 yrs.
Central incisor	6-10 mos.	6-7 yrs.

PERMANENT DENTITIO



DIAGNOSIS OF DENTAL CARIES: Dental caries is usually identified by oral examination through visual tactile method where colour change and texture would help to identify it.Radiography is used to detect dental caries where it is seen as a radiolucent triangle.Now,there are latest technologies used to diagnose dental caries at an earlier date. Laser fluorescence technology is being used in DIAGNOdent which measures bacterial end products in carious lesions, helping to detect early demineralization.The intensity of fluorescence will be displayed with a numerical value ranging from 0 to 99..[103] Fibre optic light is used inDigital Imaging Fiber-Optic Translllumination (DIFOTI) to produce an image, which is used for detecting initial areas of demineralization, cracks, or fractures, and aids in providing a quantitative characterization of the caries process.[104]In Quantitative light-induced fluorescence (QLF) the ability of human enamel toshow fluorescence is tested. Demineralizedenamel shows reduced fluorescence due to scattering, as the fluorescence is becauseof the cross-links between structural proteins .[105] The Electronic Caries Monitor (ECM)measures the changes in electrical impedance between healthy enamel and demineralized tooth , based on the principle that normal teethhave lower electrical conductivity compared to demineralized teeth .[106] But it is in the hands of the dentist to evaluate caries risk and formulate specific individualized treatment plan.

RISK FACTORS FOR THE DEVELOPMENT OF DENTAL CARIES

i. Poor socioeconomic status

Children with poor socioeconomic status suffer most from dental caries.

ii. Poor oral hygiene

Children with poor dental hygiene are prone to develop dental caries.

iii. Intake of diet rich in sucrose

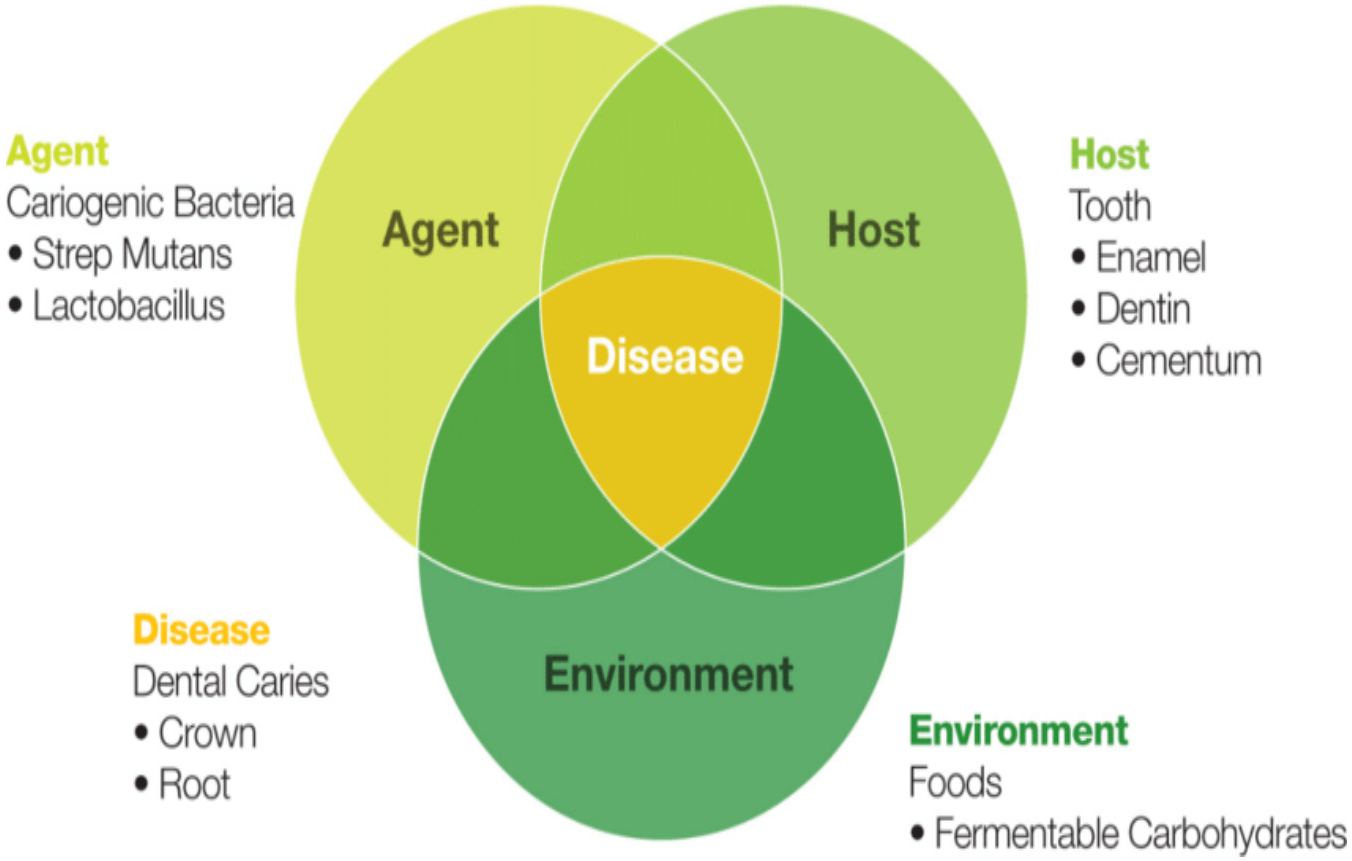
Diet rich in sucrose such as sweets and beverages lead to the fermentation by bacteria such as Streptococcus mutans producing acid resulting in the production of dental caries.

iv. Salivary characteristics

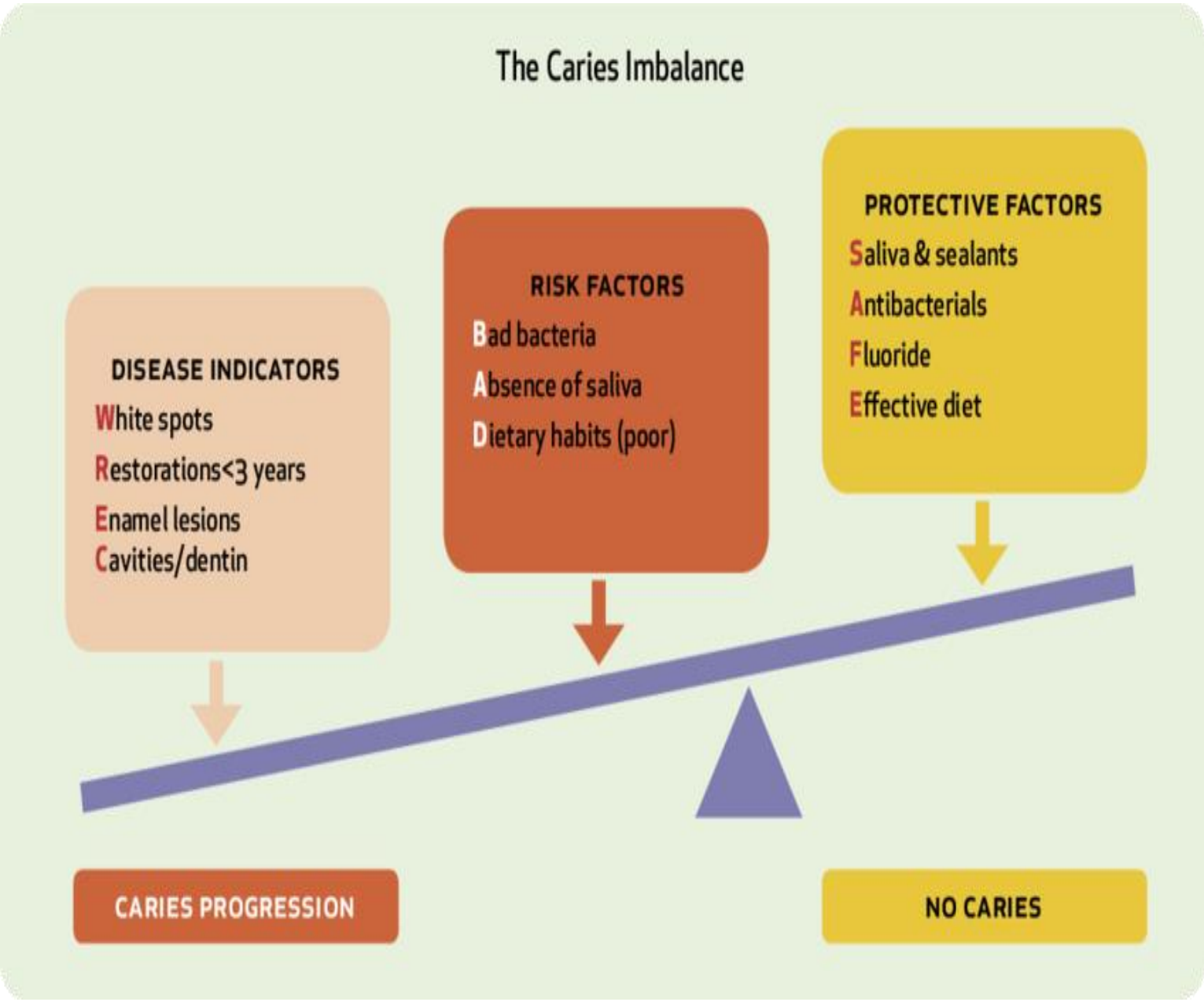
Lack of buffering capacity of saliva contributes to dental caries.**Rashkova et al in 2008** conducted a study in Bulgaria and made an inference that viscous saliva is associated with the production of dental caries. Low salivary flow rate due to medicines,radiation and diseases also promote dental caries.

v) Lack of fluoride supplementation

Fluoride absorbs to the surface of the apatite crystals during an acidic challenge thus inhibiting demineralization. **(Karger et al 2011)**Hence,lack of effective fluoride supplementation contributes to dental caries.



Above is the traditional Keye's triad for etiopathogenesis of dental caries.It was modified by Newbrun who framed a tetrad including time as the fourth factor.



Below is the caries imbalance model proposed by Featherstone in 2006.

ADA American Dental Association®
America's leading advocate for oral health

Caries Risk Assessment Form (Age 0–6)

Patient Name:				
Birth Date:			**Date:**	
Age:			**Initials:**	
		Low Risk	**Moderate Risk**	**High Risk**
Contributing Conditions		**Check or Circle the conditions that apply**		
I.	Fluoride Exposure (through drinking water, supplements, professional applications, toothpaste)	☐ Yes	☐ No	
II.	Sugary Foods or Drinks (including juice, carbonated or non-carbonated soft drinks, energy drinks, medicinal syrups)	Primarily at mealtimes ☐	Frequent or prolonged between meal exposures/day ☐	Bottle or sippy cup with anything other than water at bed time ☐
III.	Eligible for Government Programs (WIC, Head Start, Medicaid or SCHIP)	☐ No		☐ Yes
IV.	Caries Experience of Mother, Caregiver and/or other Siblings	No carious lesions in last 24 months ☐	Carious lesions in last 7–23 months ☐	Carious lesions in last 6 months ☐
V.	Dental Home: established patient of record in a dental office	☐ Yes	☐ No	
General Health Conditions		**Check or Circle the conditions that apply**		
I.	Special Health Care Needs (developmental, physical, medical or mental disabilities that prevent or limit performance of adequate oral health care by themselves or caregivers)	☐ No		☐ Yes
Clinical Conditions		**Check or Circle the conditions that apply**		
I.	Visual or Radiographically Evident Restorations/ Cavitated Carious Lesions	No new carious lesions or restorations in last 24 months ☐		Carious lesions or restorations in last 24 months ☐
II.	Non-cavitated (incipient) Carious Lesions	No new lesions in last 24 months ☐		New lesions in last 24 months ☐
III.	Teeth Missing Due to Caries	☐ No		☐ Yes
IV.	Visible Plaque	☐ No	☐ Yes	
V.	Dental/Orthodontic Appliances Present (fixed or removable)	☐ No	☐ Yes	
VI.	Salivary Flow	Visually adequate ☐		Visually inadequate ☐

Overall assessment of dental caries risk: ☐ Low ☐ Moderate ☐ High

Instructions for Caregiver:

ADA American Dental Association®
America's leading advocate for oral health

Caries Risk Assessment Form (Age >6)

Patient Name:				
Birth Date:			**Date:**	
Age:			**Initials:**	
		Low Risk	**Moderate Risk**	**High Risk**
Contributing Conditions		**Check or Circle the conditions that apply**		
I.	**Fluoride Exposure** (through drinking water, supplements, professional applications, toothpaste)	☐ Yes	☐ No	
II.	**Sugary Foods or Drinks** (including juice, carbonated or non-carbonated soft drinks, energy drinks, medicinal syrups)	**Primarily at mealtimes** ☐		**Frequent or prolonged between meal exposures/day** ☐
III.	**Caries Experience of Mother, Caregiver and/or other Siblings** (for patients ages 6–14)	**No carious lesions in last 24 months** ☐	**Carious lesions in last 7–23 months** ☐	**Carious lesions in last 6 months** ☐
IV.	**Dental Home:** established patient of record, receiving regular dental care in a dental office	☐ Yes	☐ No	
General Health Conditions		**Check or Circle the conditions that apply**		
I.	**Special Health Care Needs** (developmental, physical, medical or mental disabilities that prevent or limit performance of adequate oral health care by themselves or caregivers)	☐ No	**Yes (over age 14)** ☐	**Yes (ages 6–14)** ☐
II.	**Chemo/ Radiation Therapy**	☐ No		☐ Yes
III.	**Eating Disorders**	☐ No	☐ Yes	
IV.	**Medications that Reduce Salivary Flow**	☐ No	☐ Yes	
V.	**Drug/Alcohol Abuse**	☐ No	☐ Yes	
Clinical Conditions		**Check or Circle the conditions that apply**		
I.	**Cavitated or Non-Cavitated** (incipient) **Carious Lesions or Restorations** (visually or radiographically evident)	**No new carious lesions or restorations in last 36 months** ☐	**1 or 2 new carious lesions or restorations in last 36 months** ☐	**3 or more carious lesions or restorations in last 36 months** ☐
II.	**Teeth Missing Due to Caries in past 36 months**	☐ No		☐ Yes
III.	**Visible Plaque**	☐ No	☐ Yes	
IV.	**Unusual Tooth Morphology** that compromises oral hygiene	☐ No	☐ Yes	
V.	**Interproximal Restorations – 1 or more**	☐ No	☐ Yes	
VI.	**Exposed Root Surfaces Present**	☐ No	☐ Yes	
VII.	**Restorations with Overhangs and/or Open Margins; Open Contacts with Food Impaction**	☐ No	☐ Yes	
VIII.	**Dental/Orthodontic Appliances** (fixed or removable)	☐ No	☐ Yes	
IX.	**Severe Dry Mouth** (Xerostomia)	☐ No		☐ Yes

Overall assessment of dental caries risk: ☐ Low ☐ Moderate ☐ High

Patient Instructions:

PREVENTION OF DENTAL CARIES

1) Oral hygiene:

Dental caries progresses only in the presence of a bacterial plaque.Hence effective plaque removal by brushing,flossing should be taught to children to prevent dental caries.

2) Diet

A diet rich in sucrose has to be avoided to prevent dental caries.Frequent consumption of sweets and beverages has to be discouraged.Since it is impractical to limit sugar intake,sugar substititutes like xylitol have been developed. Xylitol is non cariogenic since it prevents the binding of sucrose molecules to streptococcus mutans therby blocking its metabolism.Xylitol also prevents the adhesion of streptococcus mutans. Thus Xylitol helps in preventing dental caries.The ADA recommends xylitol in children age 5 years or older.[107]

3) Fluoride supplementation

Flouride helps prevent demineralization and enhance remineralisation of tooth surfaces.When foods rich in sucrose are being consumed, the acid load produced due to the fermentation of sucrose by cariogenic bacterium causes the demineralization of enamel.The low pH also causes the release of fluoride at the tooth –plaque interface. This released flouoride is avidly taken up by demineralised enamel,along with calcium and phosphate thus enabling a remineralising action.This remineralised enamel is stronger with more fluoride and less carbonate and therefore becomes acid resistant. Fluoride can be added to drinking water,dietary supplements, tooth paste. It is also available as topical gels and varnishes.[108]

The recommended dietary fluoride supplement schedule is shown below:

RECOMMENDED DIETARY FLOURIDE SUPPLEMENT SCHEDULE

	Fluoride concentration in Community drinking water		
AGE	<0.3 ppm	0.3 to 0.6 ppm	>0.6 ppm
0 to 6 months	None	None	None
6 months to 3 years	0.25 mg/day	None	None
3 to 6 years	0.50 mg/day	0.25 mg/day	None
6 to 16 years	1.0 mg/day	0.50 mg/day	None

Sodium fluoride 2.2 mg contains 1 mg of fluoride ion.

Sources:

Meskin LH,ed. Caries diagnosis and risk assessment: a review of preventive strategies and management. J Am Dent Assoc 1995;126(suppl):1S-24S.

American Academy of Pediatric Dentistry. Special issue: reference manual 1994-95. Pediatr Dent 1995;16(special issue):1-96

American Academy of Pediatrics Committee. On Nutrition. Fluoride supplementation for children: interim policy recommendations. Pediatrics 1995;95:777.

4)Pit and fissure sealants:

Pits and fissures are more prone to favour the development of dental caries due to its ability to trap bacterial plaques.Hence,filling pits and fissures with restorative material prevents the accumulation of plaque and hence the formation of dental caries.Its use is therefore advocated mainly for young children with erupting teeth.[109]

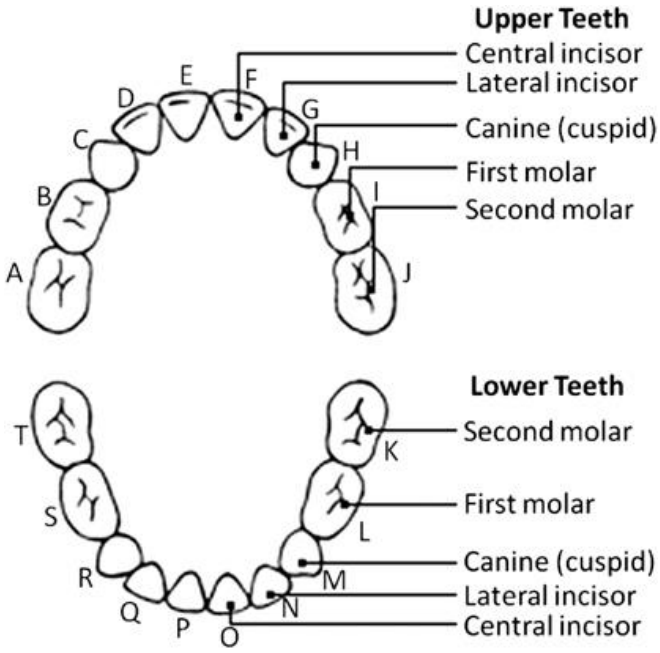
5)Vaccine:

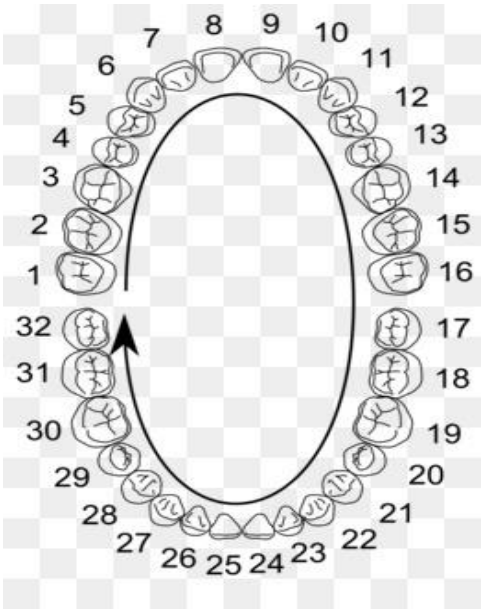
Since dental caries is a multifactorial disease caused by streptococcus mutans,attempts are made to produce vaccine against it.Ways of inactivating glucosyltransferase is also under research. So far, no vaccines for dental caries is out there in the market.[110]

DENTAL FORMULA AND NOTATION SYSYTEMS

The number and type of teeth is expressed by the dental formula.In humans, there are 20 primary teeth and 32 permanent teeth. It is denoted by universal system, palmer notation system and FDI system.

Universal System: This is called the American system.This system uses the Arabic numbers 1 through 32 for the permanent teeth and the letters A through T for the deciduous teeth. The number (1) is assigned to the most posterior upper right permanent tooth (the permanent maxillary right third molar). The highest number is given to the most posterior lower right tooth (the permanent mandibular right third molar). On the same way, the letter A is given to the most posterior lower right deciduous tooth (the upper deciduous right second molar) and the letter T to the most posterior deciduous lower right tooth (the lower deciduous right second molar)[111]





PRIMARY TEETH

PERMANENT TEETH

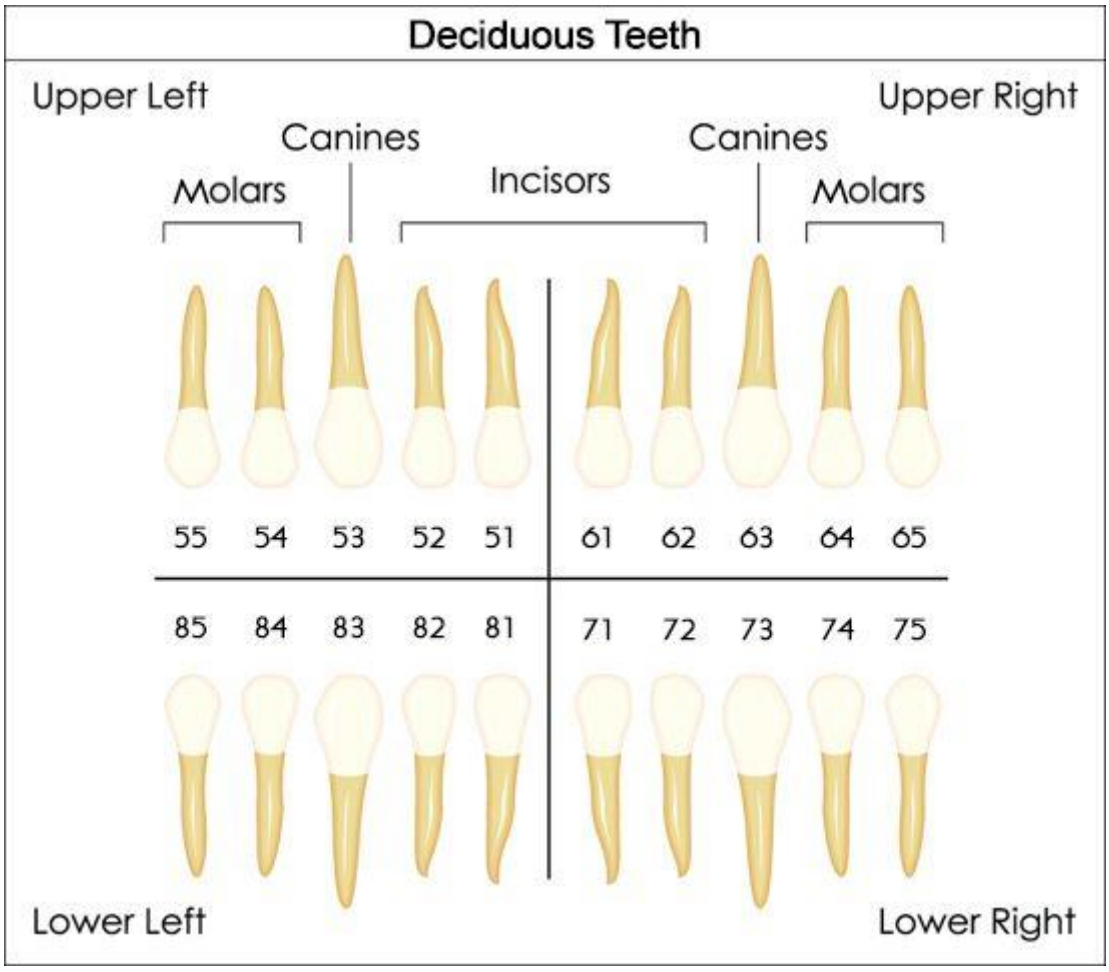
Palmer Notation System:

In this system each of the four quadrants of the mouth is given its own symbol.It is named after Adolf Zsigmondy. A cross is drown, the horizontal line of which separates the maxillary teeth above from the mandibular teeth below. The vertical line represents the midline of the mouth and separates the right from the left side.[112]

Palmer notation

Permanent Teeth															
upper right								upper left							
$8\rfloor$	$7\rfloor$	$6\rfloor$	$5\rfloor$	$4\rfloor$	$3\rfloor$	$2\rfloor$	$1\rfloor$	$\lfloor 1$	$\lfloor 2$	$\lfloor 3$	$\lfloor 4$	$\lfloor 5$	$\lfloor 6$	$\lfloor 7$	$\lfloor 8$
$8\rceil$	$7\rceil$	$6\rceil$	$5\rceil$	$4\rceil$	$3\rceil$	$2\rceil$	$1\rceil$	$\lceil 1$	$\lceil 2$	$\lceil 3$	$\lceil 4$	$\lceil 5$	$\lceil 6$	$\lceil 7$	$\lceil 8$
lower right								lower left							
Deciduous teeth (baby teeth)															
upper right								upper left							
			$E\rfloor$	$D\rfloor$	$C\rfloor$	$B\rfloor$	$A\rfloor$	$\lfloor A$	$\lfloor B$	$\lfloor C$	$\lfloor D$	$\lfloor E$			
			$E\rceil$	$D\rceil$	$C\rceil$	$B\rceil$	$A\rceil$	$\lceil A$	$\lceil B$	$\lceil C$	$\lceil D$	$\lceil E$			
lower right								lower left							

The Federation Dentaire International (FDI): It is a simple bi-digital system in which each tooth is referred to by two digits the first digit represent the quadrant of the mouth and the second digit represent the tooth. The maxillary right quadrant is given number "1", maxillary left quadrant "2", mandibular left quadrant "3", and mandibular right quadrant "4". For deciduous dentition the maxillary right quadrant is given number "5", maxillary left quadrant "6", mandibular left quadrant "7" and mandibular right quadrant "8". The type of each tooth is represented also by numbers from 1-to-5, where 1 is the central incisor and 5 is the second molar.[113] Accordingly the deciduous (upper) and permanent (lower)

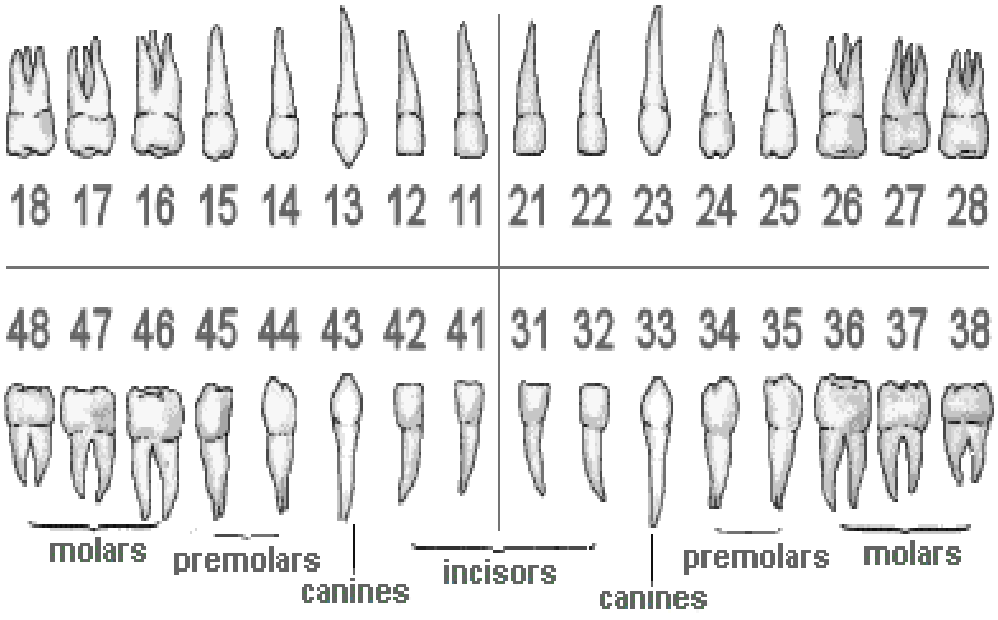


dentitions is represented here.

FDI two-digit tooth numbering system

Teeth numbering chart for adult teeth

upper right	upper left



lower right	lower left

DMFT index

The **D**ecayed, **M**issing, **F**illed (**DMF**) **T**ooth index is used as the key index of caries measurement in dental epidemiology.[114] The DMF Index is applied to the permanent dentition and is expressed as the total number of teeth or surfaces that are decayed (D), missing (M), or filled (F) in an individual. The minimum score is 0 and the

maximum score is 32.(28 if third molars are not included).When the index is applied only to tooth surfaces (five per posterior tooth and four per anterior tooth), it is called the DMFS index. Minimum score is 0 and maximum score is 148(128 if third molars are not included).[115]

When written in lowercase letters, the dmf index is applied to the primary dentition.. The dmft index expresses the number of affected teeth in the primary dentition, with minimum score of 0 and maximum of 20.. The dmfs index expresses the number of affected surfaces in primary dentition (five per posterior tooth and four per anterior tooth),with minimum score of 0 and a maximum of

88..[115] Due to the difficulty in distinguishing between teeth extracted due to caries and those that have naturally exfoliated, missing teeth may be ignored in some protocols. In such situation, it is called the df index.

Calculating DMFT:

The following teeth are not counted when calculating DMFT score:

- unerupted teeth
- congenitally missing teeth or supernumerary teeth,
- teeth removed for reasons other than dental caries
- primary teeth retained in the permanent dentition.

Counting the third molars is optional. A tooth with caries is recorded as D. A tooth with both carious lesion and a restoration is also recorded as D. When a tooth is extracted due to caries, it is recorded as M. A tooth with permanent or temporary filling or when a filling is defective but not decayed is recorded as an F. Teeth restored for reasons other than caries are not considered as F. [115]

Calculating DMFS:

Totally, there are five surfaces on the posterior teeth: facial, lingual, mesial, distal, and occlusal and four surfaces on anterior teeth: facial, lingual, mesial, and distal.The calculation is same as DMFT score.The tooth surface with carious lesion is counted as D.The tooth surface with both a carious lesion and a restoration is also counted as D.. The tooth extracted due to caries is counted as an M. The tooth surface with permanent filling , or a defective filling but not decayed one, is counted as an F. Surfaces restored on account of reasons apart from caries are not considered as an F. The total count is 128 or 148 surfaces. (excluding or including the third molars)[115]

Limitations of DMF Index:

Though DMF indices throw light on to the perspectives of dental caries, they do have certain limitations.There is inter observer bias according to some researchers' point of view.This index does not provide significant information on the teeth that is at risk for

caries .This index does not give due importance to the lost through process other than caries,such as periodontal disease.[115]

ORAL HEALTH SURVEILLANCE

The World Health Organisation aims to reduce the global average of DMFT not more than 4 by the age of 12 years by the year 2000. The World Health Day in 1994 was entirely dedicated to oral health which reflects the paramount importance given to oral health.This led to the development of oral disease surveillance systems by WHO several years ago, especially in relation to dental caries in children.

According to the WHO Oral Health Data Bank in 1980, DMFT values were available for 107 of 173 countries, where 51% had 3 DMFT or less.. In the year 2000, data were available for 184 countries, wherein, 68% had less than 3 DMFT.

In 1981, WHO and the FDI World Dental Federation together formulated goals for oral health to be achieved by the year 2000 as follows:

1. Half of the population belonging to 5-6 year-olds to be free of dental caries.

2. The global average not to be more than 3 DMFT at 12 years of age.

3. 85% of the population must retain all their teeth at the age of 18 years.

4. A 50% reduction in edentulousness among the 35-44-year-old population compared with those of the 1982 level.

5. A 25% reduction in edentulo[usness at the age of 65 years and over, compared with the 1982 range.

6. A database system for monitoring changes in oral health to be established.[116]

IRON DEFICIENCY ANEMIA ND DENTAL CARIES

Human saliva plays an enormous role in preventing dental caries. Saliva contains bicarbonates, phosphates, and urea which modulates pH and contributes to the buffering capacity of saliva.The proteins and mucin in saliva helps in the

aggregation of bacteria contributing to dental plaque metabolism.It also has calcium, phosphate, and proteins which serve in the process of mineralization.Saliva is also a storehouse of immunoglobulins, proteins, and enzymes which provide antibacterial action.[117]

. Iron is a predominant transition metal present in saliva. Salivary iron levels correlate with body iron levels.[118].It serves as a cofactor for Lactoferrin, which is known for its antibacterial property.Streptococci are those bacteria that flourish under iron deficiency states predisposing to dental caries.[119]Yet another plausible explanation is that chronic pulpitis triggers inflammatory cascade with cytokines release suppressing erythropoiesis and thus causing anaemia.[120]

It has been known that iron deficiency anaemia impairs salivary gland function thereby reducing the flow rate and buffering capacity of saliva predisposing to caries. The major buffering agent in resting saliva is inorganic phosphate and in stimulated saliva is carbonic acid / bicarbonate system.[121]

Lactoferrin in saliva has antibacterial action.Iron serves as a cofactor for lactoferrin thereby preventing the adhesion of Streptococcus mutans biofilm formation. Iron dental caries.[122]

The enzyme Glucosyl transferase produced by streptococcus mutans is a key factor in the development of dental caries.This enzyme uses sucrose as a substrate in the production of either insoluble or water soluble glucans.Divalent metal ions like iron and copper inhibits this enzyme preventing biofilm formation. In iron deficiency anaemia,glucosyl transferase enzyme is not inhibited resulting in production of glucans predisposing to dental caries.[123]Iron is said to have a restorative function on the enamel.It helps to remineralise the enamel that is lost through demineralization by forming a layer on the enamel and helping in the deposition of salivary calcium and phosphate. [124]

STUDY DESIGN

Hospital based case control study done at Institute of child health and research centre, Madurai medical college, Madurai, for a period of 12 months (august2018- September 2019).This study was approved by the Ethics Committee at Madurai Medical College.

INCLUSION CRITERIA

All children aged 1 to 12 years admitted or treated as outpatients in Government Rajaji Hospital,Institute of Child Health and Research Centre, Madurai Medical College,Madurai.

EXCLUSION CRITERIA

- Congenital diseases affecting dentition
- Children with mental retardation
- Children taking medicines which interfere with serum iron level

Diseases affecting dentition are as follows:

1. Gardner syndrome :supernumerary teeth
2. Hypomelanosis of Ito :hypodontia
3. porphyria :erythodontia
4. Papillon-Lefevre syndrome : loss of deciduous and permanent teeth by late childhood
5. Haim-Munk syndrome : loss of deciduous and permanent teeth by late childhood
6. Incontinentia pigmenti :pegged teeth

7. Tuberous sclerosis :pitted teeth
8. Hyper IgE syndrome :retention of primary teeth

MATERIALS AND METHODS:

Children who are outpatients and inpatients in Government Rajaji Hospital,Madurai were subjected to dental examination.Hundred children with dental caries were selected from the age group of 1 to 12 years. The children were examined at Government Rajaji Hospital by a single examiner.. They were examined in the dental clinic under a medical light, using a sharp probe, cotton rolls, and a dental mirror. Data

on caries prevalence in relation to the haemoglobin level were collected through previously validated studies. Dental caries assessment was done according to the WHO using DMFT score.Participants were questioned regarding breast feeding and bottle

feeding patterns,snacking habits, brushing habits,rinsing mouth after feeds, use of fluoridated tooth paste and regular visits with dentist. of the relation .Age and sex

matched controls without dental caries were also selected.2 ml of blood is collected from each of them and subjected to HB,MCV,HCT and serum iron.The mean Hemoglobin, Mean Corpuscular Volume and Haematocrit were compared between two groups. The observed data are tabulated and statistical analysis is done.

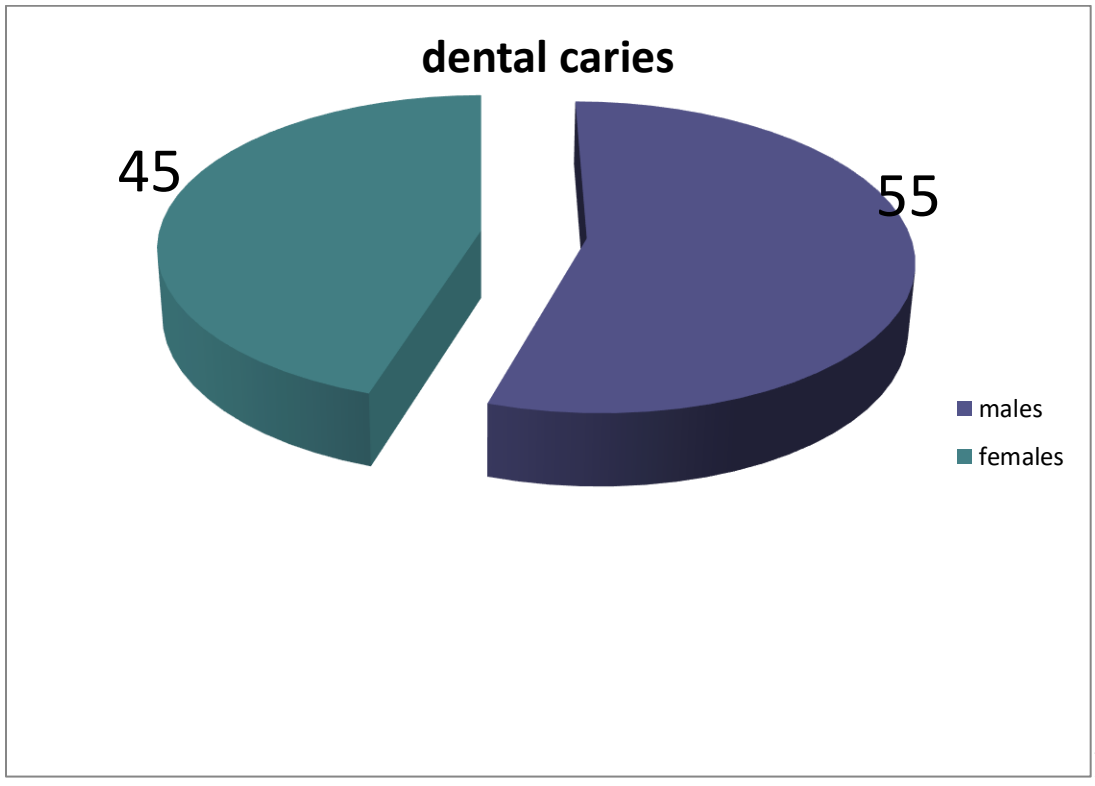
The serum iron is analysed using colorimetric method using ferrozine as reagent.

STATISTICAL ANALYSIS

The statistical test that was used for this analysis is a simple descriptive measurement. Hence, the variables were measured by means and standard deviations. In addition, the test was also assessed via counts and percentages. Chi-Square test was used to test relation between categorical variables. The Independent t-test was used to compare the study variables of two grouped means. The Pearson Correlation was used to check the dependency of two continuous variables to find a positive or negative correlation between serum iron and DMFT score.A p value less than o.o5 was the criterion for rejecting null hypothesis.The data were entered into Microsoft Office Excel 2007 spreadsheet and IBM SPSS (Statistical Package for Social Sciences) version 22 was used to perform all statistical calculations.

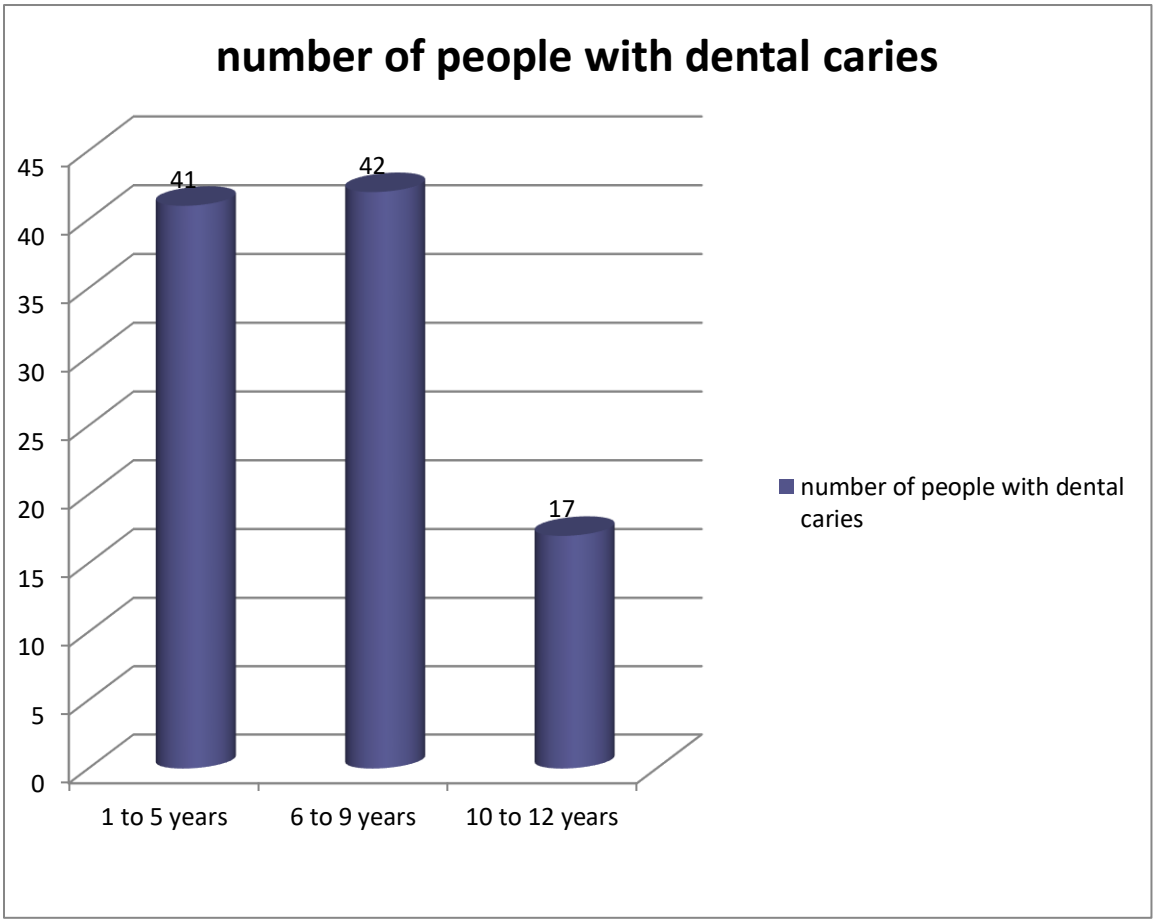
The sex distribution of people with dental caries in our study is shown below

Number of males with dental caries	55
Number of females with dental caries	45
Total	100



From the above pie chart, it is implied that among the children with dental caries, 55% are males and 45% are females.

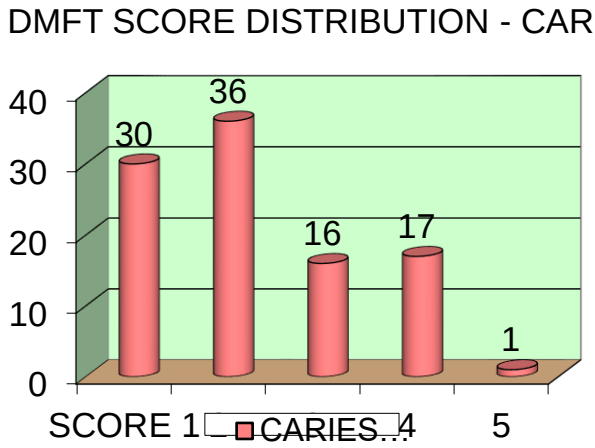
AGE GROUP	NUMBER OF PEOPLE WITH DENTAL CARIES
1 TO 5 YEARS	41
6 TO 9 YEARS	42
10 TO 12 YEARS	17



Among the 100 people with dental caries, 41 belong to the 1 to 5 years category.42 belong to the 6 to 9 years category and 17 belong to the 10 to 12 years category.

The DMFT score for 100 individuals in the group with dental caries is shown below:

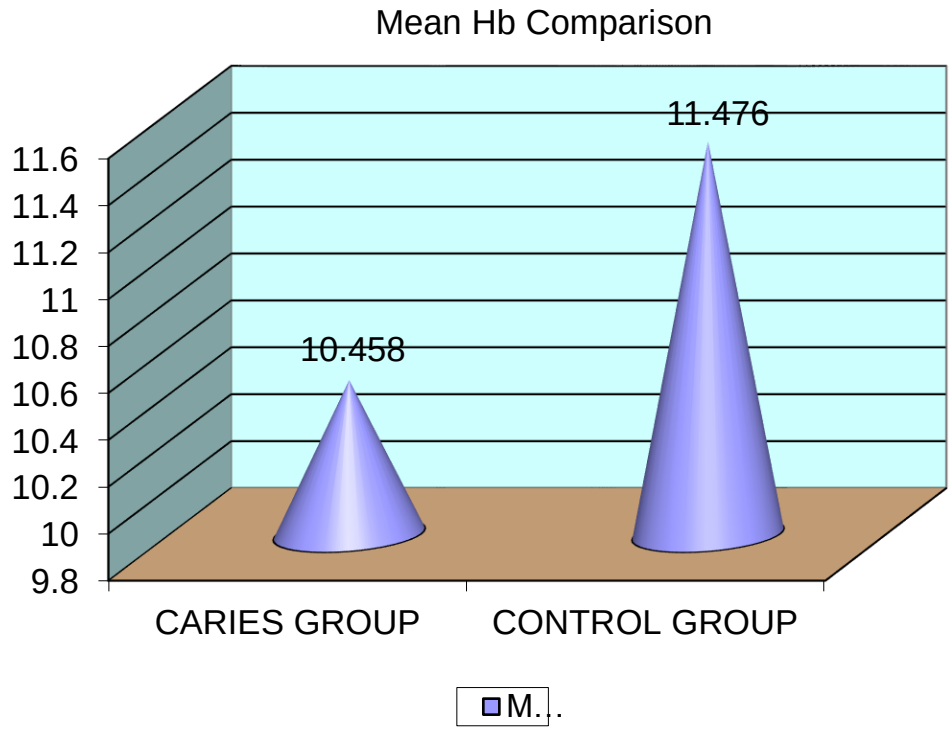
DMFT Score	CARIES GROUP
SCORE 1	30
2	36
3	16
4	17
5	1
TOTAL	100



In our study , out of 100 people, 30 had DMFT score of 1,36 had DMFT score of 2,16 had DMFT score of 3,17 had DMFT score of 4 and 1 had DMFT score of 5.Therfore, 36% of people in study had DMFT score of around 2.

The hemoglobin values in both caries and control group is as follows:

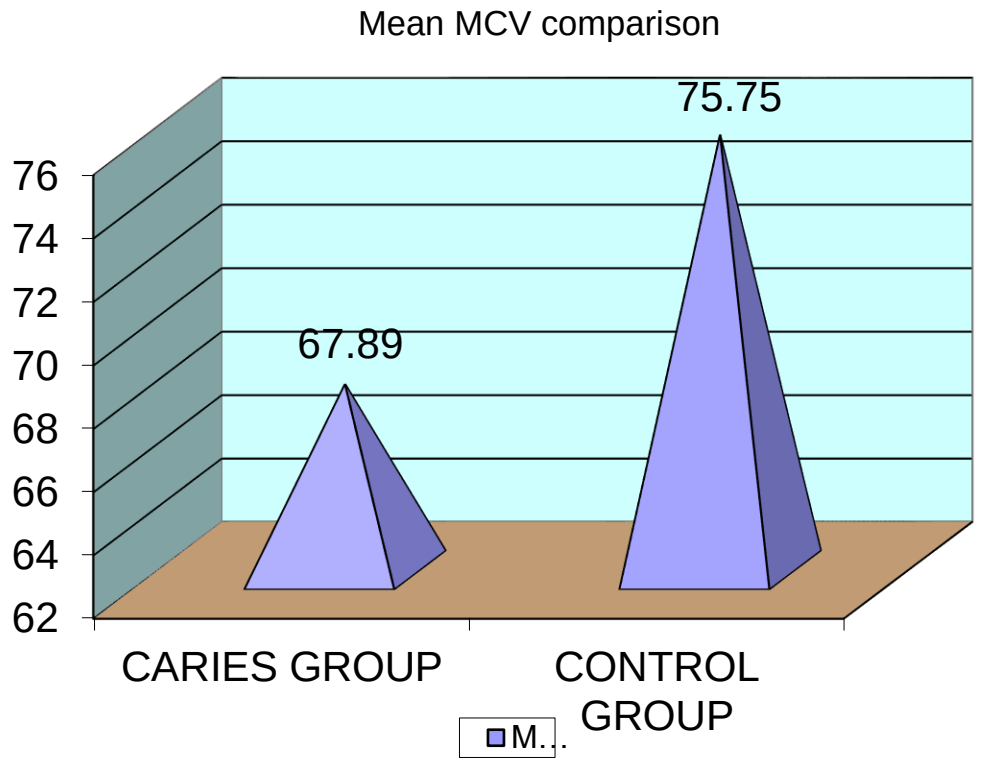
Hb	CARIES GROUP	CONTROL GROUP
Mean	10.458	11.476



It is evident from the above chart that the mean Hemoglobin is significantly less in the dental caries group compared to the control group.($p<0.001$)

The mean corpuscular volume in both caries and control group is as follows:

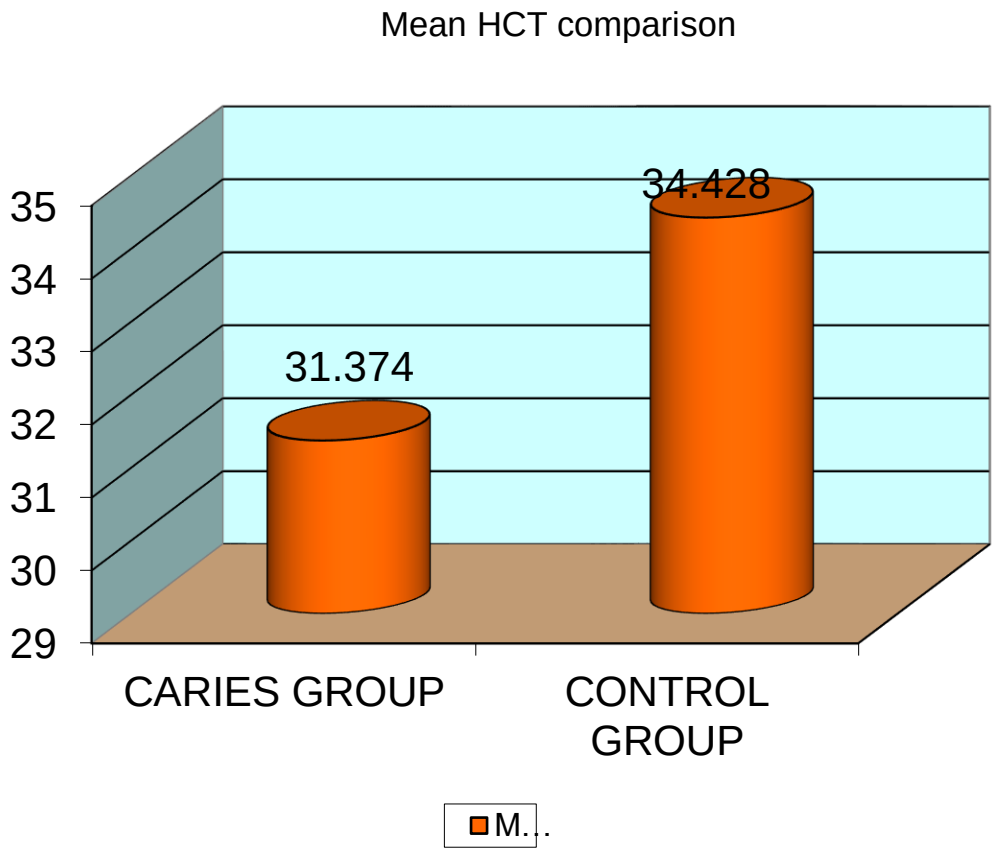
MCV	CARIES GROUP	CONTROL GROUP
Mean	67.89	75.75



In our study, it is shown that mean MCV is significantly less in caries group compared to the control group($p<0.001$)

The mean haematocrit between the caries group and the control group is as follows:

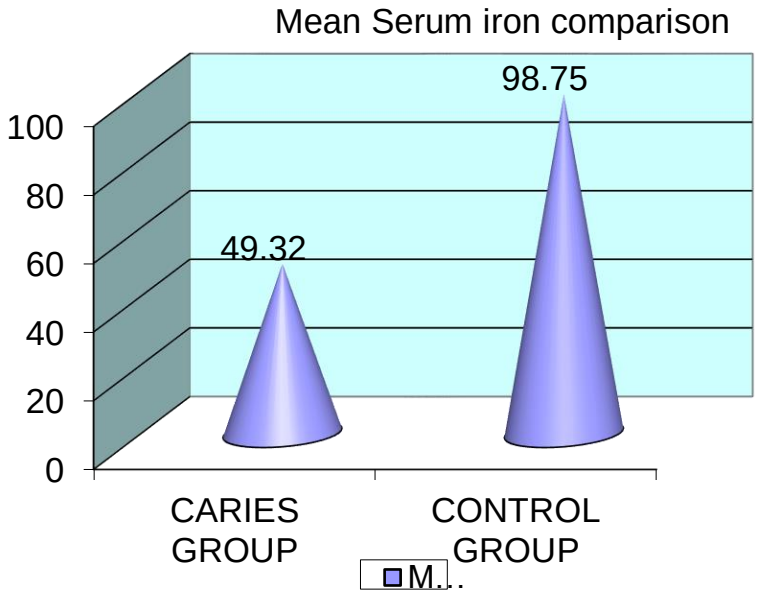
HCT	CARIES GROUP	CONTROL GROUP
Mean	31.374	34.428



In our study, mean haematocrit in dental caries group is significantly less (31.374) compared to control group (34.428)($p<0.001$)

The mean serum iron between the dental caries group and the control group is as follows:

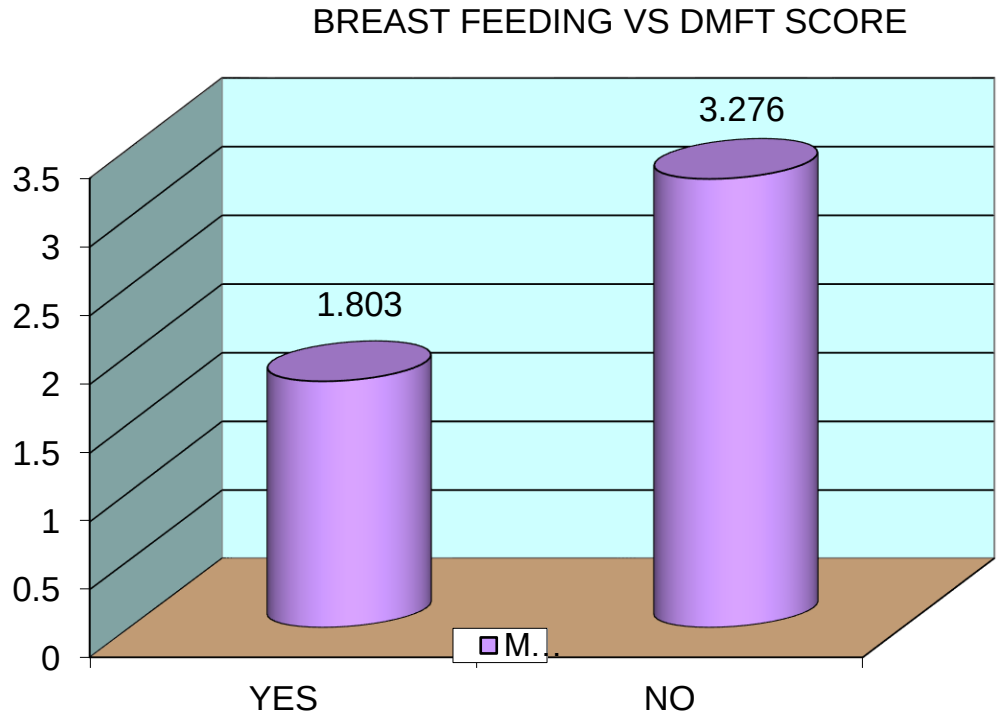
serum iron (microgram/dl)	CARIES GROUP	CONTROL GROUP
Mean	49.32	98.75



In our study, there is significant difference in serum iron between caries and control group.The mean serum iron is drastically less (49.32) in caries group comparedto control (98.75) ($p<0.001$)

The mean DMFT score in both babies being breast fed and not breast fed is being compared as shown below:

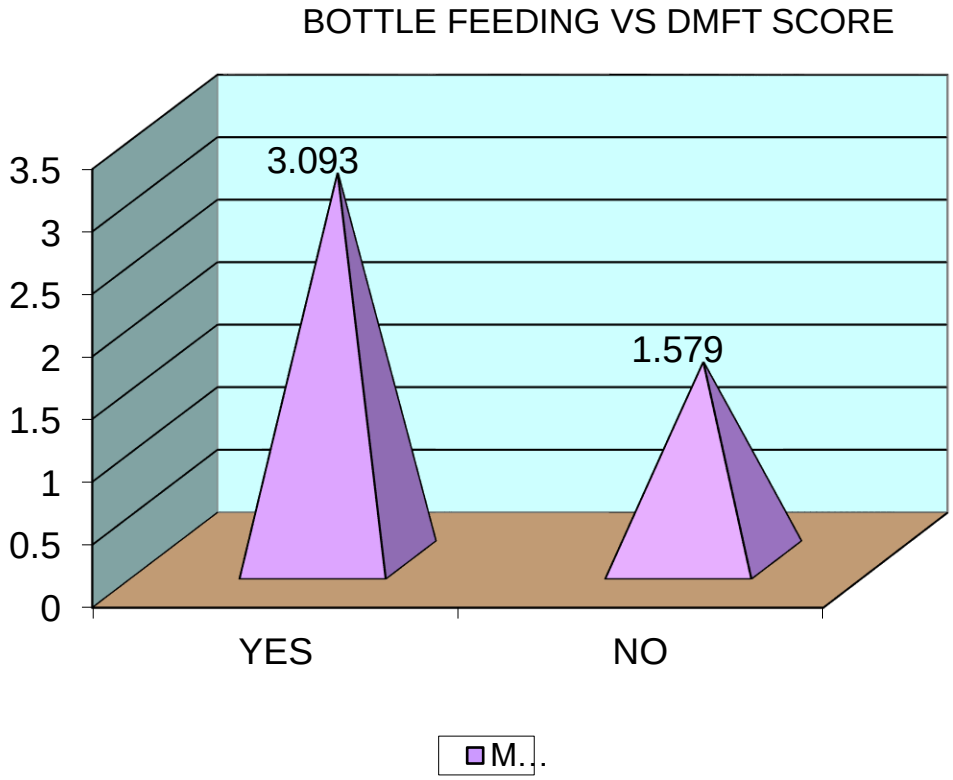
Breast Feeding VS DMFT Score		
Breast Feeding	Mean	SD
YES	1.803	0.839
NO	3.276	0.922
P' value	<0.001 Significant	



In our study, it is shown that mean DMFT score is less in breast feeding group and high in non breast feeding group.Breast feeding protects against dental caries.

The mean DMFT score is compared between babies fed through bottle and those not bottle fed.

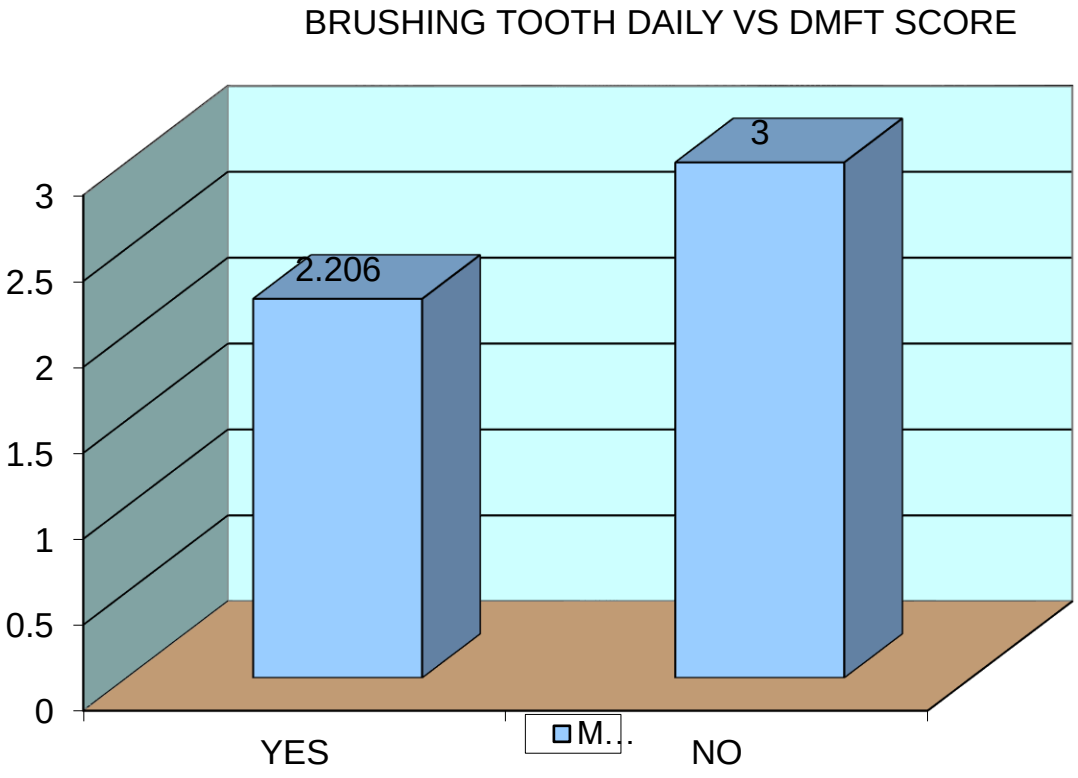
bottle feeding VS DMFT Score		
Bottle feeding	Mean	SD
YES	3.093	0.996
NO	1.579	0.596
P' value	<0.001 Significant	



In our study, it is evident that the mean DMFT score is higher in bottle feeding group.

The DMFT score is compared between those who brush daily and those who do not brush daily.

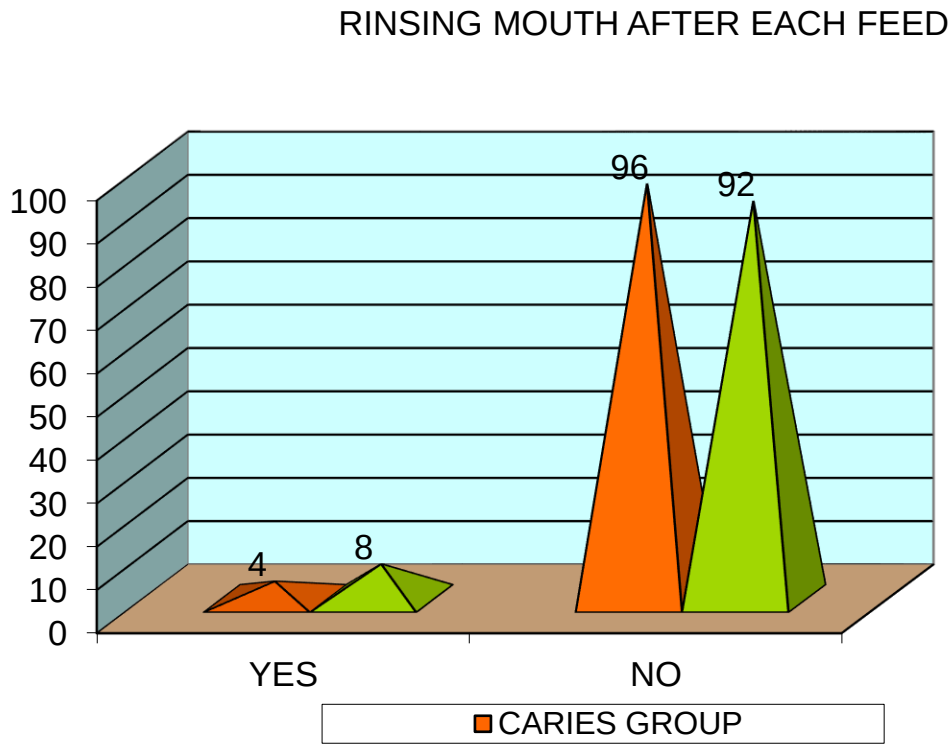
brushing tooth daily VS DMFT Score		
brushing tooth daily	Mean	SD
YES	2.206	1.089
NO	3	1
P' value	0.216 Not sig	



According to our study, there is no significant difference in the Mean DMFT score between those who brush daily and those who do not.

The number of people who rinse mouth after each feed in both caries and control group is shown below:

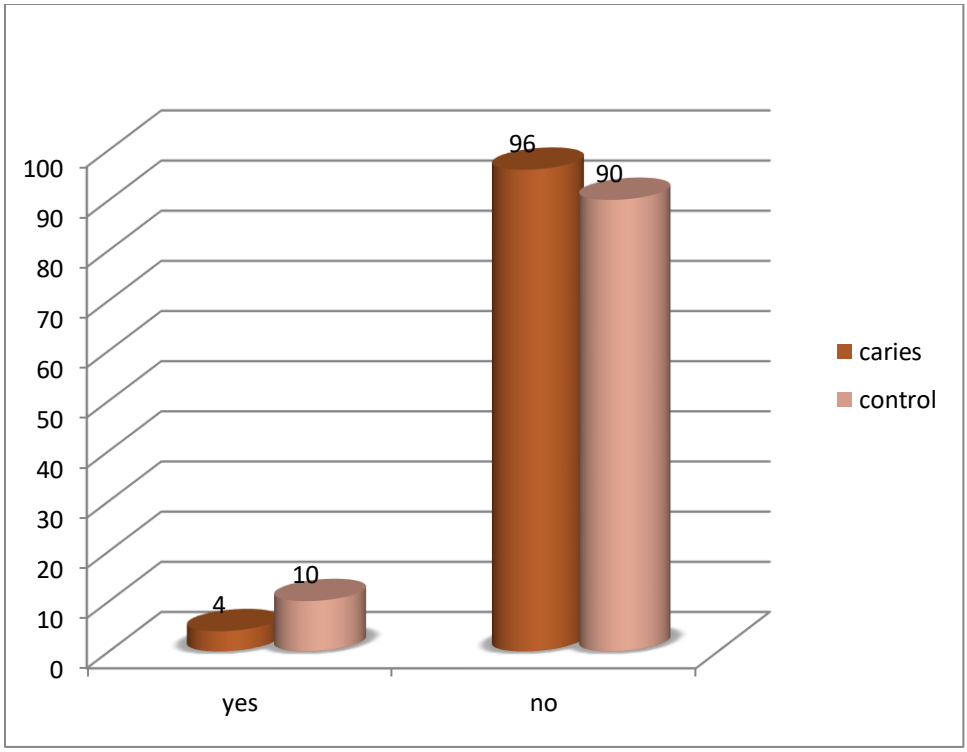
rinsing mouth after each feed	CARIES GROUP	CONTROL GROUP
YES	4	8
NO	96	92
TOTAL	100	100



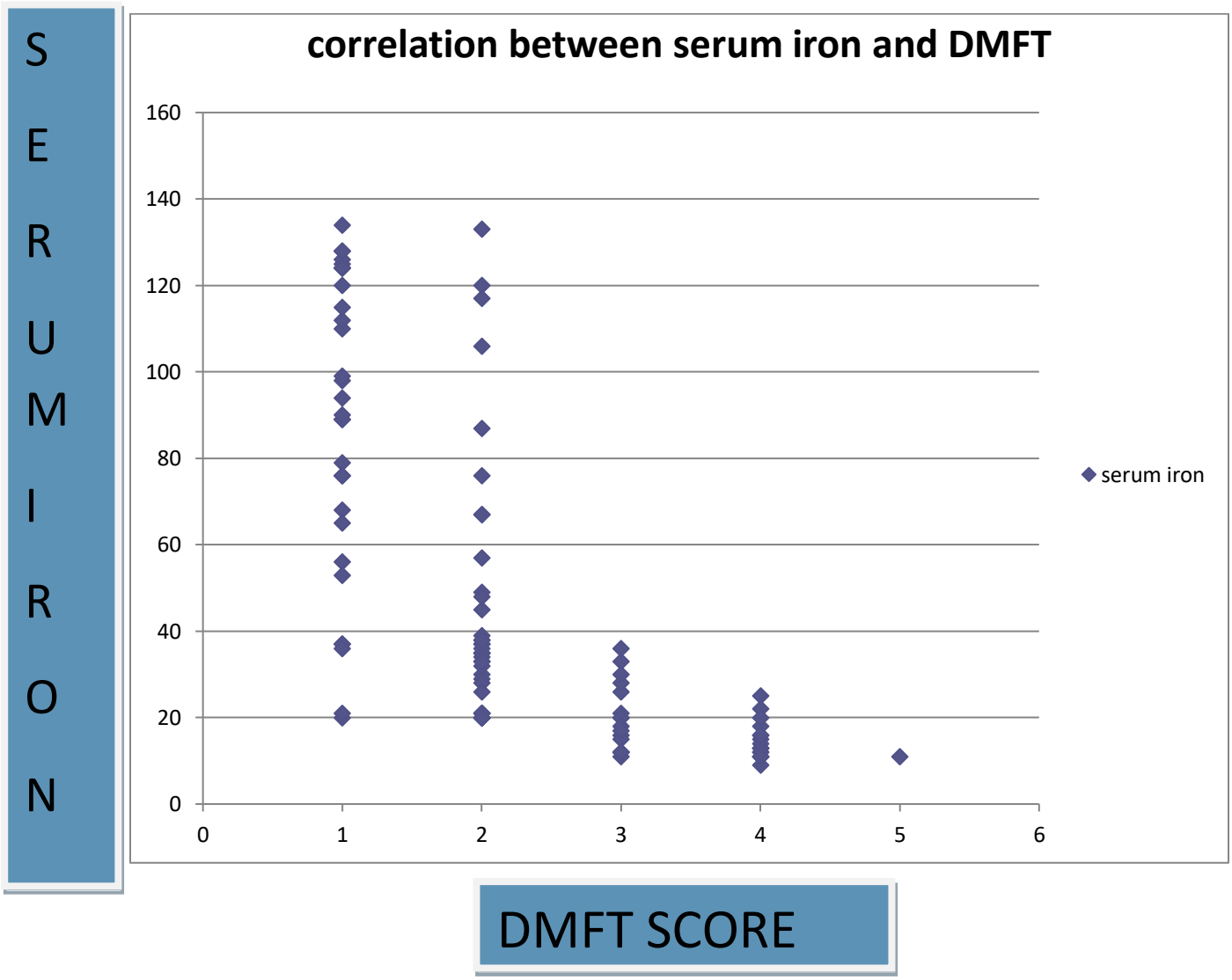
In our study, 4 % of people in caries group has the good practice of rinsing mouth after each feed compared to 8% in the control group.

The number of people who brush tooth twice daily in both caries and control group is shown below:

brushing tooth twice daily	CARIES GROUP	CONTROL GROUP
YES	4	10
NO	96	90
TOTAL	100	100

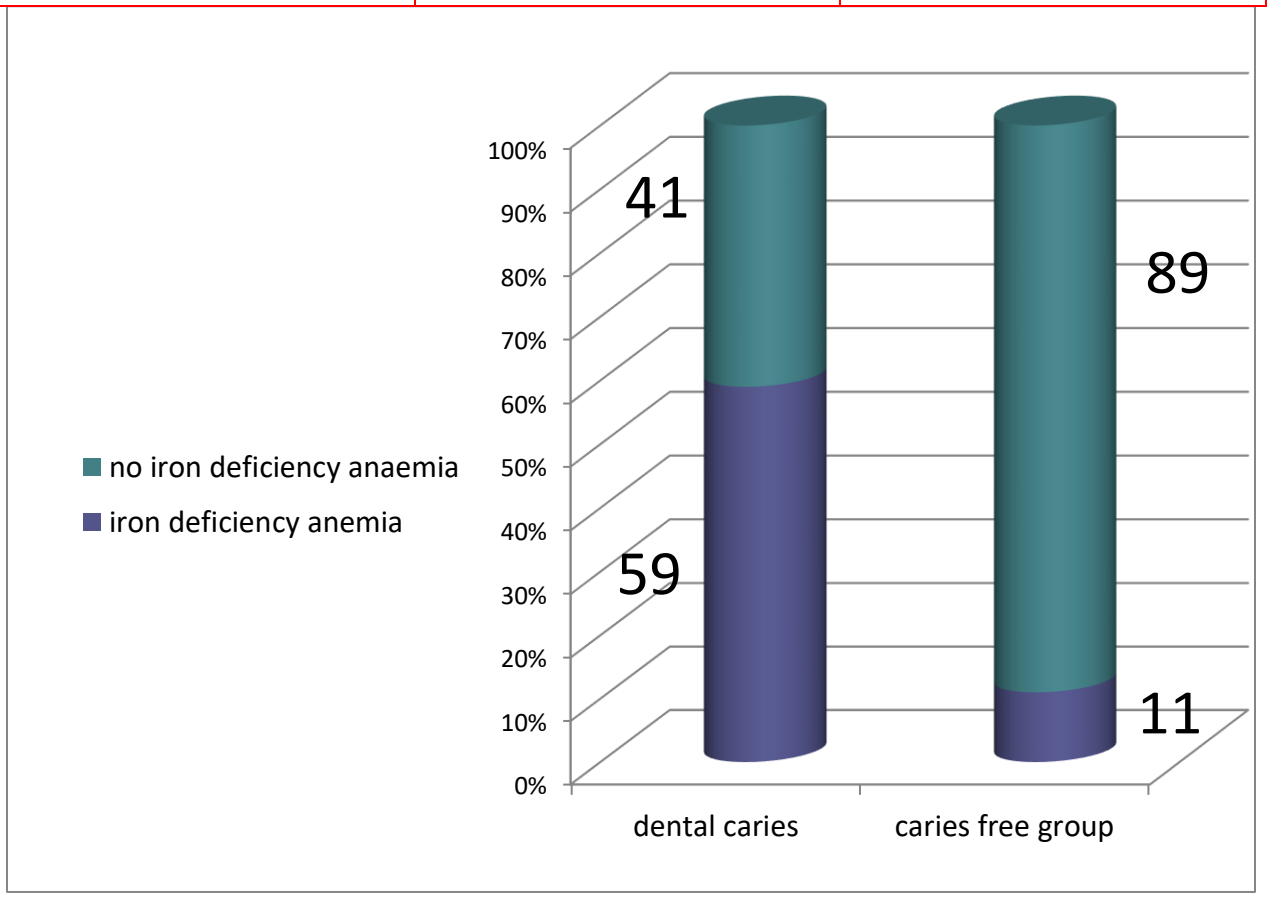


The habit of brushing tooth twice daily prevails around 10% in control group and only 4% in dental caries group.

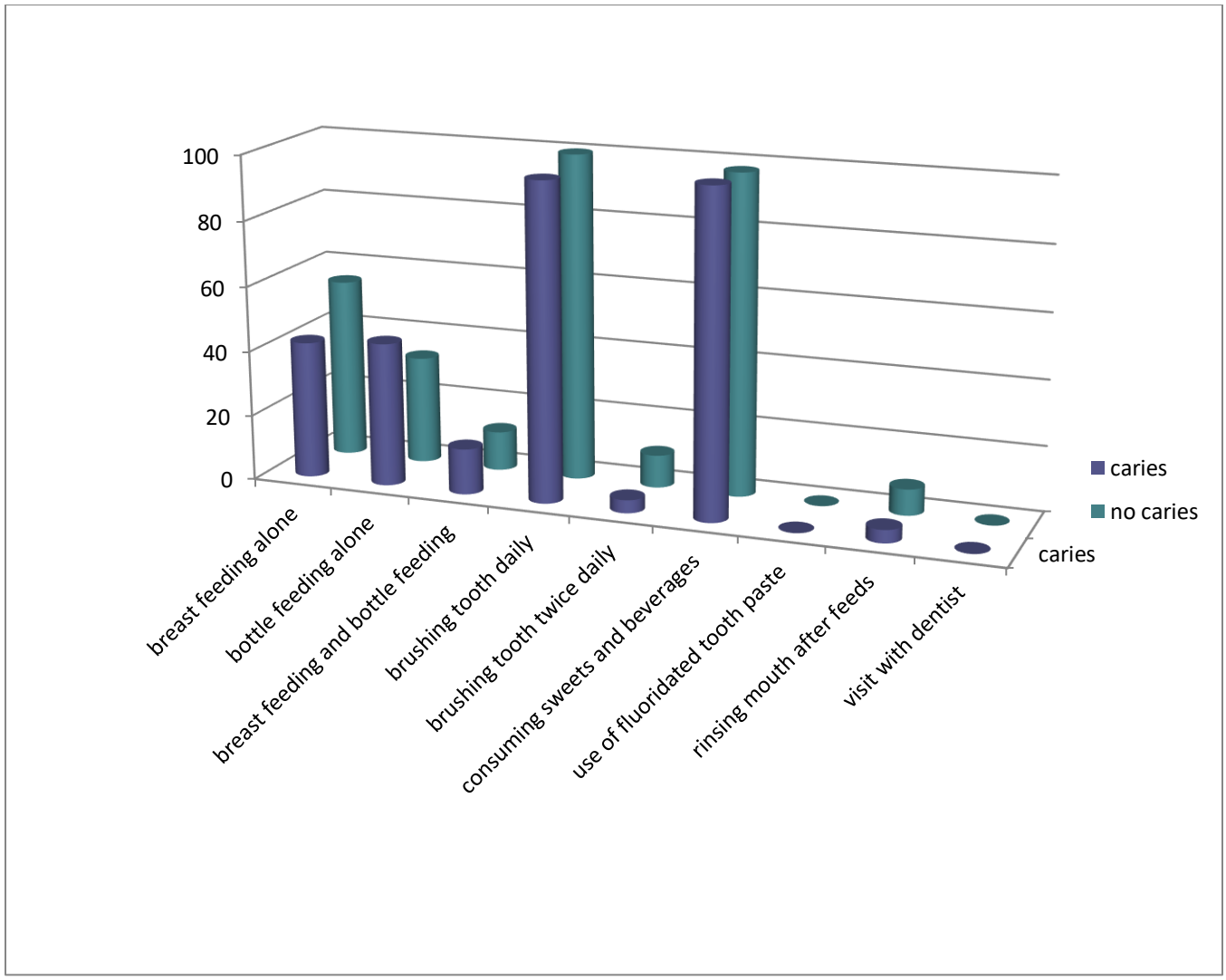


It is evident from the above scatter plot that there is an inverse relation between DMFT score and serum iron levels. That is, serum iron keeps on decreasing as the DMFT score increases.Thus severe dental caries is associated with iron deficiency anaemia.

Iron deficiency anemia	Dental caries	Caries free group
Yes	59	11
No	41	89



59 % of people with dental caries have iron deficiency anemia whereas only 11% of people without dental caries have iron deficiency anemia.41% of people with dental caries did not have iron deficiency anaemia whereas 89% of people without dental caries did not have iron deficiency anemia.



Bottle feeding is higher in children with dental caries.breast feeding alone is higher in children with no dental caries.behavioural changes like brushing tooth twice daily and rinsing mouth after feeds is higher in children with no dental caries.There is no significant difference in the consumption of sweets and beverages between the two groups.Use of fluoridated toothpaste and regular visits with dentist is nil for both groups.

RESULTS

Children attending outpatient clinic at Government Rajaji Hospital at Madurai and those admitted inpatient were screened for the presence of dental caries using DMFT score according to WHO.A total of 100 patients with dental caries is randomly selected . Age and sex matched controls without dental caries were also selected for comparison.

Among our study group of 100 people with dental caries, 55% are males and 45% are females.Majority of study group had a DMFT score of 2. The mean DMFT among the study group is 3. I n our study, DMFT score is inversely proportional to serum iron. This means, the serum iron is low for higher DMFT scores.The mean serum iron among the dental caries group is 49.32 and in control group is 98.75.The mean hemoglobin in the study group is 10.458 and 11.476 in the control group.The mean MCV in the caries group is 67.89 and in the control group is 75.75.The mean hematocrit in the caries group is 31.37 and 34.428 in the control group.This shows an association between iron deficiency anaemia and dental caries.The mean DMFT score is 1.803 in the breast feeding group and 3.276 in the bottle feeding group.This again confirms the known fact that breast feeding

protects against caries and bottle feeding promotes caries.There is no significant difference in the DMFTscore between people who brush daily or rinse their mouth after feeds.

DISCUSSION

Dental caries is the most common chronic disease of mankind among all the oral diseases. Iron deficiency anemia is the most common nutritional disorder affecting 30% of the population worldwide.Since both dental caries and iron deficiency anemia are a problem in epidemic proportions, it would be useful if an association between both can be proved thereby helping to formulate strategic plans to control both .Dental caries is an irreversible microbial disease caused by cariogenic bacterium streptococcus mutans characterized by demineralisation of inorganic and destruction of organic portions of the tooth.[125] Anemia is defined as reduction in red cell mass or hemoglobin concentration. Iron deficiency is the most common nutritional deficiency in the world. According to the Fourth National Health And Nutrition Examination Survey (NHANES IV), iron deficiency without anemia is seen in 7% of toddlers aged 1 to 2 years, 9% of adolescent girls and 16% of women belonging to child bearing age.[126] Poor socioeconomic status and bottle feeding practices play a role in the prevalence of iron deficiency anaemia.

Both iron deficiency and iron deficiency anaemia affects neurocognitive function in infancy. A detailed study of the National Health and Nutrition Examination Survey (NHANES) conducted between 1999 and 2004 reported among individuals of 20–34 years had 85.58% DMFT, 35–50 years had 94.30% DMFT, and 50–64 years had 95.62% DMFT.[127]

In our study,the mean Hemoglobin,MCV and serum iron is found to be decreased in children with dental caries.Healthy Behavioural changes like brushing teeth twice daily and rinsing mouth after feeds has a significant impact on reducing the incidence of dental caries. Breast feeding is found to protect against dental caries.Bottle feeding is shown to promote the development of dental caries. More severe the dental caries (Higher DMFT) is, more less the serum iron becomes.Thus our study also shows a significant association between dental caries and iron deficiency anaemia as demonstrated in many previous studies.

Rosalen et al in 1996 showed in his study that iron is capable of decreasing caries development in desalivated rats.[128] Devulapalle and Mooser et al in 2001 have concluded that iron ions are strong inhibitors of glucose transferase enzyme.[129] Clarke et al in 2006 portrayed the importance of iron in the development of dental

caries. [130].Berlutti et al in 2004 showed the role of iron in the protection of oral cavity from streptococcus mutans pathogenicity.[131]

Thakib et al in 2009 Showed the invitro cariostatic effects of various iron supplements.[132]Dietary sucrose is fermented with the help of oral cariogenic bacteriaforming lactic acid. This acid causes demineralization of enamel and dentine resulting in dental caries.Iron is said to have a restorative function causing remineralisation of enamel. Gels and crystals of hydrous iron oxides have the potential to nucleate salivary calcium and phosphate ions as apatites on the surface of the enamel thereby replacing the minerals which have been lost during caries process.[133]

Saliva contributes to the oral health of an individual.Saliva is a mixture of proteins, glycoproteins, electrolytes, small organic molecules and compounds transported from blood. Saliva also has buffer systems to neutralize the acid load in the oral cavity after a high carbohydrate diet. Previous studies have thrown light on the fact that high salivary flow rate and intact buffering system of the saliva helps in the prevention of dental caries.

Mahantesha et al in 2014 showed that salivary buffering capacity improved after treating patients with iron.[134]Prolonged bottle feeding is associated with the risk

of iron deficiency anemia and also dental caries .Johansson et al in 1994 pointed out that iron deficiency anaemia in rats impairs salivary peroxidase system .The lactoperoxidase system has an excellent influence in preventing early childhood caries by reducing the number of colonies formed by the cariogenic bacteria.[135]

Devulapalle et al in 2001 showed a simple method of inactivating this enzyme making use of Fenton reaction which requires the presence of iron or copper and peroxide.The hydroxyl radical ions produced in Fenton reaction inactivate Glucosyl transferase, an important factor in the production of dental caries.[136]

Buche et al in 2017 showed that salivary iron were low in children with dental caries experience..[137]Shaoul et al in 2012 showed a significant relationship between dental caries and iron deficiency anemia.[138[Koppal et al observed a definite relationship of iron deficiency anemia with Early Childhood Caries(ECC).[139]

Similar to the above mentioned studies, our study also adds on to establish an association between dental caries and iron deficiency anemia.

CONCLUSION:

The findings of the present study revealed that children with dental caries appear to be at significantly greater chance of having lower serum iron and lower hemoglobin levels compared to control group.Thus, children with dental caries appear to be at significantly greater risk for iron deficiency anemia than cavity free children.Hence our study concludes that dental caries is associated with iron deficiency anemia.Our study also shows a higher DMFT score with bottle feeding children compared to breast feeding children.Through our study, we come to know that dental caries may serve as a risk marker for iron deficiency anemia.Health behavioural interventions targeting dental caries could help prevent iron deficiency anemia thereby reducing the health burden of community.Further studies have to be done in larger populations to really refute or support an association between dental caries and iron deficiency anemia.

LIMITATIONS:

1) Our study is done only with a small sample size.Future studies aimed at large population are largely needed.
2) Randomisation from small subset of population may contribute to bias.

RECOMMENDATIONS:

On the basis of the results of the above study we made the following recommendations:

1) Hemoglobin has to be routinely checked in any child with dental caries.
2) School health programmes should also focus more on providing awareness about oral hygiene .
3) Children registered in DEIC should also get screened for dental caries by 1 year of age in addition to opthalmological and ENT screening.

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LIST OF ABBREVIATIONS USED

DMFT	:Decayed Missed Filled Teeth
SiC	:Significant Caries Index
ATP	:Adenosine Tri Phosphate
DMT1	:Divalent Metal Transporter 1
FPN1	:Ferroportin 1
RBC	:Red Blood Cell
TIBC	:Total Iron Binding Capacity
Tfr1	:Transferrin receptor 1
MFRN1	:Mitoferrin
NTBI	:Non Transferrin Bound Iron
IRP	:Iron Regulatory Protein
IRE	:Iron Responsive Elements
mRNA	:messenger Ribo Nucleic Acid
UTR	:Un Translated Region
IDA	:Iron Deficiency Anemia
CDC	:Centre for Disease Control and Prevention
MCV	:Mean Corpuscular Volume
RDW	:Red blood cell Distribution Width
CRP	:C Reactive Protein
ZPP	:Zinc Proto Porphyrin
CHr	:Cellular Hemoglobin of reticulocyte

RDA	:Recommended Dietary Allowance
WHO	:World Health Organisation
FAO	:Food and Agricultural Organisation
USA	:United States of America
AAP	:AmericaN Academy of Pediatrics
DIFOTI	:Digital Imaging Fibre Optic Transillumination
QLF	:Quantitative Light induced Fluorescence
ECM	:Electronic Caries Monitor
ADA	:American Dental Association
FDI	:Federation Dentaire International
DMFS	:Decayed Missed Filled tooth Surfaces
NHANES	:National Health And Nutrition Examination Survey
HCT	:Haematocrit
SPSS	:Statistical Package for the Social Sciences
ECC	:Early Childhood Caries
DEIC	:District Early Intervention Centre
ENT	:Ear Nose and Throat

PROFORMA

NAME :

AGE :

SEX :

INFORMANT :

RELIABILITY :

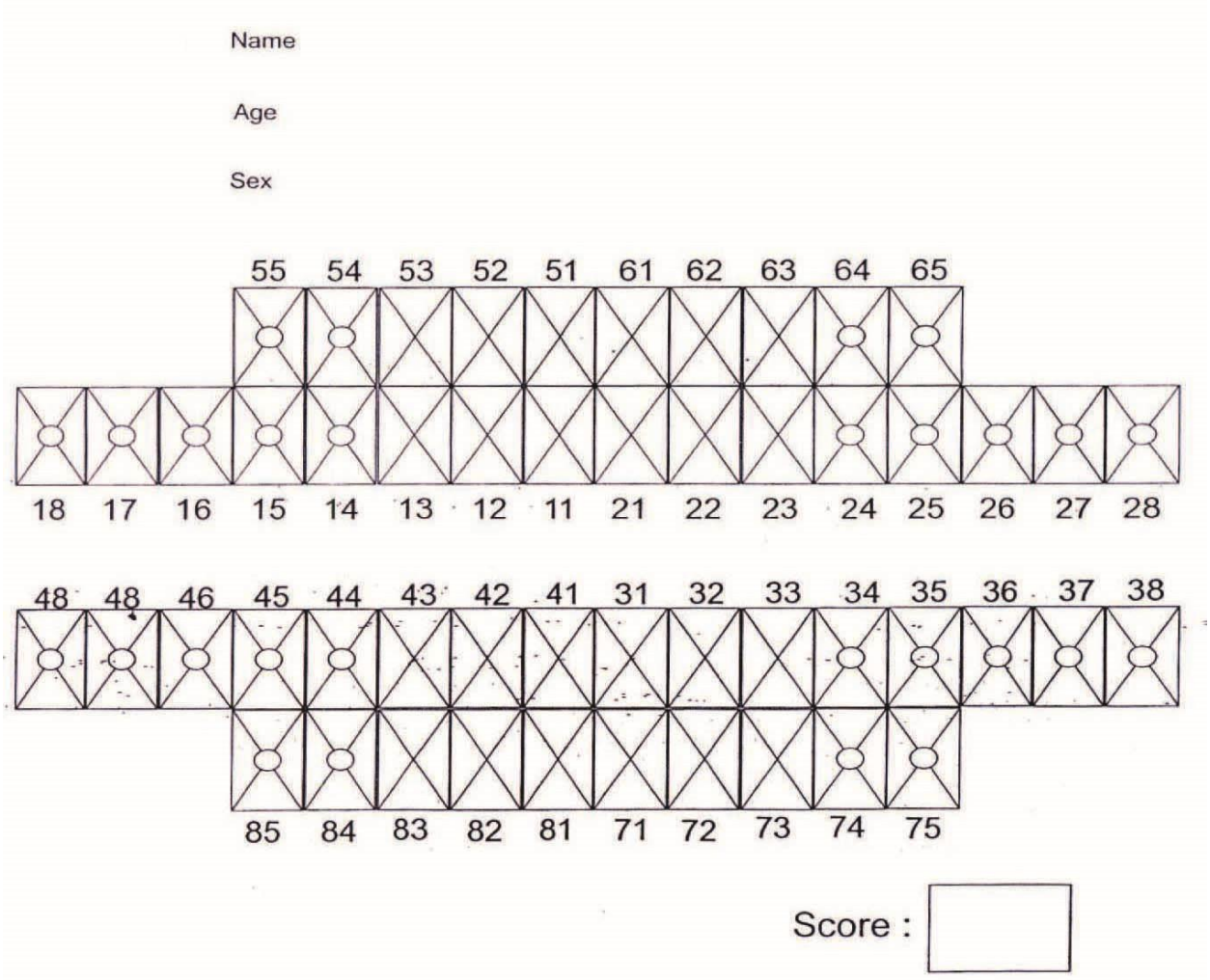
ADDRESS :

BRIEF HISTORY :

WEIGHT :

DENTAL FORMULA :

DMFT SCORE


Name
Age
Sex
55 54 53 52 51 61 62 63 64 65
18 17 16 15 14 13 12 11 21 22 23 24 25 26 27 28
48 48 46 45 44 43 42 41 31 32 33 34 35 36 37 38
85 84 83 82 81 71 72 73 74 75
Score :

INVESTIGATIONS:

Hemoglobin :

MCV :

Haematocrit :

Serum iron :

OTHER DETAILS

QUESTIONNAIRE:

Note:(parents can help filling the questionnaire) YES/NO format

1) Have you been exclusively brestfed?
2) Have you been bottle fed anytime?
3) Do you brush your teeth everyday? (caretaker brushing your teeth also included)
4) Do you brush your teeth twice daily?
5) Do you rinse your mouth after taking feeds?
6) Do you have the habit of using fluoridated toothpaste?
7) Do you have regular visits with dentist even without symptoms for screening dental caries?
8) Do you consume sweets/beverages atleast twice in a week?

CONSENT FORM

I hereby give consent to participate in the study conducted by DR.MEKHA PREM, post graduate in the Institute of Pediatrics,Madurai Medical College, Madurai and to use my personal clinical data and result of investigations for the purpose of analysis and to study the association of dental caries with iron deficiency anemia. I also give consent for further investigations.

Place: Signature of

Date: parent/guardian

MASTER CHART

119 s no	ip/op no	age	sex	DMFT score	Hb	MCV	HCT	serum iron(microgram/dl)	breast feeding	bottle feeding	brushing tooth daily	brushing tooth twice daily	consuming sweets and beverages	use of fluoridated toothpaste	rinsing mouth after each feed	visit with dentist
1	65890	3	m	4	10.6	66	31.8	15	no	yes	yes	no	yes	No	no	No
2	65990	2	m	4	10.5	68	31.5	18	no	yes	no	no	yes	No	no	No
3	65993	5	m	3	11	70	33	20	no	yes	yes	no	yes	No	no	No
4	66712	1	f	2	10.9	69	32.7	21	no	yes	no	no	no	No	no	No
5	66834	2	f	1	11	75	33	53	no	yes	yes	no	yes	No	yes	No
6	66954	6	m	3	11.3	73	33.9	33	yes	yes	yes	no	yes	No	no	No
7	66974	7	f	4	11.2	70	33.6	25	yes	yes	yes	no	yes	No	no	No
8	67234	8	m	2	11.5	73	34.5	37	no	yes	yes	no	yes	No	no	No
9	67567	3	m	4	9	56	27	13	no	yes	yes	no	yes	No	no	No
10	67890	4	m	3	10	69	30	21	no	yes	yes	no	yes	No	no	No
11	67990	3	f	3	11	73	33	26	yes	yes	yes	no	yes	No	no	No
12	68990	6	m	2	11.5	76	34.5	45	no	yes	yes	no	yes	No	yes	No
13	69345	7	f	1	12	79	36	110	yes	no	yes	yes	yes	No	yes	No
14	70213	8	m	2	12.2	80	36.6	133	no	yes	yes	no	yes	No	no	No
15	70333	10	f	2	12	76	36	30	no	yes	yes	yes	yes	No	no	No
16	70678	12	f	4	10	65	30	22	no	yes	yes	no	yes	No	no	No
17	70897	3	m	5	8.8	62	26.4	11	no	yes	yes	no	yes	No	no	No
18	70990	3	m	4	8.5	58	25.5	13	no	yes	yes	no	yes	No	no	No
19	71223	7	f	3	11	70	33	28	yes	yes	yes	no	yes	No	no	No
20	72222	9	m	2	12.5	78	37.5	120	no	yes	yes	no	yes	No	no	No
21	72456	7	f	2	12	78	36	106	no	yes	yes	no	yes	No	no	No
22	72555	5	f	4	10.2	63	30.6	20	no	yes	yes	no	yes	No	no	No
23	72678	3	m	3	9	56	27	16	no	yes	yes	no	yes	No	no	No
24	73456	4	m	3	9.2	57	27.6	20	no	yes	yes	no	yes	No	no	No
25	74567	3	m	3	9.4	55	28.2	18	yes	yes	yes	no	yes	No	no	No
26	75901	8	f	2	12	76	36	67	no	yes	yes	no	yes	No	no	No
27	76789	11	m	1	12.5	82	37.5	89	no	yes	yes	yes	yes	No	no	No
28	76889	4	m	1	11.5	77	34.5	112	no	yes	yes	yes	yes	No	no	No
29	76990	6	f	2	11.7	77	35.1	49	yes	no	yes	no	yes	No	no	No
30	76998	1	m	2	11	70	33	29	no	yes	yes	no	yes	No	no	No
31	80000	2	m	2	8.1	51	24.3	20	no	yes	yes	no	yes	No	no	No
32	80123	4	m	3	8.4	54	25.2	11	yes	yes	yes	no	yes	No	no	No
33	80167	3	f	4	8	49	24	14	yes	yes	yes	no	yes	No	no	No

34	80178	7	m	3	9.7	62	29.1	30	no	yes	yes	no	yes	No	no	No
35	80234	6	m	2	11	70	33	34	no	yes	yes	no	yes	No	no	No
36	80245	9	m	2	12.2	74	36.6	87	yes	no	yes	no	yes	No	no	No
37	80345	11	f	1	12.2	76	36.6	76	no	yes	yes	no	yes	No	no	No
38	80456	12	f	1	12.5	80	37.5	94	no	yes	yes	no	yes	No	no	No
39	80555	3	f	3	7.8	48	23.4	15	yes	yes	No	no	yes	No	no	No
40	80567	3	m	4	8	51	24	12	yes	yes	yes	no	yes	No	no	No
41	80667	5	m	2	9	55	27	20	yes	yes	yes	no	yes	No	no	No
42	80678	3	f	3	8.2	53	24.6	17	no	yes	yes	no	yes	No	no	No
43	80778	6	f	3	11	70	33	36	no	yes	yes	no	yes	No	no	No
44	80888	3	m	2	11.5	75	34.5	67	yes	no	yes	no	yes	No	no	No
45	80900	7	m	1	11.5	76	34.5	98	yes	no	yes	no	yes	No	no	No
46	80923	7	m	1	11.2	75	33.6	128	yes	no	yes	no	yes	No	no	No
47	81111	3	m	2	8	60	24	21	yes	no	yes	no	yes	No	no	No
48	81222	12	f	2	12.5	78	37.5	76	yes	no	yes	no	yes	No	no	No
49	81322	11	f	1	12.5	79	37.5	124	yes	no	yes	no	yes	No	no	No
50	81345	9	m	2	12.2	78	36.6	117	yes	no	yes	no	yes	No	no	No
51	81444	7	f	2	9.2	59	27.6	32	yes	no	yes	no	yes	No	no	No
52	81543	8	m	1	12.1	77	36.3	134	yes	no	yes	no	yes	No	no	No
53	81567	9	f	1	13	80	39	125	yes	no	yes	no	yes	No	no	No
54	81577	7	m	1	12	78	36	126	yes	no	yes	no	yes	No	no	No
55	81588	12	m	1	13	82	39	124	yes	no	yes	no	yes	No	no	No
56	81677	3	f	3	7.5	52	22.5	12	no	yes	yes	no	yes	No	no	No
57	81688	3	f	4	7.2	49	21.6	9	no	yes	yes	no	yes	No	no	No
58	81765	7	m	2	10.5	70	31.5	28	yes	no	yes	no	yes	No	no	No
59	81888	6	f	2	10.2	69	30.6	33	yes	no	yes	no	yes	No	no	No
60	81898	6	f	2	11	70	33	35	yes	no	yes	no	yes	No	no	No
61	81992	8	f	1	11.5	72	34.5	37	yes	no	yes	no	yes	No	no	No
62	81998	9	f	1	12	76	36	79	yes	no	yes	no	yes	No	no	No
63	82456	11	m	1	12	75	36	56	yes	yes	yes	no	yes	No	no	No
64	82567	12	m	1	12.2	80	36.6	128	no	yes	yes	no	yes	No	no	No
65	82578	3	m	3	7.3	47	21.9	12	no	yes	yes	no	yes	No	no	No
66	82579	6	m	2	8.5	63	25.5	35	yes	no	yes	no	yes	No	no	No
67	82580	7	m	2	9.5	65	28.5	26	no	yes	yes	no	yes	No	no	No
68	82590	3	m	4	7.4	47	22.2	16	no	yes	yes	no	yes	No	no	No
69	82660	7	f	2	9.5	68	28.5	35	no	yes	yes	no	yes	No	no	No
70	82668	4	f	4	8.2	65	24.6	11	no	yes	yes	no	yes	No	no	No
71	82669	9	m	2	12	76	36	48	yes	no	yes	no	yes	No	no	No
72	82773	11	f	1	12.2	80	36.6	68	yes	no	yes	no	yes	No	no	No
73	82774	9	f	1	12	76	36	99	yes	no	yes	no	yes	No	no	No
74	82775	11	m	1	13	82	39	120	yes	no	yes	no	yes	No	no	No

75	82832	12	f	1	13	83	39	124	yes	yes	yes	no	yes	No	no	No
76	82845	4	m	4	7.2	45	21.6	11	no	yes	yes	no	yes	No	no	No
77	82854	3	f	3	6.7	45	20.1	12	yes	yes	yes	no	yes	No	no	No
78	82863	7	f	2	11	70	33	38	no	yes	yes	no	yes	No	no	No
79	82876	3	f	4	7.1	48	21.3	13	no	yes	yes	no	yes	No	no	No
80	82897	2	m	2	8.5	56	25.5	20	yes	no	yes	no	yes	No	no	No
81	82898	2	m	2	9.5	63	28.5	21	yes	no	yes	no	yes	No	no	No
82	82902	1	f	1	9.3	62	27.9	20	yes	no	yes	no	yes	No	no	No
83	82908	8	f	2	11.5	73	34.5	36	yes	no	yes	no	yes	No	no	No
84	82909	9	f	1	12.2	78	36.6	115	yes	yes	yes	no	yes	No	no	No
85	82917	11	m	1	12.2	79	36.6	76	yes	no	yes	no	yes	No	no	No
86	82925	3	m	4	7.4	49	22.2	12	no	yes	yes	no	yes	No	no	No
87	82935	7	m	2	10	56	30	33	yes	no	yes	no	yes	No	no	No
88	82950	3	f	4	6.8	42	20.4	16	no	yes	yes	no	yes	No	no	No
89	82967	9	f	2	10	70	30	37	yes	no	yes	no	yes	No	no	No
90	82987	1	m	1	8.9	62	26.7	21	yes	no	yes	no	yes	No	yes	No
91	82990	11	m	1	12.5	78	37.5	65	yes	no	yes	no	yes	No	no	No
92	82997	12	f	1	12.3	80	36.9	89	yes	no	yes	no	yes	No	no	No
93	82998	6	m	1	11	72	33	37	yes	no	yes	no	yes	No	no	No
94	82999	7	m	1	11.2	74	33.6	36	yes	no	yes	no	yes	No	no	No
95	83001	8	m	2	12	77	36	39	yes	no	yes	no	yes	No	no	No
96	83007	3	f	2	9.5	68	28.5	20	yes	no	yes	no	yes	No	no	No
97	83009	7	f	2	10.2	65	30.6	38	yes	no	yes	no	yes	No	no	No
98	83016	9	m	2	12.2	78	36.6	57	yes	no	yes	no	yes	No	no	No
99	83019	11	m	1	12.5	80	37.5	90	yes	no	yes	no	yes	No	no	No
100	83028	3	f	4	7.6	67	22.8	11	no	yes	yes	no	yes	No	no	No

s no	ip/op no	age	Sex	DMFT score	Hb	MCV	HCT	serum iron(microgram/dl)	breast feeding	bottle feeding	brushing tooth daily	brushing tooth twice daily	consuming sweets and beverages	use of fluoridated toothpaste	rinsing mouth after each feed	visit with dentist
101	83029	3	m	0	9.5	67	28.5	37	yes	no	yes	No	yes	No	no	No
102	83038	2	m	0	11.5	76	34.5	128	yes	no	yes	No	yes	No	no	No
103	83056	5	m	0	12	78	36	123	yes	no	yes	No	yes	No	no	No
104	83098	1	f	0	11.5	73	34.5	74	yes	no	yes	No	yes	No	no	No
105	83123	2	f	0	11.4	75	34.2	86	no	yes	yes	No	yes	No	no	No
106	83156	6	m	0	12.5	80	37.5	118	no	yes	yes	No	yes	No	no	No
107	83167	7	f	0	12.5	80	37.5	129	no	yes	yes	No	yes	No	no	No
108	83267	8	m	0	12.5	78	37.5	76	no	yes	yes	No	yes	No	no	No
109	83297	3	m	0	11	73	33	35	yes	no	yes	No	yes	No	no	No
110	83300	4	m	0	8.2	60	24.6	17	yes	no	yes	No	yes	No	no	No
111	83304	3	f	0	10.5	70	31.5	20	yes	no	yes	No	yes	No	no	No
112	83309	6	m	0	9.5	68	28.5	37	yes	no	yes	no	yes	no	no	No
113	83407	7	f	0	11.7	77	35.1	132	yes	no	yes	no	yes	no	no	No
114	83500	8	m	0	12.2	79	36.6	116	no	yes	yes	yes	yes	no	no	No
115	83506	10	f	0	12.2	78	36.6	119	no	yes	yes	no	yes	no	no	No
116	83507	12	f	0	12	83	36	130	yes	yes	yes	no	yes	no	no	No
117	83519	3	m	0	8.7	62	26.1	20	yes	no	yes	no	yes	no	no	No
118	83520	3	m	0	11.2	75	33.6	68	no	yes	yes	no	yes	no	no	No
119	83529	7	f	0	12	78	36	123	yes	yes	yes	no	yes	no	no	No
120	83567	9	m	0	12.2	77	36.6	127	yes	yes	yes	no	yes	no	no	No
121	83587	7	f	0	11.7	78	35.1	124	no	yes	yes	no	yes	no	no	No
122	83597	5	f	0	11.5	77	34.5	130	no	yes	yes	no	yes	no	no	No
123	83598	3	m	0	9.5	66	28.5	20	yes	no	yes	no	yes	no	no	No
124	83599	4	m	0	11	73	33	28	yes	no	yes	no	yes	no	no	No
125	83612	3	m	0	11.5	79	34.5	47	no	no	yes	no	yes	no	no	No
126	83622	8	f	0	12.2	80	36.6	87	yes	no	yes	no	yes	no	no	No
127	83623	11	m	0	12.4	83	37.2	95	yes	no	yes	no	yes	no	no	No
128	83657	4	m	0	9.2	64	27.6	20	yes	no	yes	no	yes	no	no	No
129	83667	6	f	0	11.8	79	35.4	126	no	yes	yes	no	yes	no	no	No
130	83668	1	m	0	11.2	73	33.6	117	no	yes	yes	no	yes	no	no	No
131	83678	2	m	0	11.5	75	34.5	129	yes	no	yes	yes	yes	no	no	No
132	83690	4	m	0	11.3	75	33.9	132	yes	no	yes	no	yes	no	no	No
133	83699	3	f	0	11.4	74	34.2	127	no	yes	yes	no	yes	no	no	No
134	83700	7	m	0	12	79	36	69	no	yes	yes	no	yes	no	yes	No

135	83701	6	m	0	11.7	79	35.1	111	yes	no	yes	no	yes	no	no	No
136	83703	9	m	0	12.3	80	36.9	123	no	yes	yes	no	yes	no	no	No
137	83705	11	f	0	12.5	83	37.5	134	yes	no	yes	no	yes	no	no	No
138	83708	12	f	0	12.4	82	37.2	115	no	yes	yes	yes	yes	no	no	No
139	83709	3	f	0	8.2	56	24.6	20	yes	no	yes	no	yes	no	no	No
140	83712	3	M	0	11	73	33	89	no	yes	yes	no	yes	no	no	No
141	83715	5	m	0	11.7	77	35.1	131	yes	no	yes	no	yes	no	no	No
142	83734	3	f	0	11.2	75	33.6	120	yes	no	yes	no	yes	no	no	No
143	83745	6	f	0	11.8	79	35.4	117	yes	no	yes	no	yes	no	no	No
144	83756	3	m	0	11.3	75	33.9	118	yes	no	yes	no	yes	no	no	No
145	83765	7	m	0	12	80	36	127	yes	yes	yes	no	yes	no	no	No
146	83768	7	m	0	12.2	83	36.6	133	yes	yes	yes	no	yes	no	no	No
147	83880	3	m	0	11.2	74	33.6	99	yes	no	yes	no	yes	no	no	No
148	73882	12	f	0	12.6	82	37.8	131	yes	no	yes	no	yes	no	no	No
149	73885	11	f	0	12.3	79	36.9	122	yes	no	yes	no	yes	no	no	No
150	73889	9	m	0	12.4	79	37.2	120	no	yes	yes	yes	yes	no	no	No
151	73990	7	f	0	11.7	77	35.1	95	no	yes	yes	no	yes	no	yes	No
152	73992	8	m	0	12.8	84	38.4	79	yes	no	yes	no	yes	no	no	No
153	74210	9	f	0	12.4	79	37.2	124	yes	no	yes	no	yes	no	no	No
154	74223	7	m	0	12	78	36	121	yes	no	yes	no	yes	no	no	No
155	74226	12	m	0	13	83	39	131	yes	yes	yes	no	yes	no	no	No
156	74228	3	f	0	11.3	75	33.9	111	yes	no	yes	yes	yes	no	no	No
157	74229	3	f	0	11.5	76	34.5	102	no	yes	yes	no	yes	no	no	No
158	74230	7	m	0	11.8	78	35.4	122	no	yes	yes	no	yes	no	no	No
159	74234	6	f	0	11.9	80	35.7	130	yes	no	yes	no	yes	no	no	No
160	74244	6	f	0	8	52	24	36	yes	no	yes	no	yes	no	no	No
161	74255	8	f	0	12.3	79	36.9	114	no	yes	yes	no	yes	no	no	No
162	74265	9	f	0	12.2	79	36.6	119	yes	yes	yes	no	yes	no	no	No
163	74268	11	m	0	12.5	80	37.5	135	yes	yes	yes	no	yes	no	no	No
164	74278	12	m	0	12.5	80	37.5	111	yes	no	yes	no	yes	no	no	No
165	74288	3	m	0	9.5	69	28.5	89	no	yes	yes	no	yes	no	no	No
166	74298	6	m	0	11.9	79	35.7	56	no	yes	yes	no	yes	no	no	No
167	74311	7	m	0	12	79	36	99	yes	no	yes	yes	yes	no	no	No
168	74312	3	m	0	11	73	33	56	yes	no	yes	no	yes	no	no	No
169	74333	7	f	0	11.5	75	34.5	111	yes	yes	yes	no	yes	no	no	No
170	74346	4	f	0	11.5	76	34.5	123	yes	no	yes	no	yes	no	yes	No
171	74347	9	m	0	12.2	78	36.6	132	yes	no	yes	no	yes	no	no	No
172	74348	11	f	0	13	83	39	135	no	yes	yes	no	yes	no	no	No
173	74349	9	f	0	12.2	78	36.6	112	no	yes	yes	no	yes	no	no	No
174	74359	11	m	0	12	76	36	123	yes	no	yes	no	yes	no	no	No
175	74360	12	f	0	12	78	36	70	yes	no	yes	no	yes	no	no	No

176	74362	4	m	0	8.5	67	25.5	20	yes	no	yes	no	yes	no	no	No
177	74365	3	f	0	11.2	75	33.6	98	yes	no	yes	no	yes	no	no	No
178	74369	7	f	0	11.8	78	35.4	124	yes	no	yes	yes	yes	no	yes	No
179	74370	3	f	0	11.5	78	34.5	87	no	yes	yes	no	yes	no	no	No
180	74372	2	m	0	11.2	76	33.6	97	no	yes	yes	no	yes	no	no	No
181	74382	2	m	0	11.2	77	33.6	109	yes	no	yes	yes	yes	no	yes	No
182	74392	1	f	0	7.3	48	21.9	20	yes	no	yes	no	no	no	no	No
183	74399	8	f	0	12.2	79	36.6	132	yes	no	yes	no	yes	no	no	No
184	74400	9	f	0	12.5	82	37.5	119	yes	no	yes	no	yes	no	no	No
185	74412	11	m	0	12.4	80	37.2	105	yes	no	yes	no	yes	no	no	No
186	74432	3	m	0	11.5	76	34.5	85	yes	no	yes	no	yes	no	no	No
187	74443	7	m	0	11.8	79	35.4	120	yes	yes	yes	yes	yes	no	yes	No
188	74456	3	f	0	11.2	74	33.6	123	yes	yes	yes	no	yes	no	no	No
189	74467	9	f	0	12.6	80	37.8	113	yes	no	yes	no	yes	no	no	No
190	74489	1	m	0	11	70	33	56	yes	no	yes	no	no	no	no	No
191	74567	11	m	0	12	76	36	94	yes	yes	yes	no	yes	no	no	No
192	74587	12	f	0	12.1	79	36.3	123	no	yes	yes	no	yes	no	no	No
193	74597	6	m	0	11.7	78	35.1	120	no	yes	yes	yes	yes	no	yes	No
194	74598	7	m	0	11.6	76	34.8	120	yes	no	yes	no	yes	no	no	No
195	74623	8	m	0	12.2	78	36.6	134	no	yes	yes	no	yes	no	no	No
196	74634	3	f	0	11.5	79	34.5	67	yes	no	yes	no	yes	no	no	No
197	74645	7	f	0	11.8	76	35.4	87	no	yes	yes	no	yes	no	yes	No
198	74655	9	m	0	12.5	82	37.5	123	no	yes	yes	no	yes	no	no	No
199	74667	11	m	0	12.2	76	36.6	133	yes	no	yes	no	yes	no	no	No
200	74770	3	f	0	7.6	54	22.8	54	yes	no	yes	no	yes	no	no	no