The History of Deferiprone (L1) and the Paradigm of the Complete Treatment of Iron Overload in Thalassaemia

George J. Kontoghiorghes, Marios Kleanthous and Christina N. Kontoghiorghe.

Postgraduate Research Institute of Science, Technology, Environment and Medicine, Limassol, Cyprus.

Competing interests: George J. Kontoghiorghes, the first author of this paper, was responsible for the chemical development of iron chelators, in particular of the orally active L1 (Deferiprone), and he was the driving force in further investigations of L1 for treating patients with severe iron overload and other conditions. The Authors declare no conflict of interest. The authors have no commercial associations including consultancies, stock ownership, equity interest, patent/licensing arrangement etc. that might pose a conflict of interest in connection with the submitted article.

Abstract. Deferiprone (L1) was originally designed, synthesised and screened in vitro and in vivo in 1981 by Kontoghiorghes G. J. following his discovery of the novel alpha-ketohydroxypyridine class of iron chelators (1978-1981), which were intended for clinical use. The journey through the years for the treatment of thalassaemia with L1 has been a very difficult one with an intriguing turn of events, which continue until today. Despite many complications, such as the extensive use of L1 suboptimal dose protocols, the aim of chelation therapy- namely, the complete removal of excess iron in thalassaemia major patients, has been achieved in most cases following the introduction of specific L1 and L1/deferoxamine combinations. Many such patients continue to maintain normal iron stores. Thalassemia has changed from a fatal to chronic disease; also thanks to L1 therapy and thalassaemia patients are active professional members in all sectors of society, have their own families with children and grandchildren and their lifespan is approaching that of normal individuals. No changes in the low toxicity profile of L1 have been observed in more than 30 years of clinical use and prophylaxis against the low incidence of agranulocytosis is maintained using mandatory monitoring of weekly white blood cells’ count. Thousands of thalassaemia patients are still denied the cardioprotective and other beneficial effects of L1 therapy. The safety of L1 in thalassaemia and other non-iron loaded diseases resulted in its selection as one of the leading therapeutics for the treatment of Friedreich’s ataxia, pantothenate kinase-associated neurodegeneration and other similar cases. There are also increasing prospects for the application of L1 as a main, alternative or adjuvant therapy in many pathological conditions including cancer, infectious diseases and as a general antioxidant for diseases related to free radical pathology.

Keywords: Deferiprone; Iron overload; Thalassaemia; Chelation therapy; Iron detoxification; Deferoxamine.

Introduction. The haemoglobinopathies, which include sickle cell disease and thalassaemia, are a major group of genetic diseases affecting humans. It is estimated that about 100000 children are born annually.
with thalassaemia mainly in South-East Asia, the Middle East and Mediterranean countries. Most of the thalassaemia patients are born in South-East Asia and die untreated.1 2

Thalassaemia is endemic in some countries such as Cyprus, where 1 in 6 persons is an asymptomatic thalassaemia heterozygote carrier, and about 1 in 1000 is a thalassaemia major and intermedia patient. The prevention and treatment programmes for thalassaemia and especially chelation therapy impose a great financial burden on the health budget of many countries.1 3

The standard form of treatment of transfusion dependent thalassaemia (TDT) is regular red blood cell (RBC) transfusions every 1-4 weeks accompanied by daily chelation therapy. Multiple transfusions cause increased iron deposition and damage to the liver, heart, spleen and other organs. Iron overload in thalassaemia has the highest rate of morbidity and mortality of metal related intoxication in humans. Iron chelation therapy in thalassaemia and other transfusional iron loading conditions is carried out worldwide using the generic drugs deferoxamine (DF), deferasirox (DFRA).4 The combination of chelating drugs and especially the L1/DF combination, is also widely used in the vast majority of thalassaemia patients in Cyprus and also in some other countries.5 6

There is no worldwide consensus in the use of iron chelating drugs or of related protocols in transfusional iron loaded patients. In most cases, the selection of chelation therapy and related protocols is generally ‘random’ and based on subjective and other criteria and non specific aim. As a result, the selection and use of iron chelating drugs vary from country to country and from clinic to clinic.7

The main aim of iron chelation therapy in thalassaemia and other iron loaded conditions is the achievement and maintenance of normal iron stores, in which case patients are devoid of iron overload toxic side effects.8 This aim, including the long term prevention of iron overload, can be accomplished using effective and safe chelation protocols, involving mainly L1 and L1/DF combinations.8

Some of the unique pharmacological properties and characteristics of L1 such as the ability to penetrate most organs and remove effectively excess iron from the heart has resulted in a substantial reduction in the mortality rate of thalassaemia.4 6-9 10 Furthermore, the ability of L1 to remove excess iron from the brain has resulted in the development of L1 as one of the leading pharmaceuticals in the treatment of Friedreich’s ataxia, pantothenate kinase-associated neurodegeneration (PKAN), and other cases of neurodegeneration with brain iron accumulation.11-13

Design, Development, and Cost of Deferiprone. The design, development, and clinical use of L1 is a unique case of academic orphan drug development, which was originally based on academic efforts supported mainly by a thalassaemia patient/parent charitable organisation, namely the United Kingdom Thalassaemia Society (UKTS).

The project on chelation was initiated at the University of Essex, UK, while working on haemoglobins in 1979 and was partly supported by the British Technology Group (BTG) and the UKTS.14 15

Following a fundamental new approach on iron chelation design and testing, a new group of iron chelators was discovered by one of the authors- namely Kontoghiorghes G J (GJK), and as a result, the new classes of alpha-ketohydroxyridines including 1,2- dimethyl-3-hydroxypyrid-4-one (L1) were synthesised and tested at Essex University and University College Medical School London (UCH), UK.15 16 The latter was selected by GJK and members of UKTS because of the in vivo and clinical testing facilities.5 6

The discovery and iron removal effects of L1 could not be published for 5 years due to “embargo” on publications by BTG, and in 1985 the UKTS sponsored the continuation of the chelation project at the Royal Free Hospital Medical School (RF) London, UK from where the first publications of the iron removal effects of L1 in comparison to parenteral DF in animals were reported.19 22

A significant invention at the RF in 1986 was also the one-step novel synthesis of L1 and L1 analogues, instead of the 4-step synthesis invented in 1981, which overturned the BTG patent monopoly in many countries. The one-step synthetic method is currently utilised by all manufacturers of L1 worldwide and make L1 less expensive than DF and DFRA.23 27 Deferiprone became a generic drug about 15 years ago, and by comparison, its sale price in India, Iran and Thailand is about 5-10 times cheaper than that sold in western countries.5 6

A fierce competition against L1 and related controversies were in process from the time of the L1 discovery until today. For example, more than 60 patents were filed worldwide since the discovery of L1 and other alpha-ketohydroxyridines.15 The first patent application was filed in 1982.15 28 However, due to ‘policy changes’, BTG has submitted the remaining patents under the names of the inventor (GJK) and co-inventors using an alphabetical order format.29 30

An analogue of L1, namely 1,2-diethyl-3-hydroxypyrid-4-one (EL1NEt or CP 94) was promoted by BTG sponsored studies at Essex University and UCH.31 35 However, based on further studies and clinical trials in thalassaemia patients, EL1NEt was later abandoned.36-38 Similarly, Ciba Geigy (now Novartis) the then manufacturers of DF have also carried out animal toxicity studies with L1 and reached the conclusion that L1 was toxic.40 However, the evaluation methods used for L1, as well as the
comparative toxicity data obtained previously with DF questioned Ciba Geigy’s conclusions. Similar controversies continue until today.

Despite the opposition from different groups, the academic initiative and strategy for the development of L1 in academic institutions continued and included the general phase I to V studies as described for most other drugs.

**The First Clinical Trials with Deferiprone and Today’s Implications.** Approval for the first clinical trials with L1 in myelodysplasia and thalassaemia patients was obtained in 1987 from the local ethical committee of the RF and the Department of Health and Social Securities of the UK. Gelatine capsules were used for the oral administration of L1 because of its bitter taste (Figure 1).

In the first two clinical studies, different divided or single daily doses of L1 were administered to 11 patients (10-110 mg/kg/day) to assess efficacy and tolerability. All doses caused a net increase of urinary iron excretion (UIE), which was proportional to the dose of L1 and the iron load of the patients (Figure 2). Doses of 75-110 mg/kg/day were identified to cause negative iron balance with an increase in UIE greater than 25-33 mg, which was equivalent to that caused by DF and higher than the intake of iron from RBC transfusions. No increase in urinary excretion of other essential metals (Ca, Zn and Mg) or other toxic side effects were reported.

International multicentre clinical trials were organised, following the initial clinical trials in London and L1 was supplied in different university clinical centres worldwide e.g. Italy, Switzerland, The Netherlands, Germany, etc. The production of L1 for clinical trials was later carried out by private companies in India, Switzerland, The Netherlands and Canada etc. In 1989 the first episode of agranulocytosis was reported, as well as episodes of neutropenia, masculoskeletal and joint pains, gastric intolerance, and zinc deficiency in the RF, which were also confirmed by other centres. In the same year, no similar agranulocytosis episodes were observed in a total of 125 other patients who received L1 in 8 other countries, as reported in the first international conference on oral chelation (1st ICOC) at the RF. In this context, a mandatory weekly blood count was introduced for prophylaxis against agranulocytosis and neutropenia similar to the drug clozapine.

An application proposing a name for L1 in 1991 by the inventor (GJK), resulted in the adoption by the World Health Organisation (WHO) of the INN name deferiprone (WHO drug information list 67, volume 2 of 1992).

There was no interest from major pharmaceutical companies for the commercial development of L1. No formal animal or other preclinical toxicology studies were carried out on L1 by either Cipla or
Table 1. Deferiprone (L1) - the journey across the years.

1981: Discovery, design, synthesis and physicochemical characterisation of L1.
1981-1982: Iron binding, protein and cell studies in-vitro. Animal studies.
1982: Naming of L1 and other alpha-ketohydroxypyridines.
1983: Patented in the UK. Later patented in the USA, EU and various other countries.
1982-1986: Intensive chemical, biochemical, cell and animal studies.
1986: The UK Department of Health grants permission for clinical trials in the UK.
1987: Simple, cheap synthesis of L1. First-ever clinical trials in London, UK.
1988: Multicentre clinical trials begin worldwide.
1989: First publications on joint/musculoskeletal toxicity and agranulocytosis. Introduction of mandatory weekly white blood cell’s count.
1990: Characterisation of the pharmacokinetic and metabolic properties of L1 in patients and normal volunteers.
1992: Approved BAN and INN name for L1. (INN: Deferiprone).
1994: First ever registration of L1 in India
1995: Clinical use and multicentre clinical studies continue.
1998: It was estimated that more 5000 patients in 35 countries have been using L1, some daily for over 12 years.
1999: Registration of L1 in European, South American and Asian countries.
2000: The new, simple, one-step synthesis of L1 is patented in Greece.
2002: Worldwide interest on L1 following MRI findings regarding effective depletion of iron from the heart, which is the main cause of death in thalassaemia patients.
2003: Proposal for the use of L1 in non-iron loaded conditions including Friedreich Ataxia, Parkinson’s and Alzheimer’s diseases, cancer, HIV etc.
2003 – 2019: Clinical trials with L1 in many non iron loaded conditions
2009: Reduction in morbidity and mortality of thalassaemia patients using L1
2011: Registration of L1 in the USA
2019: Deferiprone (L1) is a leading pharmaceutical in the treatment of thalassaemia, Friedreich’s ataxia and pantothenate kinase-associated neurodegeneration (PKAN).

Apotex. The absence of such data put L1 at a disadvantage as a second line iron chelating drug in comparison to DF and DFRA. However, animal toxicology data are generally of a similar level of toxicity for all three drugs and in clinical practice, L1 is widely used to the same extent as the other two chelating drugs. In many cases, L1 is regarded as the first line iron chelating drug because of its unique properties and especially its cardioprotective effects.5-10

With regards to safety, long term studies, and continuous clinical monitoring involving thousands of thalassaemia and other categories of patients in the last 30 years have confirmed the low toxicity of L1.50,51 The most severe toxic side effects of L1 still remain the same until today and are all controllable, manageable, and reversible. These include reversible agranulocytosis (1% >) and neutropenia (5% >), while less serious toxic side effects include gastric intolerance, musculoskeletal and joint pains and zinc deficiency.50,51 Toxicity vigilance and prophylactic measures are essential steps for ensuring the safety of L1 and the other chelators. For example, zinc supplements are used for prophylaxis for patients on long term treatment with L1, DF and the L1/DF combination.21

Agranulocytosis is the most severe toxic side effect of L1 and mandatory monitoring of weekly white blood cell count is an essential prophylactic measure for its prevention during L1 therapy. Similarly, temporary withdrawal of L1 is necessary during the sore throat and other infections. The cause of agranulocytosis is still unknown but in almost all the cases, this L1 toxicity was transient and all the patients recovered following treatment with granulocyte-colony stimulating factor (G-CSF). The time of recovery of the neutrophil count in patients treated with G-CSF varies between a few days to 7 weeks.21,48 The mechanism of L1 induced agranulocytosis is thought to be related to an L1-related immune response against white cell progenitors since re-challenge on the same patients with L1 results in another episode of agranulocytosis. The patients with this L1 toxic side effect have to switch to DF or DFRA chelation.

Mechanisms of Chelation and Prevention of Iron Toxicity by Deferiprone. The properties and mechanisms of chelation by L1 and other chelating drugs have been previously reviewed.4,21,22 Three molecules of L1 are needed to bind one molecule of iron and to form a red colour iron complex similar to that shown in the urines of iron loaded patients in Figure 2. The small molecular size, neutral charge, and hydrophilicity of L1 allow substantial penetration of almost all tissues including access to all major organs such as the heart and the brain.4,21,22 As a result of the extensive distribution, L1 can act as a universal
Pharmacokinetic profile of deferiprone (L1) and its glucuronide metabolite. Serum monitoring of L1 (white circles) and its glucuronide metabolite (dark circles) following the repeated administration of 3g of L1 in a 68 kg male thalassaemia patient with serum ferritin 2500 μg/L. The timing of oral administration of L1 is shown by arrows. Rapid absorption from the stomach of L1 and elimination from blood in about 6 hours is observed. The glucuronide metabolite of L1 is cleared from blood at about 8 hours. Adapted from reference 21 (with permission).

Iron excretion in response to different doses of deferiprone (L1). The urinary and faecal iron excretion profile of a male iron loaded thalassaemia patient (40kg, serum ferritin 1200 μg/L) treated daily for one week with different doses of L1. The level of iron excretion is related to the dose of L1 and almost all of the chelated iron is excreted in the urine. Antioxidant in all conditions associated with free radical pathology by inhibiting oxidative stress damage caused by excess labile iron catalysis of free radical production.4,21

The pharmacokinetics, metabolism and route of iron elimination of L1 have also been determined (Figure 3).21,52-54 Deferiprone is readily absorbed within minutes from the stomach, metabolised to a glucuronide conjugate, cleared from the plasma within 6-8 hours, and excreted in the urine in the form of L1 iron complex, L1 glucuronide conjugate and free L1 (Figure 3).16,21,52-54 No increase in iron excretion was detected in the faeces of iron loaded thalassaemia patients treated with L1 (Figure 4).53,55

Iron mobilisation by L1 depends on the iron load of the patients and the dose of L1 (Figure 4).53 The increase of UIE in non iron loaded patients is only a few mg, which by comparison, is a small fraction of what is excreted in iron loaded patients.21,53

Iron chelation and mobilisation by L1 have been shown to occur from all the iron pools in cells including ferritin and haemosiderin and also from transferrin and NTBI in plasma (Figure 5).4,21,22,56 In contrast to the other chelating drugs, only L1 can cause the mobilisation of iron from transferrin and prevent the accumulation of excess iron in cells (Figure 5).4,21,22,56

Many factors can affect the rate of accumulation and deposition of iron in the organs of transfused iron loaded patients, with the rate of RBC transfusions being the primary factor (Table 2). Similarly, many factors can affect the rate of iron removal from regularly transfused patients with the most important being the efficacy of the iron chelation protocol (Table 2). In this context, the selected chelating drugs and dose protocols, as well as other related effects, can influence the outcome of chelation therapies (Table 3).57,88

There are many variables in the properties and mode of action of chelating drugs and the selection of any chelation protocol could have a direct effect on the mortality and morbidity of thalassaemia patients (Table 3).5-10,57-60 Optimum iron chelation therapies in the context of personalised medicine in thalassaemia patients take into consideration the most effective and less toxic monotherapy or combination therapy protocols.61 In this context, for each patient, the dose protocols are adjusted with regards to the iron load and the efficacy/tolerability of the chelation therapy.61

The benefits from the selection of a chelation protocol could easily be assessed by monitoring the levels of the iron load and also organ function. For example, the removal of excess toxic iron in thalassaemia patients by L1, and the L1/DF combination is accompanied with improvement of cardiac function, such as elevation of left ventricular ejection fraction (LVEF), endothelial function, etc.9,10,62 Improvements have also been observed in some other haematological conditions using L1 but the mechanisms have not yet been fully clarified.63-65

Recent Developments on Iron Chelation Metabolic Pathways. Congestive cardiac failure due to cardiac iron overload toxicity has been the primary cause of mortality in iron loaded thalassaemia patients for many decades.66,67 Despite that in thalassaemia the diagnostic tests previously used routinely for estimating iron stores such as serum ferritin and liver biopsies could generally reflect body iron stores, neither of these tests could reflect cardiac iron load levels.68-70 Furthermore,
Figure 5. Iron mobilization from the iron pools of iron loaded cell and plasma by deferiprone (L1). The schematic illustration shows the iron loading process of cells and the mode of action of transferrin (Tf) iron deposition via a transferrin receptor (TfR) and non-transferrin bound iron (NTBI). Deferiprone may prevent iron accumulation in cells through iron removal from transferrin in plasma (A) and the low molecular weight plasma iron pool (LMWtFe) or NTBI (B). Deferiprone may also mobilize iron from the intracellular low MWt iron pool (LMWtFe) (C), ferritin (D) and hemosiderin (E). In conditions like Friedreich’s ataxia, deferiprone (L1) can mobilise excess iron from mitochondria (in green).

such information was not sufficient for selecting appropriate chelation therapy protocols for effective removal of excess iron from the heart.

However, the relatively recent routine introduction of new diagnostic techniques such as Magnetic Resonance Imaging (MRI) T2 and T2* which identify the level of iron load in the heart, liver, spleen and other organs of thalassaemia and other iron overloaded patients, has not only increased our understanding of transfusional and other iron overload metabolic pathways but also the differential effect of chelating drugs in iron removal from various organs.60,69-73

The recent diagnostic procedures, and especially MRI T2 and T2* in the determination of iron deposition in organs, have increased the prospects of improved chelating drug targeting therapies of iron overload toxicity, as well as the introduction of personalised chelation regimens in thalassaemia and other iron overload metabolic disorders.73 Furthermore, based on these diagnostic findings the complete treatment of iron overload by removing all excess iron safely from the heart, liver and other organs of regularly TDT patients using L1, the L1/DF or other chelator combinations can nowadays be precisely monitored (Figure 6). In addition, the safe long term maintenance of normal iron stores in thalassaemia patients and prevention of chelating drug toxicity can also be regularly assessed using monthly monitoring of serum ferritin levels, as well as yearly or half yearly MRI T2 and T2* measurements.

The promising results in the treatment of iron overload in thalassaemia encouraged investigations for the use and development of chelating drugs in many other clinical conditions. Such initiatives were within the broad context of the risk/benefit assessment of therapeutic outcomes in each condition because of the absence of other effective therapeutic approaches. Most efforts were mainly focused on the use of L1 as
Table 2. Factors affecting the iron load and iron removal in transfused patients.

| Factors affecting body iron load and distribution |
|--------------------------------------------------|
| • Rate of red blood cell (RBC) transfusions. Splenectomy. |
| • Rate of iron absorption. Rate of iron excretion. |
| • Transferrin iron saturation levels. Non-transferrin bound iron levels. |
| • Cardiac iron levels. Heart is the major target organ of iron toxicity and mortality |
| • The size and function of the liver and spleen as the major iron storage organs. Organ size, vascularity and iron storage capacity in each organ. |
| • Rate of hemolysis of RBC. Red blood cell antibodies. |
| • Haptoglobin function. Antioxidant capacity. |
| • Organ specificity for excess iron uptake and storage eg liver, spleen > bone, brain. |
| • Rate of iron deposition and removal in different organs. |

| Factors affecting total body and individual organ iron removal |
|---------------------------------------------------------------|
| • Iron chelation therapy and influence on the liver/heart iron levels and serum ferritin. |
| • Dose of chelator or chelator combinations and route of administration. |
| • Differential iron removal from the iron pools and organs by the chelating drugs. |
| • Non-uniform organ distribution of stored iron during chelation therapy and identification of intense iron foci. The principle “last in/first out” mobilization of iron deposits by chelators usually apply. |
| • Absorption, distribution, metabolism, excretion and toxicity (ADMET) of chelator, iron complexes and metabolites. Chronotherapy aspects. |
| • Effects of dietary factors, metals other than iron, drugs and nutrients with chelating properties. |
| • Drug interactions. Effects of diuretics and coagulants. The effect of other drugs on iron metabolic and chelation pathways. |
| • Exercise. Sweating. |

| Genetic factors |
|-----------------|
| • Metallomics, proteogenomics, nutrigenomics, pharmacogenomics related to iron and chelating drug metabolism |

| Hormonal function |
|-------------------|
| • Erythropoietin levels. Erythropoietic activity of the bone marrow. Hepcidin levels. |
| • Male/female hormonal activity and secondary events, e.g. iron loss during menstruation and child bearing |

| Disease factors affecting organ iron load and redistribution |
|-------------------------------------------------------------|
| • Anaemia. Hypoxia. Inflammation. Malignancy. Infection. |

| Genetic and acquired conditions affecting organ iron load |
|----------------------------------------------------------|
| • Thalassaemia intermedia. Idiopathic hemochromatosis. Atransferrinaemia. Anaemia of chronic disease. Parkinson’s and Alzheimer’s diseases with brain iron accumulation. Acute iron poisoning. |

Recent developments involving the prospects of the broader use of chelating drugs have been investigated in clinical trials and clinical developments in many of these clinical conditions. In particular, the introduction of L1 for the treatment of non iron loaded patients with focal toxic iron deposits e.g. in Friedreich ataxia and toxic labile iron e.g. in diabetic and non-diabetic glomerular disease, is a reflection of the antioxidant and safety potential of L1. As in many other cases of drug development, the prospects of introduction of L1 and other chelating drugs in these diseases are based on commercial and not ethical criteria.

The Paradigm of the Complete Treatment of Iron Overload in Thalassaemia.

The removal of excess toxic iron accumulated from repeated RBC transfusions in patients with refractory anaemias was the primary aim of all investigations involved with iron removal chelation therapy in the last 50 years. In general, any form of excess iron is potentially toxic because of the ability of iron to catalyse the increased production of free radicals and cascades, which can cause molecular, subcellular, cellular, tissue, and organ damage. The extent of damage can be reversible or irreversible depending mainly on the concentration of excess deposited iron and also other factors.

With the introduction of intramuscular and then subcutaneous and intravenous DF in the early 1960’s, it became evident that the rate of iron removal by DF was not sufficient to compensate for the body iron intake from RBC transfusions in the vast majority of thalassaemia major patients mainly due to severe complications with the parenteral administration of DF. Furthermore, serious complications were also observed such as neurotoxic and other toxic side effects during the use of DF in low iron loaded thalassaemia patients and also other categories of
patients with normal iron stores. 66,74

As a result of the DF limitations, no clear strategies have become available or promoted in the last 50 years for the complete elimination of excess iron and the normalisation of the iron stores in thalassaemia and other patients.

It has been estimated previously that in the absence of chelation therapy the mean survival of thalassaemia major patients was about 20 years, and the primary cause of death was congestive cardiac failure. 5,66,74 Results from a UK registry indicate that with the introduction of DF the mean survival of thalassaemia major patients has increased to about 35 years. 76

Recently, with the introduction of L1, the mortality rate of thalassaemia major patients has decreased substantially and mean survival is approaching that of normal individuals. 5,77

It appears that the primary aim of chelation therapy in thalassaemia major and possibly other chronically transfused patients, i.e. the removal of all excess iron and inhibition/prevention of iron toxicity, as well as the associated tissue and organ damage can now be accomplished in most cases. 8,77 This aim became foreseeable and applicable very recently especially in patients that followed the ICOC protocol using L1 and L1/DF combinations. 8,73,77 Furthermore, the secondary aim of chelation therapy in chronically transfused patients i.e. the safe maintenance of normal iron stores, has also been achieved using lower dose ICOC protocols of L1 monotherapy and L1/DF combinations. 8,73,77

The Achievement and Maintenance of Normal Iron Stores in Thalassaemia. Although the efficacy of chelation monotherapies with DF, L1, and DFRA have been thoroughly studied, no normalisation of the iron stores was reported or investigated in thalassaemia major patients since in the vast majority of patients the

---

Table 3. Comparison of the mode of action and effects of chelating drugs.

| Recommended doses for the chelating drugs in thalassaemia major patients: |
|---|
| DF subcutaneously 40-60 mg/kg/day. Oral L1 75-100 mg/kg/day. Oral DFRA 20-40 mg/kg/day. 78 |

| Optimal chelation therapy protocol for the normalisation of the iron stores in thalassaemia major patients: |
|---|
| The ICOC combination dose protocol of L1 (75-100 mg/kg/day)/DF (40-60 mg/kg/day, 3-7 days per week). 58,73 |

| Compliance of iron loaded patients with chelating drugs: |
|---|
| Low compliance with subcutaneous DF in comparison to oral L1 and oral DFRA. 78 |

| Route of elimination of increased iron excretion in iron loaded patients: |
|---|
| L1: Urinary iron. DFRA: Faecal iron. DF: Mostly urinary but also faecal iron. 53,55,133 |

| Effect of chelating drugs on iron absorption: |
|---|
| Increase of iron absorption by the lipophilic maltol, 8-hydroxyquinoline and DFRA. Decrease of iron absorption by the hydrophilic DF, EDTA, DTPA and L1. 20, 134,136 |

| Iron removal from diferric transferrin and NTBI in iron loaded patients: |
|---|
| Effective transferrin iron removal only by L1 (estimated 40% iron removal from diferric transferrin at L1 concentrations > 0.1 mM), but not by DF or DFRA. 15, 52,36 All three chelating drugs are effective in the removal of non-transferin bound iron. 52,137-139 |

| Differential iron removal from various organs of iron loaded patients: |
|---|
| L1 preferential iron removal from the heart and DFRA from the liver. DF from the liver or heart. (Efficacy is related to the dose of all chelators). 98,103 |

| Efficacy in iron removal from the heart of iron loaded patients: |
|---|
| The ICOC oral L1 / intravenous DF combination > The ICOC oral L1 / subcutaneous DF combination > oral L1> intravenous DF > subcutaneous DF > DFRA. (Efficacy is related to the dose of the chelators). 8,103,140,141 |

| Iron redistribution in diseases of iron metabolism by chelating drugs: |
|---|
| L1 and to a lesser extent DF can cause iron redistribution from the reticuloendothelial system to the erythron in anemic rheumatoid arthritis patients. Enterohemepatic circulation by DFRA and metabolites. 142-144 |

| Increase excretion of metals other than iron e.g. Zn and Al: |
|---|
| Order of increased Zn excretion in iron loaded patients: DTPA> L1> DF>DFRA. 145-149 DF and L1 cause increase Al excretion in renal dialysis patients. DFRA causes an increase Al absorption. 135,150,151 |

| Iron mobilisation and excretion of chelator metabolite iron complexes: |
|---|
| Several DF metabolites have iron chelation potential and increase iron excretion but not the L1 and DFRA glucuronides. 16,52,152,153 |

| Chelating drugs minimising other drug toxicity: |
|---|
| L1 but not DFRA, inhibit doxorubicin induced cardiotoxicity. 92,154 |

| Combination chelation therapy: |
|---|
| L1, DF and DFRA combinations are more effective in iron excretion than monotherapies. Different 1-3 chelating drug combinations are under evaluation. 5,9,58,81,100,155-158 |

| Chelating drug synergism with reducing agents: |
|---|
| Ascorbate act synergistically with DF but not L1 or DFRA for increasing iron excretion. 47,159,160 |

| Chelating drug antioxidant effects: |
|---|
| L1 and DF have shown antioxidant action in in vitro, in vivo and clinical settings. The antioxidant effects of DFRA are under evaluation. Only L1 has been shown to have antioxidant effects in the brain of Friedrich’s ataxia and pantothenate kinase-associated neurodegeneration patients. 11,13,75,95,161,163 |

www.mjhid.org Mediterr J Hematol Infect Dis 2020; 12; e2020011 Pag. 8 / 17
rate of iron removal by chelation was, in general, lower by comparison to the rate of iron intake from RBC transfusions (Table 2).

The normalisation of the iron stores in thalassaemia major and other chronically transfused patients was not considered as a possible option following the introduction of DF and later DFRA, mainly because DFRA and DF were not sufficiently effective in removing all excess iron but also because in both cases there was a high risk of toxicity in non heavily iron loaded patients with serum ferritin lower than 500 μg/L, as described in their drug label information.78

Another limiting factor for not achieving normal iron stores was that no such aim had been proposed in the medical literature until recently or was described in the drug label information.78

Figure 6. Clearance of iron overload from the liver and heart of two thalassaemia patients treated with the deferiprone / deferoxamine (L1 / DF) combination using MRI assessment.

A) MRI changes during the L1 (80-100 mg/kg/day) / DF (40 mg/kg/day 1-3 days per week) combination therapy. Left MR image picture: View of heart (top arrow) and liver (bottom arrow) of a thalassaemia patient before treatment (Cardiac T2* was 14.5 ms and liver T2* 3.7 ms. Serum ferritin was 1626 μg / L). Right picture: 20.5 months after treatment (Cardiac T2* was 20.7 ms and liver T2* 18 ms. Serum ferritin was 186 μg / L). Adapted from reference 81 (with permission).

B) MRI changes during the L1 (75-85 mg/kg/day) / DF (30-60 mg/kg/day, 2-3 days per week) combination therapy. Left MR image: View of liver and heart of a thalassaemia patient before treatment. (Cardiac T2* was estimated as 9.3 ms and liver T2* as 3.8 ms. The serum ferritin was 727 μg/L). Right MR image: 9 months after treatment (Cardiac T2* was 23.0 ms and liver T2* 26.2 ms. The serum ferritin was 166 μg/L). Adapted from reference 44 (with permission).

Individual drug monotherapies are described and recommended by the chelating drug manufacturers in all three chelating drug label information, while chelating drug combinations are not described and are clearly excluded as a form of therapy. The prospect of chelating drug combinations and precisely the L1/DF combination was an academic initiative and suggested as early as 1987 and repeated in 1992.46,79 It was then mainly recommended for patients with toxicity or efficacy complications of either DF or L1.46,79

The dilemma of how to control iron load and over-chelation following the achievement of normal iron
stores has been demonstrated in several studies using the ICOC and similar protocols of tailor-made administration of L1 and L1/DF combinations and by regular monitoring of iron store levels.\textsuperscript{5,87}

The first report of the normalisation of the iron stores in iron loaded thalassaemia major patient was described following the replacement of DF with L1 due to congestive cardiac failure caused by cardiac iron overload during DF therapy.\textsuperscript{86} Several other reports followed, indicating that the use of selected combinations of L1 and DF could achieve the normalisation of the iron stores in iron loaded thalassaemia major patients.\textsuperscript{81-83} In particular, the ICOC protocol of L1 (80–100 mg/kg/day) and subcutaneous DF (40–60 mg/kg/day, at least 3 days per week) was identified as the most tolerable and effective chelation therapy protocol for achieving negative iron balance (Figure 6, Table 3).\textsuperscript{33,81}

Continuous monitoring of the iron stores, e.g. monthly serum ferritin assessment, is required for regularly transfused patients who have achieved normal iron stores.\textsuperscript{83} Furthermore, continuous adjustment of iron chelation dose protocols is necessary for maintaining the normal iron stores without the prospect of excess chelation toxicity.\textsuperscript{83} Different dose protocols of L1, DF, and L1/DF combination, are required for maintaining normal iron stores within the context of personalised medicine.\textsuperscript{83} In some cases of low serum ferritin in thalassaemia major patients, withdrawal of chelation therapy may be necessary for avoiding iron deficiency.\textsuperscript{83,84}

In addition to the more significant clinical benefits for thalassaemia patients from the maintenance of normal iron stores, there is also a substantial reduction in the cost of chelation therapy since much lower doses of chelators are generally used by comparison to iron loaded patients.\textsuperscript{58,83}

**Future Prospects of Iron Chelation Therapy.** It is conceivable that the aim of iron chelation therapy in transfusional iron overload for achieving and maintaining normal iron stores will be accomplished in many more patients in the forthcoming years, thus decreasing associated morbidity and mortality due to excess iron toxicity. Already in countries like Cyprus, many thalassaemia patients are achieving life spans approaching that of the general population, are active professionals in society and have families with children and even grandchildren.\textsuperscript{5,77}

The same aim and approach for the normalisation of the iron stores and the reduction of excess iron toxicity in thalassaemia major could be used in many other haematological conditions of iron overload including myelodysplasia, post-allogenic stem cell transplantation, non-transfusion dependent thalassaemia (NTDT), non-venesected idiopathic haemochromatosis, transfused cancer cases etc.\textsuperscript{78} Effective iron chelation therapy protocols within the context of personalised medicine and risk/benefit assessment could be used in each of these cases, similar to the ICOC protocol.\textsuperscript{61} In most of these cases, tolerant and active combination protocols of 1-3 chelating drugs may be used for effective and rapid clearance of excess iron.\textsuperscript{81-83}

The interaction between chelating drugs and chelating drugs with other drugs used for other therapeutic effects of the underlying diseases needs further investigations. Similarly, the therapeutic and toxic effects of drugs with chelating potential such as hydroxycarbamate (hydroxyurea) and iron also need further investigation.\textsuperscript{85}

The clinical application of iron chelating drugs and other chelators is likely to increase in the future involving the treatment of many other diseases in addition to transfusional iron overload and focal iron deposit toxicities.\textsuperscript{11-13,74,86} Initial clinical trials in several non iron loaded diseases with L1 are encouraging and promising.\textsuperscript{87-90} Most of these future applications include infectious diseases by withholding iron from microbes, intervention in iron metabolic pathways associated with cancer, HIV and other diseases, detoxification of environmental and diagnostic metals, and inhibition of excess toxic free radical production involved in many diseases of free radical pathology.\textsuperscript{74,75,87-91} In particular, with regards to the latter, iron chelation therapy using L1 has been considered for the reduction of anticancer drug toxicity such as doxorubicin, for ophthalmic toxicity and neurotoxicity and also many other related applications.\textsuperscript{92-95}

The selection of therapeutic protocols for thalassaemia and other diseases involving chelating drugs is crucial because it affects risk/benefit assessment and therapeutic outcome, as well as morbidity and mortality of hundreds of thousands of patients.\textsuperscript{96-99} The present state of generally ‘random’ selection of chelating drug protocols does not appear to benefit the patients. In this context, the high efficacy and safety of the ICOC L1/DF combination protocol should be considered as a first line chelation treatment for the vast majority of thalassaemia patients.\textsuperscript{8,81,83} This proposition is supported by recent detailed monitoring findings in the improvement in cardiac iron depletion rate and cardiac function by L1 and L1/DF over other therapies.\textsuperscript{60,100} Advances in the constant monitoring of iron deposition in critical organs like heart, liver, and pancreas by MRI T2* has recently allowed improvement in the tailoring iron chelation therapy and the selection of the more appropriate chelation regimens in different clinical cases, thus reducing overall patient mortality and morbidity.\textsuperscript{101-103}

The limitations in the use of L1 and the L1/DF combination in some countries may constitute an irregular action by health policy decision makers and also negligence in relation to the well being of
thalassaemia patients. This policy appears to be controversial, especially considering that drug combinations are widely used not only in other haematological conditions but also in many other diseases.

Similar controversies apply in the risk/benefit assessment for the use of chelating drugs not only in transfusion-dependent thalassaemia (TDT) but in patients with non-transfusion-dependent thalassaemia intermedia, idiopathic haemochromatosis, myelodysplasia, sickle cell disease, post-transplanted sickle cell disease and thalassaemia as well as many other categories of patients.117

With regards to personalised medicine, the characterisation of the iron metabolic or toxicity or other related targets is necessary for designing the appropriate therapeutic strategies in each condition and each patient, which can result in the optimisation of chelating drug protocol or other therapeutic interventions.117,118 In this context, the mechanisms of iron release from ferritin and haemosiderin, as well as other molecular or cellular mechanisms are of particular interest.118,119

Changes in the therapeutic strategies are necessary under special circumstances such as pregnancy, splenomegaly, and infections and also when considering the possible introduction of erythropoietic biological or other emerging therapies.120-123 Similar considerations are in progress regarding other clinical issues such as the early initiation of chelation therapy using L1 in thalassaemia children from about one year of age and also the initiation of combination therapies.124-127 There are different criteria and opinions regarding the latter, with the ICOC L1/DF combination, for example, to be available, safe and flexible in all the patient categories and cases depending on the iron load levels and the rate of body iron intake from transfusions, whereas for other groups of investigators different restrictions are implied in the use of combination protocols (Figure 6).

The academic debates on the efficacy, toxicity, historical, and other aspects of L1, DF and DFRA and their combinations are likely to continue in the forthcoming years. Such debates are mostly focused on past practises of ineffective therapies and not issues associated with the current “golden era” period of iron chelation therapy in thalassaemia, namely the achievement and maintenance of normal iron stores.81-83,128-131

The molecular, therapeutic, and other properties of L1 as a potent chelator and antioxidant with access to most tissues and organs make it a unique pharmaceutical with broad spectrum clinical applications.75,95 This prospect/dilemma is similar to that of the introduction of L1 as the first oral iron chelating drug about 30 years ago and needs further investigations to be confirmed.132 Within this context, specific therapeutic strategies have to be designed based on a risk/benefit assessment for each condition and each patient. The aim and targets of such therapeutic strategies need to be defined and evaluated in a manner similar to the case of the paradigm of the complete treatment of iron overload in thalassaemia using L1 and selected L1/DF combinations.58,81-83

References:

1. Community control of hereditary anaemias: memorandum from a WHO meeting. Bull World Health Organ. 1983; 61: 63-80.
2. Weatherall DJ, Clegg JB. Inherited haemoglobin disorders: an increasing global health problem. Bull World Health Organ. 2001; 79: 704-12.
3. Verma IC. Burden of genetic disorders in India. Indian J Pediatr. 2000; 67: 893-8.
4. Kontogiorgos GJ, Eracleous E, Economides C, Kolnagou A. Advances in iron overload therapies. Prospects for effective use of deferiprone (L1), deferoxamine, the new experimental chelators ICL670, GT56-252, L1NA11 and their combinations. Curr Med Chem. 2005; 12:2663-81.
5. Telfer PT, Warburton F, Christou S, Hadjigavriel M, Sitarou M, Kolnagou A, Angastiniotis M. Improved survival in thalassaemia major patients on switching from deferoxamine to combined chelation therapy with deferoxamine and deferiprone. Haematologica. 2009; 94: 1777-8.
6. Au WY, Lee V, Lau CW, Yau J, Chan D, Chan EY, Cheung WW, Ha SY, Kho B, Lee CY, Li RC, Li CK, Lin SY, Ling AS, Mak V, Sun L, Wong KH, Wong R, Yuen HL. A synopsis of current care of thalassaemia major patients in Hong Kong. Hong Kong Med J 2011; 17: 261-6.
7. Maggio A, Filosa A, Vitrano A, Aloj G, Gattamis A, Ceci A, Fucharoen S, Cianciulli P, Grady RW, Prossomariti L, Porter JB, Iacono A, Cappellini MD, Bonifizzi F, Cassarà F, Hamezt P, Wood J, Glud C. Iron chelation therapy in thalassemia major: a systematic review with meta-analyses of 1520 patients included on randomized clinical trials. Blood Cells Mol Dis 2011; 47:166-75.
8. Kontogiorgos GJ. The aim of iron chelation therapy in thalassaemia. Eur J Haematol. 2017;99:465-66.
9. Telfer PT, Warburton F, Christou S, Hadjigavriel M, Sitarou M, Kolnagou A, Angastiniotis M. Improved survival in thalassaemia major patients on switching from deferoxamine to combined chelation therapy with deferoxamine and deferiprone. Haematologica. 2009; 94: 1777-8.
10. Au WY, Lee V, Lau CW, Yau J, Chan D, Chan EY, Cheung WW, Ha SY, Kho B, Lee CY, Li RC, Li CK, Lin SY, Ling AS, Mak V, Sun L, Wong KH, Wong R, Yuen HL. A synopsis of current care of thalassaemia major patients in Hong Kong. Hong Kong Med J 2011; 17: 261-6.
11. Maggio A, Filosa A, Vitrano A, Aloj G, Gattamis A, Ceci A, Fucharoen S, Cianciulli P, Grady RW, Prossomariti L, Porter JB, Iacono A, Cappellini MD, Bonifizzi F, Cassarà F, Hamezt P, Wood J, Glud C. Iron chelation therapy in thalassemia major: a systematic review with meta-analyses of 1520 patients included on randomized clinical trials. Blood Cells Mol Dis 2011; 47:166-75. https://doi.org/10.1016/j.bcmd.2011.07.002
PMID:21843958
12. Kontogiorgos GJ. The aim of iron chelation therapy in thalassaemia. Eur J Haematol. 2012;87:732-3.
13. Telfer PT, Warburton F, Christou S, Hadjigavriel M, Sitarou M, Kolnagou A, Angastiniotis M. Improved survival in thalassaemia major patients on switching from deferoxamine to combined chelation therapy with deferoxamine and deferiprone. Haematologica. 2009; 94: 1777-8.
14. Au WY, Lee V, Lau CW, Yau J, Chan D, Chan EY, Cheung WW, Ha SY, Kho B, Lee CY, Li RC, Li CK, Lin SY, Ling AS, Mak V, Sun L, Wong KH, Wong R, Yuen HL. A synopsis of current care of thalassaemia major patients in Hong Kong. Hong Kong Med J 2011; 17: 261-6.
15. Maggio A, Filosa A, Vitrano A, Aloj G, Gattamis A, Ceci A, Fucharoen S, Cianciulli P, Grady RW, Prossomariti L, Porter JB, Iacono A, Cappellini MD, Bonifizzi F, Cassarà F, Hamezt P, Wood J, Glud C. Iron chelation therapy in thalassemia major: a systematic review with meta-analyses of 1520 patients included on randomized clinical trials. Blood Cells Mol Dis 2011; 47:166-75. https://doi.org/10.1016/j.bcmd.2011.07.002
PMID:21843958
16. Kontogiorgos GJ. The aim of iron chelation therapy in thalassaemia. Eur J Haematol. 2017;99:465-66.
17. Telfer PT, Warburton F, Christou S, Hadjigavriel M, Sitarou M, Kolnagou A, Angastiniotis M. Improved survival in thalassaemia major patients on switching from deferoxamine to combined chelation therapy with deferoxamine and deferiprone. Haematologica. 2009; 94: 1777-8.
18. Au WY, Lee V, Lau CW, Yau J, Chan D, Chan EY, Cheung WW, Ha SY, Kho B, Lee CY, Li RC, Li CK, Lin SY, Ling AS, Mak V, Sun L, Wong KH, Wong R, Yuen HL. A synopsis of current care of thalassaemia major patients in Hong Kong. Hong Kong Med J 2011; 17: 261-6.
19. Maggio A, Filosa A, Vitrano A, Aloj G, Gattamis A, Ceci A, Fucharoen S, Cianciulli P, Grady RW, Prossomariti L, Porter JB, Iacono A, Cappellini MD, Bonifizzi F, Cassarà F, Hamezt P, Wood J, Glud C. Iron chelation therapy in thalassemia major: a systematic review with meta-analyses of 1520 patients included on randomized clinical trials. Blood Cells Mol Dis 2011; 47:166-75. https://doi.org/10.1016/j.bcmd.2011.07.002
PMID:21843958
117. Kontogiorgos GJ. The aim of iron chelation therapy in thalassaemia. Eur J Haematol. 2012;87:732-3.
118. Telfer PT, Warburton F, Christou S, Hadjigavriel M, Sitarou M, Kolnagou A, Angastiniotis M. Improved survival in thalassaemia major patients on switching from deferoxamine to combined chelation therapy with deferoxamine and deferiprone. Haematologica. 2009; 94: 1777-8.
119. Au WY, Lee V, Lau CW, Yau J, Chan D, Chan EY, Cheung WW, Ha SY, Kho B, Lee CY, Li RC, Li CK, Lin SY, Ling AS, Mak V, Sun L, Wong KH, Wong R, Yuen HL. A synopsis of current care of thalassaemia major patients in Hong Kong. Hong Kong Med J 2011; 17: 261-6.
12. Abbuzzese G, Cossu G, Balocco M, Marchese R, Murgia D, Melis M, Galanerlo R, Barella S, Matta G, Ruffinengo U, Bonuccelli U, Forni GL. A pilot trial of deferiprone for neurodegeneration with brain iron accumulation. Haematologica. 2011; 96: 1708-11. https://doi.org/10.3324/haematol.2011.040318
PMID:21791473 PMCID:PMC3208690

13. Cossu G, Abbuzzese G, Matta G, Murgia D, Melis M, Ricci V, Galanerlo R, Barella S, Origa R, Balocco M, Pelosi E, Marchese R, Ruffinengo U, Forni GL. Efficacy and safety of deferiprone for the treatment of pantothenate kinase-associated neurodegeneration (PKAN) and neurodegeneration with brain iron accumulation (NBIA): result from a four years follow-up. Parkinsonism Relat Disord. 2014; 20: 651-4. https://doi.org/10.1016/j.parkreldis.2014.03.002
PMID:24661465

14. Kontoghiorghes GJ, Wilson MT. Structural and kinetic studies on esiema festida erthyrocruorin. In: J. Lamy ed. Invertebrate Oxygen-Binding Proteins, Structure, Active Site and Function. Marcel Dekker, New York and Basel.1981, 385-391.

15. Kontoghiorghes GJ. The design of orally active iron chelators for the treatment of thalassaemia. PhD thesis, Colchester UK, University of Essex, British Library Microfilm No D66194/86. 1982, pp 1 -243. (https://www.pri.ac.cy/files/KGJ_thesis_1982.pdf

16. Kontoghiorghes GJ, Marcus RE, Huehns ER. Desferrioxamine L1 and other α-keto-hydroxypyridinines in the treatment of transfusional iron overload in thalassemia. Ann N Y Acad Sci. 1990; 612: 339-50. https://doi.org/10.1111/j.1749-6632.1990.tb24321.x
PMID:22911562

17. Kontoghiorghes GJ, Keast CG. A simple metabolic cage for mice. Laboratory practice. 1984;33: 90-1.

18. Kontoghiorghes GJ, Marcus RE, Huehns ER. Desferrioxamine suppositories. Lancet. 1983; ii: 454. https://doi.org/10.1016/S0140-6736(83)90413-0
PMID:6893983

19. Kontoghiorghes GJ. New orally active iron chelators. Lancet. 1985; i: 817

20. Kontoghiorghes GJ, Barr J, Nortey P, Sheppard L. Selection of a new generation of orally active alpha-keto-hydroxypyridine iron chelators intended for use in the treatment of iron overload. Am J Hematol. 1993;42:340-9. https://doi.org/10.1002/ajh.2830420403
PMID:8493983

21. Kontoghiorghes GJ (ed). Oral chelation in the treatment of thalassaemia and other diseases. Drugs Today. 1992; 28 (Suppl A):1-187.

22. Kontoghiorghes GJ, Pattichis K, Neocoulous K, Kolnagou A. The design and development of deferiprone (L1) and other iron chelators for clinical use: targeting methods and application prospects. Curr Med Chem. 2004; 11: 2161-83. https://doi.org/10.2174/09286704364685
PMID:15279556

23. Kontoghiorghes GJ, Sheppard L. Simple synthesis of the potent iron chelators 1-allyl-3-hydroxy-2-methylpyridine-4-ones. Inorg Chim Acta. 1987;136: L11-L12. https://doi.org/10.1016/S0020-1693(00)85549-8
PMID:3489383

24. Pepe A, Rossi G, Bentley A, Putti MC, Frizzi L, D'Ascola DG, Cuccia L, Spasiano A, Filosa A, Caruso V, Hanif A, Saez V, Hider RC. Iron mobilization from hepatocyte monolayer cultures by chelators: the importance of membrane permeability and the iron-binding constant. Blood. 1988;72:1497-503. https://doi.org/10.1182/blood.V72.5.1497.1497
PMID:3179437

25. Hider RC, Singh S, Porter JB, Huehns ER. The development of hydroxypyridin-4-ones as orally active iron chelators. Ann N Y Acad Sci. 1990;612:327-38. https://doi.org/10.1111/j.1749-6632.1990.tb24320.x
PMID:22911561

26. Porter JB, Morgan J, Hayes KP, Burke LC, Huehns ER, Hider RC. Relative oral efficacy and acute toxicity of hydroxypyridin-4-one iron chelators in mice. Blood. 1990;76:2389-96. https://doi.org/10.1182/blood.V76.11.2389.2389
PMID:2257308

27. Porter JB, Hoyes KP, Abeyesinghe R, Huehns ER, Hider RC. Animal toxicology of iron chelator L1. Lancet. 1989;2(8655):156. https://doi.org/10.1016/S0140-6736(89)90206-7
PMID:2853385

28. Porter JB, Abeyesinghe RD, Hayes KP, Barra C, Huehns ER, Brooks PN, Blackwell MP, Aranuta M, Brittenham G, Singh S, Dobbin P, Hider RC. Contrasting interspecies efficacy and toxicity of 1,2-diethyl-3-hydroxypyridin-4-one, CP94, relates to differing metabolism of the iron chelating site. Br J Haematol. 1993;85:159-68. https://doi.org/10.1111/j.1365-2457.1993.tb08066.x
PMID:8251385

29. Epemolu RO, Ackerman R, Porter JB, Hider RC, Damani LA, Singh S. HPLC determination of 1,2-diethyl-3-hydroxypyridin-4-one (CP94), its iron complex [Fe(III)] (CP943) and glucuronide conjugate [CP94-GLUC] in serum and urine of thalassaemic patients. J Pharm Biomed Anal. 1994;356:361-70. https://doi.org/10.1016/0140-6736(94)90718-7
PMID:7887242

30. Pfannkuch F, Bentley P, Schnebel HP. Future of oral iron chelator deferiprone (L1) Lancet. 1993;341(8855):1480. https://doi.org/10.1016/0140-6736(93)90295-7

31. Hehrko C. Development of oral iron chelator L1. Lancet. 1993;341(8852):1088-9. https://doi.org/10.1016/0140-6736(93)90244-X
PMID:8421279

32. Savulescu J. Thalassaemia major: the murky story of deferiprone. Br Med J 2004;328(7436):358-9. https://doi.org/10.1136/hmj.2004.025332
PMID:14962851 PMCID:PMC341373

33. REUTERS [webpage on the Internet]. U.S. seeks up to $3.35 billion in Novartis kickback lawsuit; 2015. Available from: http://www.reuters.com/article/2015/06/30/us-novartis-lawsuit-idUSKCN0-PA1ZK20150630. Accessed October 1, 2019.

34. Kontoghiorghes CN, Kontoghiorghes GJ. New developments and controversies in iron metabolism and iron chelation therapy. World J Methodol. 2016;6:1-19. https://doi.org/10.3968/wjm.v6i1.1
PMID:27019793 PMCID:PMC4804243
in thalassaemia major patients treated with deferasirox versus deferiprone and desferrioxamine. Br J Haematol. 2018;173:783-95. 
https://doi.org/10.1111/bjh.15595
PMMid:30334574

65. Kontoghiorghes GJ. A new era in iron chelation therapy: the design of optimal, individually adjusted iron chelation therapies for the complete removal of iron overload in thalassaemia and other chronically transfused patients. Hemoglobin. 2009; 33:332-8. 
https://doi.org/10.1080/03630260903217182
PMMid:19814679

66. Fisola A, Vitran A, Rignano P, Calvaruso G, Barone R, Capra M, Cuccia L, Gagliardotto F, Pitrolo L, Prossomarti L, Casale M, Caruso V, Gerardì C, Campisi S, Cianciulli P, Rizzo M, D'Ascola G, Ciancio A, Maggio A. Long-term treatment with deferoxiprone enhances left ventricular ejection function when compared to deferoxamine in patients with thalassaemia major. Blood Cells Mol Dis. 2013;51:85-8. 
https://doi.org/10.1016/j.bcmd.2013.04.002
PMMid:23628348

67. Smeets ME, Vreugdenhil G, Holdrenit RS. Improvement of erythropoiesis during treatment with deferoxiprone in a patient with myelofibrosis and transfusional hemosiderosis. Am J Hematol. 1996;51:243-4. 
https://doi.org/10.1002/(SICI)1096-8652(199603)51:3<243::AID- AJH12>3.0.CO;2-H

68. Chang YH, Shaw CF, Wu KH, Hsieh KH, Su YN, Lu PJ. Treatment with deferoxiprone for iron overload alleviates bone marrow failure in a Fanconi anemia patient. Hemoglobin. 2009;33:346-51. 
https://doi.org/10.1080/03630260903212563
PMMid:19814681

69. Srivastana T, Vivithanaporn P, Paiboonsukwong K, Rattanawonsakul K, Srijurin S, Sihmnooh N. Deferoxipine increases endothelial nitric oxide synthase phosphorylation and nitric oxide production. Can J Physiol Pharmacol. 2018;96:879-85. 
https://doi.org/10.1139/cjpp-2018-0012
PMMid:29806986

70. Zarno MG, De Stefano P, Borgna-Pignatti C, Di Palm A, Piga A, Melevendi C, Di Gregorio F, Burattini MG, Terzoli S. Survival and causes of death in thalassaemia major. Lancet. 1989;2: 27-30. 
https://doi.org/10.1016/S0140-6736(89)90064-X

71. Kyrracou K, Michaelides Y, Senkus R, Simmanon K, Pavlides N, Antoniades L, Zambartas C. Ultrasonic pathology of the heart in patients with beta-thalassaemia major. Ultrasonic Pathol. 2000; 24: 75-81. 
https://doi.org/10.1016/S0191-312X(00)00018-3
PMMid:10808552

72. Kolnagou A, Economides C, Ereucleos E, Kontoghiorghes GJ. Low serum ferritin levels are misleading for detecting cardiac iron overload and increase the risk of cardiomyopathy in thalassaemia patients. The importance of cardiac iron overload monitoring using magnetic resonance imaging T2 and T2*. Hemoglobin. 2006;30:219-27. 
https://doi.org/10.1080/0363026060642542
PMMid:16798647

73. Papakonstantinou O, Alexoupoulou E, Economopoulos N, Benekos O, Kattamis A, Kostaridou S, Ladis V, Efthastopoulos E, Gouliamos A, Kelekis NL. Assessment of iron distribution between liver, spleen, pancreas, bone marrow, and myocardium by means of R2 relaxometry with MRI in patients with beta-thalassaemia major. J Magn Reson Imaging. 2009;29:583-9. 
https://doi.org/10.1002/mri.21707
PMMid:19306409

74. Kolnagou A, Natsiopoulos K, Kleanthous M, Ioannou A, Kontoghiorghes GJ. Liver iron and serum ferritin levels are misleading for estimating cardiac, pancreatic, splenic and total body iron load in thalassaemia patients: factors influencing the heterogenic distribution of excess storage iron in organs as identified by MRI T2*. Toxicol Mech Methods. 2013; 23: 48-56. 
https://doi.org/10.1515/tmm.2013.076
PMMid:22943064

75. Mavrogeni SI, Gotsis ED, Markussis V, Tsekos N, Politis C, Vretou E, Kermastinos D. T2 relaxation time study of iron overload in b-thalassaemia. MAGMA. 1998;6:7-12. 
https://doi.org/10.1002/(SICI)1096-8652(199603)1:3<243::AID-AJH12>3.0.CO;2-H

76. Anderson LJ, Holden S, Davis B, Prescott E, Charrier CC, Bunce NH, Firmin DN, Wonke B, Porter J, Walker JM, Pennell DJ. Cardiovascular T2-star (T2*) magnetic resonance for the early diagnosis of myocardial iron overload. Eur Heart J. 2001;22: 2171-9. 
https://doi.org/10.1053/euhj.2001.2822
73. Kolnagou A, Yazman D, Economides C, Enacleous E, Kontoghiorgis GJ. Uses and limitations of serum ferritin, magnetic resonance imaging T2 and T2* in the diagnosis of iron overload and in the ferritin kinetics of normalization of the iron stores in thalassemia using the International Committee on Chelation deferiprone/deferoxamine combination protocol. Hemoglobin. 2009;33:312-22. [https://doi.org/10.3109/03630260903213231]

PMid:19814677

74. Kontoghiorgis GJ, Neicleous K, Kolnagou A. Benefits and risks of deferiprone in iron overload in thalassemia and other conditions: comparison of epidemiological and therapeutic aspects with deferoxamine. Drug Safety. 2003;26:553-84. [https://doi.org/10.2165/00002018-20032608-00003]

PMid:12825969

75. Kontoghiorgis GJ, Kontoghiorghe CN. Prospects for the introduction of targeted antioxidant drugs for the prevention and treatment of diseases related to free radical pathology. Expert Opin Investig Drugs. 2019;28: 593-603. [https://doi.org/10.1080/13543784.2019.1631284]

PMid:31185180

76. Modell B, Khan M, Darlison M. Survival in beta-thalassemia major in the UK: data from the UK Thalassaemia Register. Lancet. 2000; 355: 2051-2. [https://doi.org/10.1016/S0140-6736(00)02357-6]

77. Kolnagou A, Kontoghiorghe CN, Kontoghiorgis GJ. Transition of thalassaemia and Friedreich ataxia from fatal to chronic diseases. World J Methodol. 2014;4:197-218. [https://doi.org/10.5662/wjm.v4.i4.197]

PMid:25541601 PMCID:PMC4724580

78. Kontoghiorgis GJ, Kontoghiorgis CN, Kontoghiorgis GJ. Efficacy and safety of iron-chelation treatment with deferiprone, deferoxamine, and deferasirox for the treatment of iron-loaded patients with non-transfusion-dependent thalassemia syndromes. Drug Des Devel Ther. 2016;10: 465-81. [https://doi.org/10.2147/DDDT.S79458]

PMid:28893541 PMCID:PMC4745840

79. Kontoghiorgis GJ. Advances in oral iron chelation in man. Int J Hematol. 1992;55: 27-38.

80. Kolnagou A, Michaelides Y, Kontos C, Kyriacou K, Kontoghiorgis GJ. Myocyte damage and loss of myofibers is the potential mechanism of iron overload toxicity in congestive cardiac failure in thalassemia. Br J Haematol. 1992;82:27-38. [https://doi.org/10.1111/j.1365-2141.1992.tb09700.x]

PMid:19912219

81. Kolnagou A, Kontoghiorgis CN, Kontoghiorgis GJ. Prevention of Iron Overload and Long Term Maintenance of Normal Iron Stores in Thalassaemia Major Patients using Deferiprone or Deferiprone Deferoxamine Combination. Drug Res (Stuttg). 2017; 67: 404-11. [https://doi.org/10.1055/s-0043-102691]

PMid:28320041

82. Forni GL, Gamberini MR, Ghilardi R, Piga A, Romeo MA, Zhao H, Cnaan A. Cardiac morbidity and mortality in deferoxamine- or deferasirox-treated patients with thalassemia major. Br J Haematol. 2013;163:400-3. [https://doi.org/10.1111/bjh.12517]

PMid:24033185

83. Konstantinou E, Pashalidis I, Kolnagou A, Kontoghiorghes GJ. Antioxidant targeting by deferiprone in diseases related to oxidative damage. Front Biosci (Landmark Ed). 2014;19: 862-85. [https://doi.org/10.2741/4253]

PMid:24896322

84. Origa R, Danjou F, Cossa S, Matta G, Bina P, Dessi C, Defraia E, Foschini ML, Leoni G, Morittu M, Galanello R. Impact of cardiac magnetic resonance imaging on chelation choices, compliance with treatment and risk of heart disease in patients with thalassaemia major. Br J Haematol. 2013;163:400-3. [https://doi.org/10.1111/bjh.12517]

PMid:24033185

85. Platis O, Anagnostopoulos G, Farmaki K, Posantzi M, Gottis E, Toli G. Glucose metabolism disorders improvement in patients with thalassaemia major after 24-36 months of intensive chelation therapy. Pediatr Endocrinol Rev. 2004; 2 Suppl 2: 279-81.

86. 98] Borgna-Pignatti C, Cappellini MD, Del Stefano P, Del Vecchio GC, Forini GL, Gamberini MR, Ghihardi R, Piga A, Romeo MA, Zhao H, Cnaan A. Cardiac morbidity and mortality in deferoxamine- or deferasirox-treated patients with thalassemia major. Blood. 2006; 107: 1733-7. [https://doi.org/10.1182/blood-2005-07-2933]

PMid:16373663

87. Tanner MA, Galanello R, Dessi C, Smith GC, Westwood MA, Agus A, Pitiri M, Nair SV, Walker JM, Pennell DJ. Combined chelation therapy in thalassemia major for the treatment of severe myocardial siderosis with left ventricular dysfunction. J Cardiovasc Magn Reson. 2008; 10: 12. [https://doi.org/10.1182/1532-429X-10.12]

PMid:18298856 PMCID:PMC2289829

88. Pepe A, Meloni A, Rossi G, Cuccia L, D’Ascola GD, Suntodreroz M, Cianciulli P, Caruso V, Romeo MA, Filosa A, Pirolo L, Putti MC, Pulello A, Campisi S, Missere M, Midiri M, Gulino L, Positano V, Lombardi M, Ricchi P. Cardiac and hepatic iron and ejection fraction in thalassemia major: multicenter prospective comparison of combined
deferiprone and deferoxamine therapy against deferiprone or deferoxamine monotherapy. J Cardiovasc Magn Reson. 2013;15:1. https://doi.org/10.1186/1532-429X-15-1

PMid:23324167 PMCid:PMC3599638

101. Pepe A, Meloni A, Capra M, Gianferrari M, Misdra M, Muscato M, Valeri G, Sorrentino F, D'Androsa DG, Spasiano A, Filosa A, Cuccia L, Dalle Iacono N, Forini G, Caruso V, Maggio A, Pitrillo P, Pesotia A, De Marchi D, Postiano V, Wood JC. Prediction of cardiac complications for thalassemia major in the widespread cardiac magnetic resonance era: a prospective multicentre study by a multi-parametric approach. Eur Heart J Cardiovasc Imaging. 2018;19:299-309. https://doi.org/10.1093/ehjci/ejx012

PMid:28200076

102. Pepe A, Meloni A, Capra M, Gianferrari M, Misdra M, Muscato M, Valeri G, Sorrentino F, D'Androsa DG, Spasiano A, Filosa A, Cuccia L, Dalle Iacono N, Forini G, Caruso V, Maggio A, Pitrillo P, Pesotia A, De Marchi D, Postiano V, Lombardi M, Maggio A. Deferasirox, deferiprone and desferrioxamine treatment in thalassemia major patients: cardiac iron and function comparison determined by quantitative magnetic resonance imaging. Haematologica. 2011;96:41-7. https://doi.org/10.3324/haematol.2009.019042

PMid:20884710 PMCid:PMC3001276

103. Pennell DJ, Udesho JE, Arai AE, Bozkurt B, Cohen AR, Galanello R, Hoffman TM, Kieman MS, Lerakis S, Piga A, Porter JB, Walker JM, Wood J; Cardiovascular function and treatment in β-thalassemia major: a consensus statement from the American Heart Association. Circulation. 2013;128: 281-308. https://doi.org/10.1161/CIRCULATIONAHA.111.161897

PMid:23775258

104. Steensma DP. Myelodysplasia parainao: iron as the new radon. Leuk Res. 2009;33:1158-63. https://doi.org/10.1016/j.leukres.2008.10.017

PMid:19036443

105. Lucania G, Vitro A, Filosa A, Maggio A. Chelation treatment in sickle-cell-anemia: much ado about nothing? Br J Haematol. 2011;154: 545-55. https://doi.org/10.1111/j.1365-2141.2011.08769.x

PMid:21707578

106. Marsella M, Borgna-Pignatti C. Transfusional iron overload and iron chelation therapy in thalassemia major and sickle cell disease. Hematol Oncol Clin North Am. 2014;28:703-27. https://doi.org/10.1016/j.hoc.2014.04.004

PMid:25604709

107. Lucarelli G, Angelucci E, Giardini C, Bariconiani D, Galimberti M, Polchi P, Bartolucci M, Muretto P, Albertini F. Fate of iron stores in thalassemia after bone-marrow transplantation. Lancet. 1993;342(8844):1388-91. https://doi.org/10.1016/0140-6736(93)90275-G

PMid:1060140-673609302753-G

108. Pol F, Angelucci E. Iron Toxicity and Hemopoietic Cell Transplantation: Time to Change the Paradigm. Mediterr J Hematol Infect Dis. 2019;11:e2019019. https://doi.org/10.4084/mjhid.2019.019

PMid:30858957 PMCid:PMC6402552

109. Kolagou A, Kontogiorghe CN, Kontogiorghe GJ. New targeted therapies and diagnostic methods for iron overload diseases. Front Biosci (Schol Ed). 2018;10:1-20. https://doi.org/10.2741/s498

PMid:28930516

110. La A, Nguyen T, Tran K, Sauble E, Tu D, Gonzalez A, Kidane TZ, Nagori C, Morgan J, Doan M, Tran K, Wang CY, Koutson MD, Linder MC. Mobilization of iron from ferritin: new steps and details. Metallomics. 2018;10:154-68. https://doi.org/10.1039/C7MT002841J

PMid:29260183

111. Kolagou A, Kontogiorghe CN, Kontogiorghe GJ. Iron mobilization from ferritin using alpha-oxohydroxy heteroaromatic chelators. Biochem J. 1986;233:299-302. https://doi.org/10.1042/bj3332029

PMid:3954731 PMCid:PMC1153022

112. Origa R, Comiti F. Pregnancy in Thalassemia. Mediterr J Hematol Infect Dis. 2019;11:e2019019. https://doi.org/10.4084/mjhid.2019.019

PMid:30858957 PMCid:PMC6402552

113. Kolagou A, Kontogiorghe CN, Kontogiorghe GJ. Transfusion-Related Acute Lung Injury (TRALI) in two Thalassaemia Patients Caused by the Same Multiparous Blood Donor. Mediterr J Hematol Infect Dis. 2017;9:2017060. https://doi.org/10.4084/mjhid.2017.060

PMid:29198137 PMCid:PMC5667526

114. Kolagou A, Michaelides Y, Kontogiorghe CN, Kontogiorghe GJ. The importance of spleen, spleen size and splenectomy for determining total body iron load, ferritins and iron toxicity in thalassemia major patients. Toxicol Mech Methods. 2013:23:34-41. https://doi.org/10.3109/15376516.2013.735278

PMid:23039902

115. Piga A, Perrotta S, Gamberrini MR, Voskaridou E, Melignano A, Filosa A, Caruso V, Pietrangeli A, Longo F, Tartaglione I, Borgna-Pignatti C, Zhang X, Laadem A, Sherrman A, Attie KM. Luspatercept improves hemoglobin levels and blood transfusion requirements in a study of patients with β-thalassemia. Blood. 2019;133:1279-89. https://doi.org/10.1182/blood-2018-10-879247

PMid:30617198 PMCid:PMC6640181

116. Elafi MS, Adly A, Awad H, Tarif Salam M, Barchous H, Tschida F. Safety and efficacy of early start of iron chelation therapy with deferoxamine in young children newly diagnosed with transfusion-dependent thalassemia: A randomized controlled trial. Am J Hematol. 2018;93:262-8. https://doi.org/10.1002/ajh.24966

PMid:29119631

117. Chuaemanut A, Songdej D, Sirachainan N, Wongwerawattanakoon P, Kadeagsem P, Sasanuk W. Safety profile of a liquid formulation of deferoxamine in young children with transfusion-induced iron overload: a 1-year experience. Paediatr Int Child Health. 2016;36:209-13. https://doi.org/10.1016/j.pediatr.2015.02.021

PMid:25723197

118. Dashti crop M, Kiani C, Dalaei P, Obeureka N, Simmons D, Gatto NM, Zariff M. Iron Accumulation Is Not Homogenous among Patients with Parkinson's Disease. Parkinsons Dis. 2015;2015:324843. https://doi.org/10.1155/2015/324843

PMid:25945281 PMCid:PMC4402185
deferoxamine therapy. Blood. 1996;88:705-13.

can prevent iron-related toxicity in children with thalassemia major. Eur J Pediatr. 2005;164:45-8.

Kontoghiorghes GJ, Spyrou A, Kolnagou A. Iron chelation therapy in hereditary hemochromatosis and thalassemia intermedia: regulatory and non regulatory mechanisms of increased iron absorption. Hemoglobin. 2010;34:251-64.

Harmatz P, Vichinsky E. Combined chelation therapy with deferasirox (deferoxamine) does not protect myocytes against doxorubicin. Free Radic Biol Med. 2003;35:1469-79.

Canadan D, Ormeci AR, Vural H, Aylak F. The effects of deferoxamine on zinc levels in patients with thalassemia major. J Trace Elem Med Biol. 2013;27:109-11.

Eghbali A, Shokri P, Afzal RR, Bagheri B. A 1-year randomized trial of deferoxamine treatment on ocular toxicity induced by desferrioxamine in patients with thalassemia. In Vivo. 2014;28:645-9.

Eghbali A, Shokri P, Afzal RR, Bagheri B. A 1-year randomized trial of deferoxamine alone versus deferasirox and deferoxamine combination for the treatment of iron overload in thalassemia major. Transfus Apher Sci. 2019;58:429-33.

Song TS, Hsieh YW, Peng CT, Chen TL, Lee HZ, Chung JG, Hour MJ. Hour MJ. Combined versus monotherapy or concurrent therapy for treatment of thalassaemia. In Vivo. 2014;28:645-9.
158. Cassinerio E, Orofino N, Roghi A, Duca L, Poggiali E, Fraquelli M, Zanaboni L, Cappellini MD. Combination of deferasirox and deferoxamine in clinical practice: an alternative scheme of chelation in thalassemia major patients. Blood Cells Mol Dis. 2014;53:164-7. https://doi.org/10.1016/j.bcmd.2014.04.006 PMid:24846580

159. Elalfy MS, Saber MM, Adly AA, Ismail EA, Tarif M, Ibrahim F, Elalfy OM. Role of vitamin C as an adjuvant therapy to different iron chelators in young β-thalassemia major patients: efficacy and safety in relation to tissue iron overload. Eur J Haematol. 2016;96:318-26. https://doi.org/10.1111/ejh.12594 PMid:26018112

160. Conte D, Brunelli L, Ferrario L, Mandelli C, Quatrini M, Velio P, Bianchi PA. Effect of ascorbic acid on desferrioxamine-induced urinary iron excretion in idiopathic hemochromatosis. Acta Haematol. 1984;72:117-20. https://doi.org/10.1159/000206370 PMid:6437113

161. Kontogiorghes GJ, Jackson MJ, Lunec J. In vitro screening of iron chelators using models of free radical damage. Free Radic Res Commun. 1986;2:115-24. https://doi.org/10.3109/10715768609088062 PMid:3505236

162. Mostert LJ, Van Dorst JA, Koster JF, Van Eijk HG, Kontogiorghes GJ. Free radical and cytotoxic effects of chelators and their iron complexes in the hepatocyte. Free Radic Res Commun. 1987;3:379-88. https://doi.org/10.3109/10715768709088079 PMid:3508452

163. Walter PB, Macklin EA, Porter J, Evans P, Kwiatkowski JL, Neufeld EJ, Coates T, Giardina PJ, Vichinsky E, Olivieri N, Alberti D, Holland J, Harmatz P; Thalassemia Clinical Research Network. Inflammation and oxidant-stress in beta-thalassemia patients treated with iron chelators deferasirox (ICL670) or deferoxamine: an ancillary study of the Novartis CICL670A0107 trial. Haematologica. 2008;93:817-25. https://doi.org/10.3324/haematol.11755 PMid:18469351