KDIGO Controversies Conference on Optimal Anemia Management in CKD
- Public Review Comments -

As of November 14, 2019
Industry comments are highlighted in blue

Gunnar Heine - Saarland University
Dear Prof. Drüeke, dear Prof. Babitt, since you plan to cover the occurrence of interaction with CKD-MBD parameters [FGF23]) of currently available intravenous agents, we would like to make you aware of HOMe AFers study that compared the occurrence of high FGF-23 and subsequent low phosphorus after intravenous application of either ferric carboxymaltose (FCM) or iron isomaltoside in 25 otherwise healthy women with iron deficient anemia. We found a very high incidence of hypophosphatemia after FCM. The results have been presented at the ASH Meeting 2018, and we are currently drafting a manuscript for peer review. If considered to be of importance by you, we will be most happy to share our findings with you.

Irene Mewburn – Australia (Patient)
Great scope. Personal experience is that diagnosis takes too long (in emergency situations at least 30 hours). Once a patient reaches stage 3 or 4, anemia is inevitable and needs greater recognition in medical circles for diagnosis to happen sooner rather than later. Most patients make errors with EPO medications and can be difficult to self-administer. Need more training. Not to mention the fear of needles in many patients, but once a nurse gave me a practice with saline it was easy and if I had to go back to it give me the needle anytime.

Eleni Frangou - Limassol General Hospital
The Scope of Work is well written with clear aims. During the conference, emphasis should be given to new research findings, new molecular insights and studies on novel agents. Uncertainties should be discussed, and guidelines should be clear regarding both iron and ESA administration. I would be happy to help and be part of this.

Deepak Sharma - Ketav Kalp Healthcare and Research Private Limited
Well researched. Almost all the relevant issues included.

Craig Langman - Northwestern University
This is a long-overdue controversies conference, so I am happy to see it now scheduled. The issue of the treatment of anemia in children on maintenance forms of dialysis is poorly studied and there are few contemporary data, as evidenced by the references noted. As a pediatric practitioner, I hope that the issue is addressed in regards to CV outcomes, general measures of well-being, and even school performance.

Geoffrey Block - Reata Pharmaceuticals, US Renal Care
Congratulations on an outstanding SOW. Really well done! I would add just a few comments to consider: - should we have some common/accepted criteria for defining anemia which is NOT tied /related to the use of ESA's at various stages of CKD. Current practice waits until < 10, often < 9 due to anxiety/concern about ESA effect but whether this is right is another matter. should we actually use WHO definitions and encourage their adoption for assessing/treating anemia in CKD - 'off-target' effects of iron repletion- not to be too self-serving but our recent paper showing reduction in hospitalization and time to RRT with ferric citrate serves as example - establishing a consensus CLINICAL definition of 'iron overload'- MRI imaging definition, bone marrow Fe staining are essentially irrelevant for clinicians and use of ferritin is both wrong and misleading. there needs to be a clear statement about this. - Is 'reducing ESA exposure' an outcome measure? it has clinical and cost implications for sure but is it acceptable/reasonable as an endpoint? - can we aim to establish some standardization of what it means to be epo-'resistant' and suggest a single definition to be used for clinical trial endpoints of iron, HIF, inflammation, hepcidin clinical trials - I think we need a CLEAR set of research goals-prioritized- what are the main current unmet clinical questions that need to be answered. Top 5 or 10. Laundry lists of wish-lists and minutiae don't help guide trial design. - what is standard of care control for iron trials- placebo/ oral iron? - how long should someone be treated with oral iron before they are considered 'failure'. In many clinical trials, 'intolerance' of oral iron was used to allow entry and lumped together with 'oral iron unresponsive'. How to handle?

Simon Roger - University of Newcastle
Should the target haemoglobin be re-explored in the word of HIF stabilisers vs epoetin use? Is it high dose epoetin that causes the problems or achieving it with the new drugs won't cause the problems?

Bill Strauss - AMAG Pharma

Most interested in the topic of hyperphosphatemia and how some IV irons have this reaction and not others. Under group 3 questions 2 and 5, I believe one of the potential adverse effects of some, but not all IV irons is their inducing of hypophosphatemia (via impact on intact FGF23). Since there is growing evidence that this is neither "transient" nor "benign" as once thought, with increasing number of cases being published, I believe this would be a valid subject for Q2, and measuring of serum phosphate in patients at risk in Q5. Note-although patients with CKD have a lower likelihood of hypophos due to their obvious challenge in clearing Phos, they still have been shown to have a significant rate of "severe HypoP" by CTCAE grade 3

Carlo Francisco Santos Gochuico - St. Luke's Medical Center Philippines

Anemia in adults with congenital heart disease and congestive heart failure is a known risk factor for mortality and morbidity. In the article by Konstantinos Dimopoulos et al. entitled "Anemia in Adults With Congenital Heart Disease Relates to Adverse Outcome" submitted in the Journal of the American College of Cardiology Volume 54, Issue 22, November 2009, it was stated that anemia is associated with a 3-fold increased risk of death. In the FAIR HF trial, Piotr Ponikowski et al. studied the impact of intravenous ferric carboxymaltose on renal function (Eur J Heart Fail. 2015 Mar; 17(3): 329–339). They concluded that treatment of iron deficiency in CHF patients with i.v. FCM was associated with an improvement in renal function. FCM therapy was effective and safe in CHF patients with renal dysfunction. There was an improvement in the NYHA, QoL, 6MWD, Fatigue score, and reduced risk of hospitalization for worsening heart failure.

Rolando Claure-Del Granado - Universidad Mayor de San Simon

Group 1. Evaluate if different types of I.V. or oral iron supplementation have different effects on CKD outcomes in non-HD (ND) patients. Does the evidence support the use of i.v. iron over oral iron to treat deficiencies in ND-CKD patients?

Yusuke Tsukamoto - Itabashi Chuo Medical Center, Japan

Regarding the study on outcome, diversity of impact between high mortality country like US and low one like Japan should be compared. For this discussion, I like to recommend additional references: 1) Ishigami, J et al. The impact of hyporesponsiveness to erythropoietin-stimulating agents on time-dependent mortality risk among CKD stage 5D patients: a single-center cohort study Clin Exp Nephrol (2013) 17:106–114 (I am a chief investigator). 2) Hamano T, et al. Kidney International Supplements (2015) 5, 23–32;Thresholds of iron markers for iron deficiency erythropoiesis—finding of the Japanese nationwide dialysis registry

Aleix Cases - Hospital Clinic Barcelona

Dear Sirs, I suggest some additional thoughts to your thorough review on iron and anemia in CKD. I hope you can find them useful Best regards Aleix Cases Question: Is there an evidence of how much iron is too much in CKD, ESRD ¿ The study of Eisenga et al (ref 8) the results of the FIND-CKD and especially the PIVOTAL trial suggest a benefit of iron repletion vs iron deficiency, which is also in line with the results in the general population (Gill D et al. The Effect of Iron Status on Risk of Coronary Artery Disease: A Mendelian Randomization Study-Brief Report. Arterioscler Thromb Vasc Biol. 2017 Sep;37(9):1788-1792) that ID is associated with worse outcomes, but the recent study of the VA suggest that both ID and iron overload are harmful in this population (Cho M et al. An increased mortality risk is associated with abnormal iron status in diabetic and non-diabetic Veterans with predialysis chronic kidney disease.Kidney Int. 2019 Sep;96(3):750-760. Kidney Int. 2019 Sep;96(3):750-760), experimental studies (Vinchi F. Atherosclerosis is aggravated by iron overload and ameliorated by dietary and pharmacological iron restriction. Eur Heart J. 2019 Mar 20. pii: ehz112. doi: 10.1093/eurheartj/ehz112) and an editorial on a possible U-shaped relationship (Kempf T, Wollert KC. Iron and atherosclerosis: too much of a good thing can be bad.Eur Heart J. 2019 Jul 19. pii: ehz506. doi: 10.1093/eurheartj/ehz506. [Question: Are new oral iron formulations better than the classical iron salts for the treatment of ID in CKD ¿ Ferric maltol, ferric citrate, sucrosomial iron, etc

Maria José Romeo - Hospital del Vall d´Hebron

Group 1: Iron, anemia, and outcomes in CKD 1. What is the evidence that anemia and/or iron deficiency cause adverse outcomes in CKD patients? 2. What are the known or expected benefits from iron administration (e.g., reduction in mortality and/or morbidity, such as heart failure, cardiovascular disease, hospitalizations, exposure to ESAs, quality of life, fatigue, cognitive function *** I suggest adding sexual life**)? 3. What are the known or expected harms from iron administration: (e.g., infection, cardiovascular disease, anaphylaxis, oxidant-mediated tissue injury, diabetes, neurodegenerative disorders, kidney disease progression, cancer,*add liver disease**)?

Warm regards, Maria José Soler

Thabit Shahbal - Saudi Pharmaceutical Industries & Medical Appliances Corporation (SPIMACO)

I think the best practice is what mentioned Among patients undergoing hemodialysis, a high-dose intravenous iron regimen administered proactively was superior to a low-dose regimen administered reactively and resulted in lower doses

of erythropoiesis-stimulating agent being administered. (Funded by Kidney Research UK; PIVOTAL EudraCT number, 2013-002267-25. opens in new tab.)

Rumeyza Turan Kazancioglu - Bezmialem Vakif University
All the suggested topic cover the needs Thanks

Pablo Urena - AURA Nord Saint Ouen
The use of iron-based intestinal phosphate binders, such as ferric citrate, as well s the use of iron into the dialysate bath should also be include in Group 3. The hormone erythroferrone, which is produced by erythroblast and acts on hepatocytes to suppress hepcidin production, and thereby increase dietary iron absorption, should be include among the parameters to discuss in Group 4.

Vanessa Cullen - KDIGO Consumer
Please include considerations and discussion of haem and non-haem iron intake and absorption through diet. Food combining can inhibit or promote absorption from both foods and supplements, and some diets and lifestyles are more or less inflammatory. Different cultural diets should be taken into account in assessment of the iron state of CKD patients, in different parts of the world, and in providing advice to patients to improve their iron status. Also please consider anemia prevention by establishing and maintaining good iron intake/absorption and status in CKD Stages 1 and 2 as early adoption of lifestyle interventions can lead to patient empowerment and better outcomes. Also consider when to administer iron infusions based on ferritin status and loss patterns rather than purely on anemia. Thank you

Alessandra Naghettini - Goiás Federal University
important to define criteria for pediatric patients

Roberto Ramírez Marmolejo - Professor, nephrologist
Measure hepcidin if oral iron prescription is your option. The oral iron prescription should be the second option when the use of parenteral iron is not safe for the patient. The decrease in the dose of erythropoietin should be gradual, not greater than 10% at a time, since its variability puts the patient at risk of secondary anemia.

Sunil Bhandari - UK
a few references to consider Nuhu F, Seymour AM, Bhandari S. Impact of iv iron on oxidative stress and mitochondrial function in experimental CKD. Anti-oxidants 2019 press Ziedan A, Bhandari S. Protocol and baseline data for a prospective open-label explorative randomized single-center comparative study to determine the

effects of various intravenous iron preparations on markers of oxidative stress and kidney injury in chronic kidney disease (IRON-CKD). Trials. 2019 Apr 4;20(1):194. doi: 10.1186/s13063-019-3291-x. Sivakumar C, Jubb VM, Lamplugh A, Bhandari S. Safety of Intravenous Iron – Cosmofer and Monofer Therapy in Peritoneal Dialysis and Non-Dialysis-Dependent Chronic Kidney Disease Patients, Peritoneal Dialysis International 2019, 39(2):192-195, 2019 Mar-Apr. https://doi.org/10.3747/pdi.2018/00125 Sunil Bhandari, Dora I.A Pereira, Helen F. Chappell and Hal Drakesmith. Intravenous irons: From basic science to clinical practice. Pharmaceuticals 2018 111, 103; doi:10.3390/ph11040104 Faisal Nuhu, Sunil Bhandari. Oxidative stress and cardiovascular complications in CKD, the impact of anaemia. Iron as Therapeutic Targets in Human Diseases, Pharmaceuticals 2018, 11 (4), 103; 1-15 doi:10.3390/ph11040103 Muñoz M, Gómez-Ramírez S, Bhandari S. The safety of available treatment options for iron-deficiency anaemia. Expert Opinion in Drug Safety 2018 https://doi.org/10.1080/14740338.2018.1400009 Mikhail A, Brown C, Williams JA, Mathrani V Shrivastava R; Evans J; Isaac H; Bhandari S. Renal association clinical practice guideline on Anaemia of Chronic Kidney Disease. BMC Nephrology. 18(1):345, 2017 Nov 30. Munoz M, Gómez-Ramírez S, Besser B, Pavía J, Gomollón F, Liumbruno GM, Bhandari S, Cladellas M, Shander A, Auerbach M. Current misconceptions in diagnosis and management of iron deficiency. Blood Transfusion 2017;15: 422-37 DOI 10.2450/2017.0113-17 Zeidan A. Bhandari S. Anaemia in peritoneal dialysis patients; Iron repletion, current and future therapies Peritoneal Dialysis International 2017 1-2;37(1):6-13. doi: 10.3747/pdi.2016.00193. Kalra PA and Bhandari S. Safety of intravenous iron use in chronic kidney disease. Curr Opin Nephrol Hypertens. 2016; 25 (6); 529-535. Hazara AM, Owen SJ, Bhandari S. The Impact of Lowering Haemoglobin Targets on Patterns of Erythropoiesis-Stimulating Agent Use in Patients on Haemodialysis. Blood Purif. 2016;41(4):287-92. doi: 10.1159/000442280. Epub 2016 Jan 28. Syed A, Bhandari S. "Correction of iron deficiency anaemia using IV CosmoFer in CKD patients with asthma: a prospective study." QJM: monthly journal of the Association of Physicians 2016; 109 (3): 187-90. doi:10.1093/qjmed/hcv117

Leyre Martin-Rodríguez - Hospital Universitario Puerta de Hierro

Dear colleagues, Thank you for opening up the opportunity to contribute to the Conference. I have had the opportunity to read the Conference Overview document with interest. I hereby include my humble comments for your kind consideration. Group 1: - Not every IV iron supplement is equivalent regarding free iron and labile iron, therefore a distinction among different compounds should be addressed when comparing outcomes such as hypersensitivity reactions, infections, or oxidative stress. - I would specifically like to hear about the potential harmful effects

of iron supplementation and vascular calcification. - I would like to hear about the potential effects of oral iron supplements on intestinal microbiota and inflammation. Group 2: - After several studies FAIR-HF, PIVOTAL,...I think there should be a clear statement that Iron correction should be a goal itself, independent of Hb. - Evidence demonstrates fairly enough the lower limits in iron status, but I would like to know "how much iron is too much". - According to the evidence (Pivotal, FIND-CKD...) Should a low frequency high dose--strategy be openly recommended? - Is there a role for different schemas of oral iron supplements such as double dose every other day? Probably related with an overexpresion of ferroporting enabling the overcome of the hepcidin block. Still scarce evidence, but maybe disruptive? Ref: (1)Iron absorption from oral iron supplements given on consecutive versus alternate days and as single morning doses versus twice-daily split dosing in iron-depleted women: two open-label, randomised controlled trials DOI:https://doi.org/10.1016/S2352-3026(17)30182-5. (2) Poster communication in 2019 Congress of the Spanish Society of Nephrology . Alternate day-oral seems to be eficacious in patients with End-Stage CKD. D. Herrero et al. P316. - I would like to hear about the usefulness of evaluating iron deposits in myocardium and liver, and the management of CKD, normally end-stage patients with documented iron overload. What is the adequate use of deferasirox/deferoxamine, including dialysis patients? - I would like to hear about the management of CKD patients with porphyiria. Group 3: - Is there still a role/need for iron supplement with the forthcoming HIF stabilizers? - Is there a role for artificial intelligence models (such as neural networks) in the diagnosis, in the prediction of treatment response or prediction of outcomes? Ref (1) Artificial intelligence for optimal anemia management in end-stage renal disease. Brier ME et al. Kidney Int. 2016 Aug;90(2):259-261. (2) Personalized Anemia Management and Precision Medicine in ESA and Iron Pharmacology in End-Stage Kidney Disease. Brier ME et al. Semin Nephrol. 2018 Jul;38(4):410-417. I hope my comments are useful. Do not hesitate to contact me in case you would wish any further expounding of my observations. Thank you very much for your hard work.
Kind regards Leyre Martin, MD.

Patricia Abreu - UNIFESP

Dear colleagues, thanks a lot for being part of this public review. The key issues that will be discussed involve the global aspects of anemia, especially iron. In Brazil, iron deficiency by parasitosis is common in pre dialysis. In dialysis, anemia usually occurs by non-distribution of ESA. This new KDIGO will help us as an official document to facilitate the discussion with Ministry of Health.
Best regards Patricia Abreu

Rommel Bataclan - University of the East Ramon Magsaysay Medical Center
Five years is an ample time to review new evidences available with regards to this chronic problem in CKD patients. I think we should also determine if pure red cell aplasia is more frequent with the ESA and when do we have to entertain other causes of anemia, necessitating other diagnostics and a multi-specialty approach (i.e. Hematologist, Gastro). It will also be interesting if biosimilar ESA had an impact on safety and compliance among CKD patients, whether on RRT or pre-dialytic.

Joanna Hudson - University of Tennessee
I am excited to learn there will be a conference to address the new information regarding management of anemia of CKD. I find that when referring to the guidelines I am often left saying that the new information/data has not been considered in the current guideline statements. In the area of anemia management of CKD there are many health care providers involved including pharmacists. As a nephrology clinical pharmacist and faculty member I am engaged with many other practitioners with expertise and insight into this topic and issues related to appropriate management of anemia of CKD. I hope this multidisciplinary panel includes clinical pharmacists (ideally more than one to participate in the different work groups) to add to the breadth of insight on this topic. Pharmacologists are listed in the description but not a clinical pharmacist. I look forward to seeing the recommendations of this important conference.

Elizabeth Lindley - Leeds Teaching Hospitals NHS Trust
I'm missing discussions of Hb cycling and ESA resistance but these topics are probably in the scope for the 2020 conference.

Baris Afsar - Süleyman Demirel University
Group 1: Iron, anemia, and outcomes in CKD 1-What is the evidence that anemia and/or iron deficiency cause adverse outcomes in CKD patients? Comment 1: Anemia in CKD is associated with reduced health-related quality of life, increased cardiovascular and all-cause mortality and a higher risk of progression to end-stage kidney disease (ESKD) 2-What are the known or expected benefits from iron administration (e.g., reduction in mortality and/or morbidity, such as heart failure, cardiovascular disease, hospitalizations, exposure to ESAs, quality of life, fatigue, cognitive function Comment 2: New data suggest that liberal iron administration (to be terminated when the ferritin concentration was >700 μg per liter or the transferrin saturation was ≥40%) has beneficial effects in terms of lowering erythropoietin dosage, reduction in the risk for myocardial infarction, need for hospitalization for heart failure and reduction in recurrent cardiovascular events. These outcomes were observed without increased infection risk. (Macdougallet al. N

Engl J Med. 2019 Jan 31;380(5):447-458). However, this study was carried out in patients with low CRP levels, with shorter dialysis vintage, with short follow-up time. Besides the patients in the higher iron dosage arm only received 264mg iron where the planned dosage was 400mg. Thus more studies are needed with extended follow-up, with higher dosages and including more heterogeneous patients (inflamed, functional anemia etc) 3- What are the known or expected harms from iron administration: (e.g., infection, cardiovascular disease, anaphylaxis, oxidant-mediated tissue injury, diabetes, neurodegenerative disorders, kidney disease progression, cancer Comment 3: In theory, iron may be responsible for increased infection rate, anaphylaxis, oxidant-mediated tissue injury. But in everyday daily practice these are very hard to measure. For example it is not very easy to determine oxidant-mediated tissue injury due to iron administration in daily practice. Regarding heart failure and diabetes iron deficiency may be more problematic. One of the most important factors regarding the iron toxicity is free iron or ''non-transferrin bound iron''. To measure free iron, sophisticated methods are needed and I think more studies are needed in these issues 4- Are there data to support the known or expected benefits of iron administration, as defined in #2? Are there differential effects by the route of administration or dosing strategy? Comment 4: As suggested in comment 2 there is now big study showing the proactive high dose iv. Iron therapy has beneficial effects. The researchers have to be celebrated to perform this study. However this study has also been criticized with many aspects. Thus there are more randomized studies are needed to reach firm conclusions. Selection of IV iron dosing regimen and formulation is another important issue. For patients with non-dialysis CKD patients, who are seen less frequently by a nephrologists and for whom the preservation of potential vascular access sites is important, the use of high-dose, low-frequency IV iron dosing may be preferred. In the FIND-CKD study high-ferritin IV iron (ferric carboxymaltose) is shown to be more effective then low dose and oral iron (Macdougall et al. Nephrol Dial Transplant 2014; 29: 843–850). In dialysis patients, more frequent (weekly or biweekly) administration of lower doses of IV iron during regularly scheduled dialysis sessions may be preferred and higher doses may be indicated if serum ferritin and TSAT levels fall below thresholds (Kshirsagar et al.Am J Med 2013; 126: 541) Observations in dialysis patients have shown that bolus: bolus dosing has been associated with higher levels of Hb, TSAT and ferritin and the use of lower ESA doses compared with maintenance dosing. However, this action must not be performed in the presence of active infection.(Brookhart et al. J Am SocNephrol 2013; 24: 1151–1158) 5. Are there are data to support the known or expected harms of iron administration, as defined in #3? Are there differential effects by the route of administration or dosing strategy? Comment 5: The adverse effects of ıron treatment are well-known concern. However, there concerns were based on

anecdotal reports and observational studies and few randomized trials in dialysis patients. A recent meta-analysis including seven randomized, controlled trials and 15 observational studies showed that higher-dose intravenous iron does not seem to be associated with higher risk of mortality, infection, cardiovascular events, or hospitalizations in adult patients on dialysis. However the authors admit that Strength of this finding is limited by small numbers of participants and events in the randomized, controlled trials and statistical heterogeneity in observational studies (Hougen et al. Clin J Am SocNephrol. 2018 Mar 7;13(3):457-467). Of course according to my view, this does not mean that excess iron is not harmful and there is there is likely an upper limit of IV iron dose above which iron loading becomes harmful. But this is not single value and it may change according to patient's characteristics. 6. What is the differential risk of anaphylaxis for the currently available iron formulations? Can we develop a table of reported anaphylactic risk for all available iron formulations to help guide selection? Comment 6: A recent retrospective cohort study conducted by FDA researchers using Medicare data suggested the risk of anaphylaxis may differ among IV iron products, and the risk was maximum for iron dextran and minimum for iron sucrose. (Wang et al. Jama 2015; 314:2062–2068). On the other hand some authors strongly disagree that the risk of anaphylaxis is significantly higher for iron dextran compared to iron sucrose (Auerbach et al. Am J Hematol. 2016, Dec;91(12):E497-E498) However in another meta-analysis by Avni et al. no safety signal with intravenous iron was reported. Minor infusion reactions were increased with intravenous iron but the adverse event profile was markedly greater with oral iron. The formulations of intravenous iron included in the meta-analysis were iron sucrose, ferric gluconate, low molecular weight iron dextran, ferumoxytol, and ferric carboxymaltose (Avni et al. Mayo Clin Proc 2015; 90: 12-23) In March 2015, FDA issued a Drug Safety Communication after a search of the FDA Adverse Event Reporting System (FAERS) database identified 79 cases of anaphylactic reactions associated with ferumoxytol administration. Of the 79 cases, 18 were fatal. Based on these data, FDA strengthened an existing warning that fatal and serious hypersensitivity reactions can occur, regarding these serious risks for ferumoxytol. At the same time the ferumoxytol dosage and administration section underwent a significant modification with the recommendation to dilute the drug, and administer it over at least 15 minutes, in contrast to the original recommendation to infuse undiluted over 17 seconds. Given the March 2015 labeling recommendation to infuse ferumoxytol over a minimum of 15 minutes, it is noteworthy that 18 of the 19 patients who received IV iron by push administration received ferumoxytol, but also that three deaths associated with ferumoxytol occurred following an infusion of at least 15 minutes. Since deaths occurred after the first, second, and third administrations of IV iron, the risk of fatality may exist at any dose (McCulley et al.

Am J Hematol. 2016 Dec;91(12):E496-E497) According to these conflicting findings, FDA's own publication concluded that due to absent head to head studies, it is impossible to determine any relative rates of adverse events among available formulations using current reporting mechanisms (Wysowski et al. . Am J Hematol. 2010 Sep;85(9):650-4). Thus in my opinion, there is not enough evidence to compare the differential risk of anaphylaxis for the currently available iron formulations 7. Are there special populations for which intravenous iron supplementation would be beneficial or should be avoided or minimized? What is the evidence to inform the withholding of IV iron supplementation in the context of active infections, hepatitis B or C, dialysis vintage greater than 4 years, use of a catheter rather than a fistula or graft, or other specialized populations? Comment 7: There is theoretical concern that during active infection microorganisms can use iron and iron should not be used. However a recent study showed that, receipt of intravenous iron among hemodialysis patients hospitalized for infection, showed that there was no difference among patients receiving iron or not receiving iron with regard to iron use, length of hospital stay, 30-day mortality, readmission for infection or death within 30 days of discharge (Ishida et al. Clin J Am Soc Nephrol. 2015 Oct 7;10(10):1799-805).There is also concern of use of iron when high ferritin levels exist. However ''DRIVE'' study showed that iron administration may be still valuable in patients with high ferritin levels (Coyne et al. J Am Soc Nephrol. 2007 Mar;18(3):975-84). To the best of my knowledge there are no guidelines regarding to withhold supplementation in the context of active hepatitis B or C, dialysis vintage greater than 4 years, use of a catheter rather than a fistula or graft. 8. How do iron status, anemia, and/or intravenous iron formulations impact CKD mineral and bone disorder? Comment 8: Iron deficiency stimulates FGF23 transcription but does not cause hypophosphatemia because increased FGF23 production is coupled to increased FGF23 cleavage within osteocytes. The parallel increases in FGF23 production and cleavage in untreated iron deficiency results in secretion of large amounts of C-terminal fragments into circulation that register as high cFGF23 but normal or decreased iFGF23 concentrations. There is one exception to this: only ferric carboxymaltose (saccharated iron oxide) leads to inhibition of FGF23 cleavage relative to production and the biological consequences of excess iFGF23 is observed with ferric carboxymaltose. Due to this alteration ferric carboxymaltose may cause hypophosphatemia, low vitamin D levels, high PTH levels. Hematopoietic stem cells including BFU-E (Burst Forming Unit-Erythroid), CFU-E and proerythroblasts, showed higher amounts of FGF23mRNA. EPO (endogenous or exogenous) increases the total amount of circulating FGF23 (iFGF23 and cFGF23) and alters the iFGF23/cFGF23 ratio in favor of cFGF23. Thus EPO results increased intracellular cleavage of iFGF23 resulting diminished FGF-23 activity. 9. Do iron status, anemia, and/or iron supplementation affect the host immune response or host microbiome?

Comment 9: This probably the one of the most unknown areas. Given the fact that gut microbiota seems to effects various systems and organ functions in the body it needs to be determined and studies are needed with regard to anemia, iron supplementation and different iron products on host immunity and microbiota. Iron has also effects on innate immune system especially on macrophages. Increased intracellular iron polarizes the macrophages toward a pro-inflammatory macrophages (M1 macrophages) (Pereira et al. Cell Rep. 2019 Jul 9; 28(2): 498–511). Excess iron also result in an impairment of the host's immune functions in terms of the T cell and polymorphonuclear leukocyte (PMNL) response. In terms of peripheral blood leukocytes, therapeutic concentrations of iron in media diminished CD4+ lymphocyte survival through the intracellular oxidative stress caused by iron, which leads to apoptosis (Gupta et al. BMC Nephrol 2010;11:16). High doses of IV iron impair the phagocytic activity and diminish the hydrogen peroxide production capacity and microbial killing capability of polymorphonuclear leukocyte (PMNLs). [Patruta et al. J Am Soc Nephrol 1998;9:655-663], Iron sucrose treatment led to impaired phagocytic function and increased apoptosis of PMNLs [Ichii et al. Am J Nephrol 2012;36:50-57]. Thus iron seems to affect the functions of macrophages, lymphocytes and PMNLs. However, the data regarding this issue is relatively scant especially considering CKD patients.

Group 2: Pathogenesis and diagnosis of iron deficiency and anemia in CKD 1. What new insights in systemic iron homeostasis have been obtained in the last decade? What is their relevance for new diagnostic and treatment strategies for iron deficiency in the CKD setting? Is this different for inflamed and non-inflamed patients? Comment 1: It is now clear that the iron hormone hepcidin and its receptor and cellular iron exporter ferroportin control the major fluxes of iron into blood plasma: intestinal iron absorption, the delivery of recycled iron from macrophages, and the release of stored iron from hepatocytes. Hepcidin is in turn feedback-regulated by plasma iron concentration and iron stores (when tissue and plasma iron increase hepsidin functions to decrease plasma iron), and negatively regulated by the activity of erythrocyte precursors, the dominant consumers of iron. In addition a hormone named ''erythroferrone'' produced by human erythroid precursors acts directly on the liver to decrease hepcidin synthesis. Hepcidin and ferroportin also play a role in host defense and inflammation, and hepcidin synthesis is induced by inflammatory signals including interleukin-6 and activin B. The failure to regulate hepcidin homeostasis underlies genetic disorders of iron overload and deficiency, including hereditary hemochromatosis and iron-refractory iron deficiency anemia. In addition in CKD, it was suggested that one of the reasons for anemia (apart from other causes) is the decreased expression of TfR1 in erythroblasts as well as increased hepcidin levels in circulation. This change may hamper erythroblast differentiation by decreasing the iron supply, as iron is an

indispensable component of erythroblast differentiation. Thus the treatment strategies may differ according to underlying pathology. In patients with absolute iron deficiency, the main issue is to replace the iron. However, in functional iron deficiency, it may not be the case. For example one may pay attention to underlying chronic inflammation first in these setting. 2. What is the best definition of iron deficiency and anemia in the CKD setting? Is the definition/diagnosis of iron deficiency still relevant considering the large iron use? Comment 2: According to my view the anemia definition is relevant. Indeed, it is ongoing discussion whether to adjust anemia definition according to age in non-CKD patients (Physiologic anemia of elderly). However as far as I know, the anemia definition has not changed. Thus I think there is not enough data also to change anemia definition in CKD patients. With regard to iron deficiency there is some change. For example even before ''PIVOTAL'' study most patients receive enough iron and Ferritin levels are high in routine daily practice. Thus the safe ferritin levels suggested by KDIGO may be discussed to up-titrate. It is not very clear which laboratory parameter is the best for anemia definition. In a very recent study, Eisenga et al. showed that TSAT, especially TSAT< 10%, is most strongly associated with the risk of adverse outcomes in CKD patients irrespective of serum ferritin level, suggesting that clinicians should focus more on TSAT rather than ferritin in this patient setting. (Eisenga et al. BMC Nephrol. 2018 Sep 12;19(1):225). However, there are also conflicting findings. It is also known that Ferritin is acute-phase reactant. Thus all these factors hinder one parameter to diagnose anemia and iron deficiency perfectly. 3. What is the prevalence of iron deficiency and anemia in CKD? Is this different for various parts of the world? Comment 3: There are various studies showing anemia prevalence in CKD patients. In USA, it was suggested that anemia was twice as prevalent in people with CKD (15.4%) as in the general population (7.6%). The prevalence of anemia increased with stage of CKD, from 8.4% at stage 1 to 53.4% at stage 5. A total of 22.8% of CKD patients with anemia reported being treated for anemia within the previous 3 months–14.6% of patients at CKD stages 1–2 and 26.4% of patients at stages 3–4 (Stauffer et al, Plos one, 2014; 9(1): e84943). A recent study using data from the CKDopps, authors evaluated the monitoring frequency, prevalence and management of anemia and iron deficiency in patients with CKD Stages 3-5 non-dialysis patients in Brazil, France, Germany and the USA. The study sample was comprised of 6766 CKD Stages 3-5ND patients from 135 CKD clinics across the four countries. Within all CKD stages, mean hemoglobin was highest in France, intermediate in Brazil and Germany, and lowest in the USA. In all countries, the prevalence of anemia was higher among patients with lower eGFR. Prescription of iron supplementation and ESAs was more common among patients with lower eGFR and lower hemoglobin levels. Among patients with hemoglobin<10 g/dL, 48% in the USA, 58% in Brazil, 66% in France and 70% in Germany were

prescribed an ESA or iron in the 3 months following hemoglobin measurement (wong et al, Clinical Kidney Journal, August, 2019 epub). In South Africa it was suggested that almost half of the CKD participants were anemic (nalado et al. Int J Nephrol Renovasc Dis. 2019 Feb 18;12:19-32). A study from Catalonia showed anemia prevalence was 58.5%, However, only 14.9% of patients had hemoglobin levels <11 g/dL. Among the patients with anemia (n=295), 36.3% had iron deficiency (Cases-Amenós et al. Nefrologia.2014;34(2):189-98). A study from Chinese showed that anemia was established in 51.5% patients: (51.3% men and 48.7% women) in non-dialysis dependent CKD patients (Li et al. Medicine (Baltimore). 2016 Jun; 95(24): e3872). Lastly, in our country (Turkey) among 60,643 hemodialysis patients, %4 has Hb lower then 8gr/dl, %16.27 has Hb between 8-9.99gr/dl, %21.08 has Hb between then 10-10.99 gr/dl, %29.07 has Hb between 11-11.99 gr/dl and %29.58 has Hb greater 12 gr/dl (registry of the nephrology, Dialysis and Transplantation in Turkey). Thus all these evidence suggest that anemia prevalence changes from country to country. 4: How can iron deficiency and anemia be diagnosed? What laboratory parameters should be used and what are their limitations? Is there a role for functional tests? Is there a clinical relevance for distinguishing absolute iron deficiency from functional iron deficiency and how should they be defined? Is there a role for novel diagnostic tests? Comment 4: For the diagnosis of anemia and iron deficiency various test are used. However in my opinion none of them is %100 satisfactory. For example, transferrin in negative acute phase reactant and effected by nutritional status whereas ferritin is positive acute phase reactant. TIBC is also not perfect since it depends on both transferrin and iron levels. Besides TIBC has wide margin and one cannot be sure regarding iron deficiency unless it decreases much. Although to reticulocyte hemoglobin and hypochromic red cells and Soluble Transferrin Receptor can be used they are not widely available. 5. What are the criteria to initiate therapy with ESA/iron? Should we use clinical or laboratory based criteria or both? Comment 5: I think the suggestions set by KDIGO 2012 guideline are still valid. Given the fact that full normalization of HB is not recommended these suggestions are proper. Besides, as KDIGO 2012 also suggested, there may be a group of patients who may benefit with hb>11.5. Thus individualization of therapy is also important and must be taken into consideration. 6. Are there differences in prevalence, pathophysiology, diagnosis, treatment initiation criteria for iron deficiency and anemia between patients with CKD (non-dialysis) vs on hemodialysis vs on peritoneal dialysis vs pediatric patients vs kidney transplant recipients? Comment 6: The prevalence, pathophysiology, diagnosis differs among patients with non-dialysis CKD, hemodialysis and peritoneal dialysis. In 2012 KDIGO suggested some global recommendations for the use of iron in the treatment of anemia in CKD. However different countries and institutions may have distinct budgetary, regulatory and practical constraints that

impact treatment choices available to physicians. The KDIGO guideline states that in anemic patients with CKD, iron therapy may be required to increase hemoglobin (Hb) levels without the use of ESAs, to boost iron stores prior to initiation of ESA therapy or enhance the response to ESA therapy once initiated or to treat iron deficiency resulting from ESA therapy. Iron stores are mostly assessed by Ferritin and transferrin saturation which both have inherent limitation especially in CKD patients which have high degree of co-morbidity. Beginning with oral iron is not satisfactory for CKD dialysis patients. For non-dialysis CKD patients either oral or iv. iron can be tried but the decision must be based on various factors (severity of anemia, availability of venous access, response to prior therapy, patient adherence and cost). However it is generally agreed that iv. iron is more effective in non-dialysis patients (with respect to elevation of Hb, reaching Hb targets more quickly, lower ESA dose in iv iron). Thus iv iron rather than oral administration of iron may therefore be more appropriate for CKD patients with more severe anemia and iron deficiency, as well as for those receiving ESA therapy. Oral iron may be preferred in early stages of CKD and less anemia. In addition, for preservation of the vasculature to allow for the creation of an arteriovenous fistula formation oral iron may be favored over IV iron. Both the high pill burden and unpleasant side effects associated with oral iron therapy can lead to adherence issues and in these patients high dose with extended interval iv. dosage can be preferred. Patients with high phosphate levels oral phosphate iron binders may also be valuable. Meta-analyses of RCTs assessing IV iron therapy in patients with dialysis and non-dialysis CKD patients demonstrated no appreciable differences in the rates of mortality and adverse events (including CV events and infection) for IV and oral iron treatment groups, but majority of studies evaluated in these analyses had comparatively short follow-up periods (roger et al. Clinical Kidney Journal, 2017, vol. 10, Suppl 1, i9-i15). However studies with longer follow-up also showed no difference. (Macdougall et al. IC, Nephrol Dial Transplant 2014; 29: 843–850 and Roger et al. Nephrol Dial Transplant 2017; 32: 1530-1539). On the other hand one study was terminated early due to increased risk of attributable to iv.iron (Macdougall et al.? Kidney Int 2015; 88:1445-1446). With regard to iron dosing regimen, for non-dialysis CKD patients, who are seen less frequently by a nephrologist and for whom the preservation of potential vascular access sites is important, the use of highdose, low-frequency IV iron dosing may be preferred. One study showed this approach is effective ((Macdougall et al. IC, Nephrol Dial Transplant 2014; 29: 843–850) In dialysis patients the regimen of iv. dosing mostly depends on protocols in dialysis facility. In general, more frequent (weekly or biweekly) administration of lower doses of IV iron during regularly scheduled dialysis sessions is preferred in such patients, although higher doses may be indicated if serum ferritin and TSAT levels fall below thresholds prescribed by the treatment protocol.

Group 3: Use of iron agents in CKD anemia management 1. What are the properties, efficacy (e.g., hemoglobin, iron status, functional, and clinical endpoints), and safety profiles (occurrence of hypersensitivity reactions; occurrence of interaction with CKD-MBD parameters [FGF23]) of currently available oral iron agents to be used in anemia of CKD? How do oral iron agents compare with each other? with IV iron agents? How do we define effectiveness? How do we assess equal or unequal effectiveness? Comment 1: The oral iron preparations would not be satisfactory in hemodialysis patients both due to amount of loss and absorption issues. An older study by Wingard et al. examined the efficiency of different oral iron preparations in stable HD patients receiving erythropoietin. At the end of the study authors concluded that even with a high compliance rate, it is likely that at some point in time the use of intravenous iron would have become necessary. (Wingard et al. Am J Kidney Dis. 1995 Mar;25(3):433-9). In non-dialysis CKD patients, oral ıron may be tried first but this decision is also subject to individualization according to hb levels, iron status, symptoms and co-morbidities. Another study in peritoneal dialysis patients have shown that oral iron treatment either with ferrous sulfate or heme iron polypeptide is not effective in augmenting transferrin saturation or decreasing EPO needs. Thus iv. iron is the preferred method for dialysis patients. Iv iron is both effective for absolute and functional iron deficiency (Pandey et al. SeminNephrol. 2016 Mar;36(2):105-11). Intravenous administration of iron has been demonstrated to be more effective than oral administration with respect to the elevation of Hb, ferritin and TSAT levels in patients with CKD-5D and in those with CKD-ND. Patients receiving IV iron have also been shown to achieve target Hb levels more quickly. A recent retrospective cohort study conducted by FDA researchers using Medicare data suggested the risk of anaphylaxis may differ among IV iron products, and the risk was maximum for iron dextran and minimum for iron sucrose. (Wang et al. Jama 2015; 314:2062–2068). On the other hand some authors strongly disagree that the “risk of anaphylaxis is significantly higher for iron dextran compared to iron sucrose (Auerbach et al. Am J Hematol. 2016, Dec;91(12):E497-E498) However in another meta-analysis by Avni et al. no safety signal with intravenous iron was reported. Minor infusion reactions were increased with intravenous iron but the adverse event profile was markedly greater with oral iron. The formulations of intravenous iron included in the meta-analysis were iron sucrose, ferric gluconate, low molecular weight iron dextran, ferumoxytol, and ferric carboxymaltose (Avni et al. Mayo Clin Proc 2015; 90: 12-23) In March 2015, FDA issued a Drug Safety Communication after a search of the FDA Adverse Event Reporting System (FAERS) database identified 79 cases of anaphylactic reactions associated with ferumoxytol administration. Of the 79 cases, 18 were fatal. Based on these data, FDA strengthened an existing warning that fatal and serious hypersensitivity reactions can occur, regarding these serious risks for ferumoxytol.

At the same time the ferumoxytol Dosage and Administration section underwent a significant modification with the recommendation to dilute the drug, and administer it over at least 15 minutes, in contrast to the original recommendation to infuse undiluted over 17 seconds. Given the March 2015 labeling recommendation to infuse ferumoxytol over a minimum of 15 minutes, it is noteworthy that 18 of the 19 patients who received IV iron by push administration received ferumoxytol, but also that three deaths associated with ferumoxytol occurred following an infusion of at least 15 minutes. Since deaths occurred after the first, second, and third administrations of IV iron, the risk of fatality may exist at any dose (McCulley et al. Am J Hematol. 2016 Dec;91(12):E496-E497) According to these conflicting findings, FDA's own publication concluded that absent head to head studies it is impossible to determine any relative rates of adverse events among available formulations using current reporting mechanisms (Wysowski et al. Am J Hematol. 2010 Sep;85(9):650-4). Thus in my opinion, there is not enough evidence to compare the differential risk of anaphylaxis for the currently available iron formulations.
Selection of IV iron dosing regimen and formulation is another important issue. For patients with non-dialysis CKD patients, who are seen less frequently by nephrologists and for whom the preservation of potential vascular access sites is important, the use of high-dose, low-frequency IV iron dosing may be preferred. In the FIND-CKD study high-ferritin IV iron (ferric carboxymaltose) is shown to be more effective then low dose and oral iron (Macdougall et al. Nephrol Dial Transplant 2014; 29: 843–850). In dialysis patients, more frequent (weekly or bi-weekly) administration of lower doses of IV iron during regularly scheduled dialysis sessions may be preferred and higher doses may be indicated if serum ferritin and TSAT levels fall below thresholds (Kshirsagar et al.Am J Med 2013; 126: 541) Observations in dialysis patients have shown that bolus dosing has been associated with higher levels of Hb, TSAT and ferritin and the use of lower ESA doses compared with maintenance dosing. However, this action must not be performed in the presence of active infection.(Brookhart et al. J Am Soc Nephrol 2013; 24: 1151–1158) With regard to FGF-23, iron deficiency stimulates FGF23 transcription but does not cause hypophosphatemia because increased FGF23 production is coupled to increased FGF23 cleavage within osteocytes. The parallel increases in FGF23 production and cleavage in untreated iron deficiency results in secretion of large amounts of C-terminal fragments into circulation that register as high cFGF23 but normal iFGF23 concentrations. There is one exception to this: only ferric carboxymaltose (saccharated iron oxide) leading to inhibition of FGF23 cleavage relative to production and the biological consequences of excess iFGF23. Due to this alteration ferric carboxymaltose may cause hypophosphatemia, low vitamin D levels, high PTH levels. Hematopoietic stem cells including BFU-E (Burst Forming Unit-Erythroid), CFU-E and proerythroblasts, showed higher amounts of FGF23

mRNA. EPO (endogenous or exogenous) increases the total amount of circulating FGF23 (iFGF23 and cFGF23) and alters the iFGF23/cFGF23 ratio in favor of cFGF23. Thus EPO results increased intracellular cleavage of iFGF23 resulting diminished FGF-23 activity 2. What are the properties, efficacy (e.g., hemoglobin, iron status, functional, and clinical endpoints), and safety profiles (occurrence of hypersensitivity reactions; occurrence of interaction with CKD-MBD parameters [FGF23]) of currently available intravenous iron preparations to be used in anemia of CKD? What is the evidence based data directly comparing efficacy and/or safety among different intravenous iron preparations (e.g., modern versus classic iron preparations and their stability and ligand properties)? Comment 2: As well-known all iv. iron complexes consist of a polynuclea rFe(III)-oxyhydroxide/oxide core that is stabilized with a compound-specific carbohydrate, which strongly influences their physico-chemical properties (e.g. molecular weight distribution, complex stability, and labile iron content). Thus, the carbohydrate determines the metabolic fate of the complex, affecting its pharmacokinetic/pharmacodynamic profile and interactions with the innate immune system. To the best of my knowledge, in nephrology practice no iron preparation is suggested over others with respect to efficiency. Although there is general assumption that iv. Iron dextran is more anaphylactic compared to iv. Iron sucrose this is not accepted as general rule. As Epobio-similars, there are also iron biosimilars however unlike EPO biosimilars, some literature showed that iron biosimiliars were inferior with respect to original iron preperations (Aguera et al. PLoS One 2015; 10: e0135967 and Lee et al. Curr Med Res Opin 2013; 29: 141-147) 3. What should be the optimal treatment strategy with iron supplementation (e.g., how do we define different dosing regimens/strategies: high dose, low dose, maintenance, bolus, reactive versus proactive)? What are the optimal doses, frequency of administration, dosing strategies? Is there a maximal allowable dose? Comment 3: As PIVOTAL study suggests proactive iv. iron administration has favorable effects without increasing infection risk. However, a previous study by Brookhart et al. showed that bolus iv. Iron but not maintenance iv. iron was related with infection-related mortality. The difference between these studies may be due to study design, patient characteristics, dosage of iv. iron etc. Of course there should be a upper limit of Ferritin levels in dialysis patients as guidelines suggest. However, I am not sure that should we up-titrate fe

Guy Rostoker - Hôpital Privé Claude Galien

Why iron balance should be neutral in hemodialysis patients and iron overload avoided ? Almost all hemodialysis patients worldwide are treated by parenteral iron to compensate blood losses and allow full therapeutic effect of erythropoiesis stimulating agents (ESA). Intravenous iron therapy together with ESA actually forms

the backbone of anemia treatment in end-stage kidney disease (ESKD) due to its convenience (infusion during the dialysis sessions), superiority over oral preparations (poorly tolerated) and ability of cost savings of about 20% on expensive ESA molecules [1]. Iron overload in ESKD was considered to be a classical complication of iterative blood transfusions in the pre-ESA era and was believed until recently to be exceptional among hemodialysis patients in the actual ESA-era but is now an increasingly recognized clinical situation diagnosed by quantitative magnetic resonance imaging (MRI) [2]. Since the liver is the main iron storage site in humans, and because liver iron concentration (LIC) correlates closely with total body iron stores in patients with genetic hemochromatosis and secondary hemosiderosis, hepatic magnetic resonance imaging (MRI) has become the gold standard method for estimating and monitoring iron stores in non-renal patients with iron-related disorders [3]. The three MRI modalities for liver iron quantification eg signal-intensity ratio (SIR), R2 relaxometry and R2* relaxometry have been validated in cohorts of non-renal patients with genetic hemochromatosis, hepatic disorders and secondary hemosiderosis requiring liver biopsy for biochemical iron assay [4]. Of note, a recent pilot study in ESKD compared Scheuer's histological classification and Deugnier and Turlin's histological quantitative grading of iron overload by Perls staining with SIR-MRI values obtained with the Rennes University algorithm in 11 hemodialysis patients in whom liver biopsy was indicated in their medical follow-up and showed their close correlations; of note only two of these patients had hepatitis C [5]. Thus, taking into account the fact that the quantitative histological scoring of Deugnier and Turlin has been validated in both hemochromatotic and non-hemochromatotic iron overload disorders, this pilot study strongly suggests that liver iron determination based on SIR-MRI with the Rennes algorithm accurately identifies iron load in hemodialysis patients[5]. Hemodialysis-associated hemosiderosis was recently shown in a pool analysis to be encountered in up to 66% of 500 patients [99% CI: 0.60–0.71] living in various countries, studied by non-invasive radiological methods (Magnetic susceptometry study, n=1; MRI studies; n=10) [1, 6,7, 8]. Moreover, the French study published in 2012 showed severe hepatic iron overload by MRI (> 200 μmol/g liver dry weight, as usually seen in genetic hemochromatosis) in 36 patients (30.2%) of 119 stable hemodialysis patients treated according to the accepted guidelines [8] and iron pancreatic involvement (a marker of severity of iron overload) was investigated in the eight most motivated Israeli dialysis patients with R2* relaxometry and was found in three cases (37%) [7]. The recent PIVOTAL trial has demonstrated strong evidence of a benefit of IV iron in the treatment of anemia in ESKD, with decreased hospitalization rates related to cardiac insufficiency and reduced ESA cost [9]. Conversely, while the PIVOTAL study will result in an increased use of parenteral iron in clinical practice, there is a need to take into account the double-edged sword

of iron therapy [10], especially hepatic iron accumulation in the setting of dialysis, which has been associated with increased hepcidin production [7,8] and a risk of destabilizing atheromatous plaques and triggering cardiovascular events [11] and even deaths [12] and inducing or worsening fatty liver disease [13], together with the increased mortality shown by DOPPS in ESKD patients receiving more than 300 mg monthly IV iron [14]. It is very likely that, major progress in the management of iron status in dialysis patients may soon come from investigational drugs that selectively inhibit hypoxia-inducible factor prolyl hydroxylases (HIF-PH) and stabilize hypoxia-inducible factor (HIF)[2,15]. HIF, a key regulatory protein, stimulates erythropoietin and transferrin production, reduces hepcidin production, and thereby modulates iron absorption and metabolism [2,15]. HIF-PH may also protect against ischemia-reperfusion damage)[15]. It is of interest to note that, phase III trials of HIF-PH in dialysis patients seek to manage iron stores cautiously and physiologically, using oral iron and adopting a target ferritin value of at least 100 µg/L [2,15]. Therefore, the twin risks of iron deficiency and iron overload must still be tightly controlled in dialysis patients on iron therapy until HIF-PH will be used in current practice. References 1) Rostoker G, Vaziri ND. Risk of iron overload with chronic indiscriminate use of intravenous iron products in ESRD and IBD populations. Heliyon. 2019 Jul 12;5(7):e02045. doi: 10.1016/j.heliyon.2019.e02045. eCollection 2019 Jul. 2) Rostoker G, Vaziri N, Fishbane S. Iatrogenic iron overload in dialysis patients at the beginning of the 21st century. Drugs. 2016 May;76(7):741-57. doi: 10.1007/s40265-016-0569-0. 3) Rostoker G. The changing landscape of iron overload disorders at the beginning of the 21st century. Presse Med 2017;46(12 Pt 2):e269-e271. doi: 10.1016/j.lpm.2017.10.011. 4) Paisant A, d'Assignies G, Bannier E, Bardou-Jacquet E, Gandon Y. MRI for the measurement of liver iron content and for the diagnosis and follow-up of iron overload disorders. Presse Med 2017;46(12 Pt 2):e279-e287. doi: 10.1016/j.lpm.2017.10.008. 5) Rostoker G, Laroudie M, Blanc R et al.. Signal-intensity-ratio MRI accurately estimates hepatic iron load in hemodialysis patients. Heliyon. 2017 Jan 5; 3 (1): e00226. doi 10.1016/j.heliyon.2016.e00226.eCollection 2017 jan 6) Ferrari P, Kulkarni H, Dheda S, et al. Serum iron markers are inadequate for guiding iron repletion in chronic kidney disease. Clin J Am Soc Nephrol 2011;6:77-83. doi: 10.2215/CJN.04190510 7) Ghoti H, Rachmilewitz EA, Simon-Lopez R, et al. Evidence for tissue iron overload in long-term hemodialysis patients and the impact of withdrawing parenteral iron. Eur J Haematol 2012;89:87-93. doi: 10.1111/j.1600-0609.2012.01783.x. 8) Rostoker G, Griuncelli M, Loridon C, et al. Hemodialysis-associated hemosiderosis in the era of erythropoiesis-stimulating agents: a MRI study. Am J Med 2012;125:991-999. doi: 10.1016/j.amjmed.2012.01.015. 9) Macdougall IC, White C, Anker SD, et al.; PIVOTAL Investigators and Committees. Intravenous iron in patients undergoing

maintenance hemodialysis. N Engl J Med 2019;380:447-458. doi: 10.1056/NEJMoa1810742. Erratum in: N Engl J Med. 2019 Jan 14. 10) Chinnadurai R, Macdougall IC, Kalra PA. Treatment of anaemia in end-stage renal disease: A double-edged iron sword? EBioMedicine 2019 Jan 16. pii: S2352-3964(19)30005-2. doi: 10.1016/j.ebiom.2019.01.005. 11) Van der Weerd NC, Grooteman MPC, Bots ML, et al. Hepcidin-25 is related to cardiovascular events in chronic haemodialysis patients. Nephrol Dial Transplant 2013;28:3062-3071. doi: org/10.1093/ndt/gfs488 12) Sun L, Zou LX, Lu Y et al. Effect of serum hepcidin on predicting mortality in hemodialysis patients : a prospective cohort study. Iran Red Crescent Med J. 2019 March; 21(3):e87091. doi: 10.5812/ircmj.87091. 13) Rostoker G, Loridon C, Griuncelli M et al. Liver Iron Load Influences Hepatic Fat Fraction in End-Stage Renal Disease Patients on Dialysis: A Proof of Concept Study. EBioMedicine. 2019 Jan;39:461-471. doi: 10.1016/j.ebiom.2018.11.020. Epub 2018 Nov 2 14) Bailie GR, Larkina M, Goodkin DA, et al. Data from the Dialysis Outcomes and Practice Patterns Study validate an association between high intravenous iron doses and mortality. Kidney Int 2015;87:162-168. doi: 10.1038/ki.2014.275. 15) Locatelli F Del Vecchio L. Are prolyl-hydroxylase inhibitors potential alternative treatments for anaemia in patients with chronic kidney disease ? Nephrol Dial Transplant. 2019 Mar 7. pii: gfz031. doi: 10.1093/ndt/gfz031. [Epub ahead of print]

Nicole Casadevall - Saint Antoine Hospital

The scope of coverage of the 4 groups is very detailed and specific. I wonder (although I am not a specialist) if we could not go a little into activin traps ?
With my best, Nicole

Alvaro Garcia Garcia - Grupo Trasplante San Vicente de Paúl

Group 1: Iron, anemia, and outcomes in CKD 1-What is the evidence that anemia and/or iron deficiency cause adverse outcomes in CKD patients? It has been widely elucidated the association of anemia, and increased risk of cardiovascular morbidity and mortality in patients with CKD, even in the early stages of the disease; in a retrospective study of 5885 patients with CKD; when comparing those with anemia (Hb < de 10,5 gr/dl) with patients with normal Hb, it could be demonstrated (Hb< de 10,5); they presented a high mortality risk (HR 5.27, CI: 4.37 -6,35); the risk of hospitalization for cardiovascular pathology was (HR 2.18, CI 1,76- 2.70) and the progression of the CKD to End-stage renal disease ESKD was of (HR= 5.46, CI 3,38 – 8,82) in patients with Hb < de 10.5 g/dl; which demonstrates that anemia is an adverse etiological agent because it increases mortality in general, the risk of hospitalization for cardiovascular pathology and increases the deterioration of the initial stages from CKD to ESKD. (1, 2). The association of anemia and mortality was fully demonstrated in a retrospective work of patients in stage G3-4, (pre dialysis);

in its follow-up during 2 years it was evidenced that its HR decreased when the levels of Hb improved: When the patients had Hb < 11 g/dl their risk was (HR 2.06, 95% CI 1.35 – 3.13) when compared with patients with Hb of 13.9 g/dl; when anemia improved over time to Hb 11- 12 gr/dl, their risk of mortality decreased to (HR 1.8, 95% CI 1.23-2.63) when compared with patients with Hb of 13 g/dl. Lower time –averaged Hb levels also correlated with statistically significant increased risk in the composite end point of predialysis mortality and ESRD as follows: HR 2.57 (95% 1.85-3.58) for Hb < 11 gr/dl, HR 1.97 (95% CI 1.45-2.66) for Hb < 12 g/dl, again, compared with Hb > for13 g/dl (3). In a Japanese study with 1974 pre-dialysis patients, this showed an increase in cardiothoracic radio (CTR) directly related to blood iron levels. This medical pathology can be corrected with the supply of iron which prevents cardiac remodeling (4). The adverse effects of iron deficiency, we can see in another prospective study in 975 patients with CKD, during 8 years of follow-up, 164 patients developed anemia; risk for all-cause mortality was HR 2.83, (95% CI, 1.53-5.24), cardiovascular mortality HR, (4.15; 1.78- 9.66), and the HR from developing anemia was 3.07, (1.69-5.57), which was most evident with a TSAT >10p%. (5). Anemia is a risk factor for all causes of mortality in the CKD patient, as demonstrated by a Japanese study of 62,931 patients studied between 2008-2012; during that time 828 patients died (1.3%), of these classified as non-anemic. (Hb 13 g/dl Men, and 12 Hb g/dl in Women), the % of mortality 1.2 in the (normal) vs. 2.3, en los anemic with a $p < 0.01$, and HR was determined in 2.25 (95% CI, 1.89 – 2.67) with a $p < 0.01$(6) Finally, there is a Korean study in 510,620 patients, followed from 2003 to 2013; 575 women progressed to CKD and 1047 men; at the multivariable –adjusted HRs associate, evidenced that the decrease in 1 g/ Hb in patients with e-GFR ≥ 60, 30 – 59 y < 30 ml/min/1.73 mts^2, were HR 1.34 (CI, 1.17 -1.54), HR 1.55 (1.38-1.74) and HR (1.75 81.47-2.09) respectively; showing that anemia is a high risk for CKD and progression of it. (7)- As we have been able to show, anemia is a determining factor in the risk of morbidity and mortality in patients with CKD. 2. What are the known or expected benefits from iron administration (e.g., reduction in mortality and/or morbidity, such as heart failure, cardiovascular disease, hospitalizations, and exposure to ESAs, quality of life, fatigue, and cognitive function)? From 1985 onwards, nephrology underwent a worldwide 180-degree shift with the discovery of human recombinant erythropoietin. With its systematic and protocolized use in patients with renal anemia, ESAs and iron came to solve a series of comorbidities typical of patients with CKD: Hemosiderosis was decreased by a lower number of transfusions, that of transmissible diseases, HBV, HCV, HIV among others, hyper sensitization to the HLA system (candidates for transplants); the contribution of tissue and therefore muscle strength, cognitive capacity was increased; as well as sleep disorders, the progression of CKD, and cardiovascular diseases (hypertrophy of the left ventricle,

some causes of heart failure) and their mortality were decreased; Daily or labor fatigue improved, as did sexual activity and fertility, as well as decreased costs, the number of hospitalizations in the CKD patient and the risks of anemia treatment (from inpatient therapy) to self-administered outpatient treatment in most cases. In cross-sectional study (survey) in Europe in patients with CKD, describes the association between quality of life and anemia: data related to quality of life and health were obtained by completing the forms EQ-5D, SF-12y, KDQol-36: the presence of anemia was associated with impaired activity levels, in stage 3 CKD, anemic and non-anemic (37.5% vs. 28.4% respectively p=0.0044) in stage 4 (48.1% vs. 39.9% respectively p=0.0292) and in dialysis patients (52.0% vs. 45% respectively p=0.0732), it should be noted that the deterioration was more evident in patients without dialysis, initial stages of CKD (8). In 2 works carried out with the supply of ESAs: the first with the use of darbopoetin α in patient with CKD in total 4038: 2012 with Darbopoetin α and 2026 controls, at the end of the study no change was evidenced on all causes of death, progression of CKD, or episodes of stroke, in the two groups (9). The second work - use of ESAs, to improve anemia in patients with CKD: 603 pts. were taken randomly with e-GFR between 15 to 35 ml/min/1.73 mts; mild anemia was defined Hb 11 to 12.5 g/dl (Group 1), patients with Hb < 10.5 (group 2 with ESAs). During the development of the work, cardiovascular events were similar in the two groups: 58 group 1 vs. 47 group 2, HR 0.78 (95% CI 0.53 -1.14, p = 0.02); ventricular mass was similar, e-FGR was 24.9 vs. 24.2, and the number of patients admitted to hemodialysis 177 (group 1), 111(group 2), p=0.63, in conclusion, ESAs do not improve cardiovascular events in patients with CKD. (10) It has been shown that the treatment of iron deficiency and anemia can reduce the overall mortality of the CKD patient, as well as the risk of hospitalization for cardiovascular disease and deterioration of the early stages from CKD to ESKD, (1, 2). Finally, patients with ESRD are associated with accelerated cerebrovascular disease due to disorders of cerebral circulation caused by uremic toxins; anemia compromises circulation by the vessel dilation itself and the increase in consumption of 02, further deteriorates cerebral circulation, causing loss of cognitive capacity. This work evaluates the effects of anemia correction by measuring: flow velocity, resistance index (RI), pulsating index (PI) in the average brain and correlates with cognitive changes over the time of the study. For it in 120 pts with ESRD, they were classified according to their blood levels of Hb, between 8 - 10 g/dl (state1), these measurements were made and compared when the Hb increased to levels of 10-11, 5 and 11,5- 12, 5 g/dl; all the parameters improved the same thing that the cognitive capacity when going from state 2 (Hb 10 - < 11,5) to 3 (Hb 11,5 ≤ of 12.5 g/d), with a p< of 0.001 (11), which demonstrates that the correction of anemia improves the cognitive capacity of the patient. In addition iron supplementation, increases Hb, improves mean corpuscular volume, increases

ferritin and transferrin saturation TIBC, decreases Erythropoiesis - estimating agent dose and erytpoiesis - estimating agent resistance index value. Serum values of phosphorus, calcium, PTH, do not change significantly, but values of I-FGF-23, increased significantly in the intravenous group (iv), when compared with the oral route (vo); levels of C-FGF23 were significantly reduced with the two methods of iron supply; but levels of interleukin-6, Tumor necrosis factor α, were increased with the supply with the two methods of iron supply (12). 3. What are the known or expected harms from iron administration: (e.g., infection, cardiovascular disease, anaphylaxis, oxidant-mediated tissue injury, diabetes, neurodegenerative disorders, kidney disease progression, and cancer)? The main causes of systemic iron overload are: hereditary hemochromatosis, anemia with iron overload (thalassemia, congenital deseritropoyetic anemia, sidereoblatic anemia, and myelodysplastic syndromes), transfusion overload or other secondary forms. In these cases the iron exceeds the buffering capacity of transferrin, and this free iron is transported by NTBI, which is highly reactive; these forms of free iron when absorbed by the liver, heart, endocrine glands, cause the following pathologies: At hepatic level, when the storage capacity and its anti-oxidant power are exceeded, cirrhosis and hepatocellular carcinoma can be produced; in other diseases such as fatty liver, viral hepatitis are also associated with iron overload; the decrease of hepcidine is part of the mechanism of tissue damage as an important factor in its physiopathology. Diabetes Mellitus is one of the endocrinopathies associated with iron overload; it should be noted that in clinical practice the decrease in circulating iron improves the control of diabetes. Restrictive cardiomyopathy has always been associated with high levels of iron; its etiology is multiple, including injury by oxidants, as an important cause of this pathology. High levels of iron at brain level cause neurodegenerative diseases, caused by pathological deposits of iron in specific places which cause neurodegenerative disorders. In this excellent review of iron metabolism the mechanism of AKI acute renal damage is revealed (iron-induced reactive oxygen species are involved in all AKI models) and otherwise high levels of iron are also part of the increase or persistence some infections by specific bacteria which are called siderophylls like it (Vibrio vulnificus and Yersinea enterocolitics); the other way to avoid a natural anti-bacterial defense mechanism is the sequestration of Hepcidine - Ferroportina by macrophages to decrease iron and prevent the growth of bacteria, but this measure does not protect at all, because it can promote the growth of intracellular forms such as (salmonella). Another important point to take into account is that high levels of iron can alter functions of macrophages / Monocytes themselves as the production of interleukins, and cell migration factors in the immune response. (13). There are no clinical studies that clearly specify the damage caused by the use of iron iv, it is always questionable that there may be that possibility of damage from its use, but the important thing is to

determine how much iron levels measured by ferritin and transferrin saturation are permissible under normal conditions, which do not produce these alterations? 4. Are there data to support the known or expected benefits of iron administration, as defined in #2? Are there differential effects by the route of administration or dosing strategy? Iron deficiency is an independent mortality factor, as we can see in the follow-up of 700 post-transplanted patients (post Tx) the HR for all types of mortality was 1.13-2.78 with a p = 0.01; therefore it is postulated that the (anemia) is a modifiable factor that when corrected improves patient survival. (14). Another similar work in patient with post-tx, evidenced that mortality in the 10 years of follow-up, is related to the degree of anemia: patients with mild anemia Hb 10-11.9 g/dl, had 6 fold higher risk of mortality, when compared with normal patients, also in those with severe anemia < 10 g/dl this risk increased to 10 times more, even in the initial stages 1, 2 of CKD. The conclusion of the study demonstrates that a correct evaluation and treatment of anemia reduces mortality in the initial stages of CKD and in the post Tx (15). With the widespread introduction of ESAs, it became evident that iron supplements are necessary to optimize the response to Hb, decrease doses and costs of ESAS, and possible complications with their use. Iron supplementation is more effective when the intravenous route is used (iv), when compared to the oral route (vo); the use (iv) quickly became popular due to the advantages offered; but is there a latent concern that iron overload can produce oxidative stress, hypersensitivity rations and increase the number of infections (16). 5. Are there are data to support the known or expected harms of iron administration, as defined in #3? Are there differential effects by the route of administration or dosing strategy? Most of the pathologies associated with iron overload are congenital or hereditary and sometimes due to overdoses of iron ir supplied (transfusions, iron, etc.); the physiopathology of these entities is directly related to alterations in the natural regulators of systemic iron, such as hepcidine, ferroportin, or its transporting protein, transferrin. The deregulation of this system of iron control by the organism, allows: overloading in some organs, or tissues deferent to the natural reserve (iron deposition) causing specific pathologies. Other mechanism of damage is presented by oversaturation of the iron transported by (transferrin), which allows the circulation of free iron which is highly toxic, and with a great oxidizing power, which causes tissue damage lesions (oxidative stress) (13). The use of iron iv, partly avoids the action of hepcidine and ferroportin at the level of the intestinal gastrointestinal tract, can expose the patient to high loads of uncontrolled iron. It is worth noting that iron supplementation became the cornerstone in the anemia therapy of the CKD patient; iv iron supplementation is superior to oral iron in patients with RRT (Hemodialysis or PD). On the other hand, iv iron may promote cytotoxicity, tissue injury, exacerbate oxidative stress, and thus produce endothelial dysfunction, as well as inflammation and progression of CKD

and cardiovascular disease. However, anemia correction is effective in correcting oxidative stress and consequently cardiovascular risk (13, 16, 17). 6. What is the differential risk of anaphylaxis for the currently available iron formulations? Can we develop a table of reported anaphylactic risk for all available iron formulations to help guide selection? Lifetime iron supplementation ir is frequently used when iron supplementation is not tolerated or its administration is not effective. This is a cohort study in a single center where 2 types of iron of 3 generation are compared, after administering 1000 mg iv iron isomaltoside vs. iron carboxymaltose in patients with iron deficiency and comparing the hypersensitivity reactions (HSR) that occurred, which were subdivided into severe and non-severe using the Ring and Messner classification: HSR were presented in 18/836 (2.1%), with ferric carboxymaltose and 43/496(8.7%) with isomalt administration. The gross risk of HSR was 75% less after treatment with ferric carboxymaltose (RR=0.248, 95% CI 0.145-0.426 $p < 0.0001$); and the risk of grade II HSR was 88% lower after ferric carboxymaltose (RR=0.123, 95% CI 0.051-0.294). As can be seen, the probability of HSRs was 3.4 times greater with administration of isomalt iron (95% CI 1.910-6.093, $p < 0.0001$). One thing to highlight in this work is that patients with previous comorbidities have a higher possibility of risk for HSR, Regardless of the type of iron used (95% CI 1.899-6.739, $p < 0.0001$)-, Conclusion iron ferric carboxymaltose is associated with a lower risk of 75% when compared with isomalt. (18) In another retrospective study in: 688, 183 patients of the Us-free-for-service Medicare program from January 2003 to December 2013, the risk of anaphylaxis was evaluated, with the IV use of (Iron dextran, Gluconate, Sucrose or ferumoxytol): 274 cases of anaphylaxis were presented in the first exposure, and 170 more cases in the subsequent administration ir of iron. The % of pts with anaphylaxis during the first administration, was 68 per 100,000 for iron dextran (95% CI, 57.8-78.7 per 100,000) with an adjusted odds ratio (OR) of 2.6 (95% CI, 2.0-3.3; $p< 0.001$), at first exposure, when compared with iron sucrose the OR for Iron dextran was 3.6 (95% CI, 2.4-5).4); for iron Gluconate 2.0 (95% CI, 1.2, 3.5); and for ferumoxytol 2.2 (95% Ci, 1.1-4.3); a cumulative risk of anaphylaxis was estimated at 12 weeks after a repletion of 1000 mg with: iron dextran (82 per 100,000 persons, 955 CI, 70.5-93.1) and lower with Iron sucrose 821 per 100,000 persons, 95% CI, 1.3 -26.4). (19) The High Molecular weight iron dextran, represents the first (I) generation of iron, with high risk of anaphylaxis with its ir application; the second generation (II), compose it: ferrous Gluconate and iron sucrose, with a lower risk of anaphylactic reactions, but its biochemical preparation, requires to be administered in small doses every 2 weeks, to reach the required or prescribed dose; the III generation of parenteral iron (isomaltoside vs. ferrous carboxymaltose) have several advantages, it can be used in high doses quickly supplying the needs in one or two applications, its biochemical structure is stable and with very little anaphylactic power (20). 7. Are

there special populations for which intravenous iron supplementation would be beneficial or should be avoided or minimized? What is the evidence to inform the withholding of IV iron supplementation in the context of active infections, hepatitis B or C, dialysis vintage greater than 4 years, use of a catheter rather than a fistula or graft, or other specialized populations? The anemia of the patient in CKD, can be absolute or relative; the absolute is determined, when the TSAT ≤ 20 % and ferritin levels are ≤ 100 ng/ml in pre-dialysis, or, peritoneal Dialysis (PD); and in Hemodialysis (HD), when the TIBC ≤ 20% and ferritin ≤ 200 ng/ml. It should be noted that in patients with normal function and iron deficiency anemia, ferritin is ≤ 20 ng/ml. Functional iron dysfunction (relative) is characterized by TSAT ≤ 20% and one high ferritin 800 ng/ml (29-30). This variability in the diagnosis of iron deficiency, recommends the use of iron IV, stage 5D, patient with ASAs and oral iron use in: CKD ND, or stages 3-5, for 1 to 3 months; if patients with oral iron do not reach the levels of ferritin and transferrin saturation, it is recommended to prescribe iron IV for a time. (21, 22). There are few reports of IV iron use and infection in the medical literature: in a study from Taiwan's National Health Insurance Research, database, the first infection was determined in HD patients in a period of 1.5 years in 1410 pts and these cases were compared with patients receiving IV iron. During the statistical analysis of the problem cases, no difference was found (odds ratio. 1,000, 95%, confidence interval 0.75-1.33) in patients with or without IV iron, including patients with pathologies such as: with diabetes mellitus, chronic lung disease, venous catheter for HD (23). The recorded medical information of the use of IV iron in patients with hepatitis B or C, even in patients with cirrhosis, in the majority are warnings for the risk that can be generated when decompensating or aggravating their previous pathology, if the levels of ferritin and TIBC are not taken into account according to the stage of the CKD, it can increase the overload of iron (from deposit) in the liver deteriorating further its function. In the second instance, the saturation of transferrin with high IV iron loads should be avoided, this allows free iron to circulate which is highly toxic, with a high oxidant power which produces direct tissue damage or increases oxidative stress as another mechanism of damage (13); there are some reported cases of discrete increases in transaminase or some energy with the hepatitis B vaccine (anecdotal reports), and the use of iron in these patients. In the IV iron supply schemes, it has always been taken into account, not to apply, this medicine by fistula or by jugular catheter due to the risk of thrombosis or the compound's own reactions, always mixed with 0.9% saline solution, in a time of 1 to 2 hours with II generation iron compounds; with III generation compounds it is recommended to pass in 15-20 minutes, they are more stable compounds, relatively new, therefore more information must be awaited for their use in order to standardize their application.(18,19) 8. How do iron status, anemia, and/or intravenous iron formulations impact CKD mineral and bone

disorder? In patients with CKD, systemic inflammation and anemia contribute to the increased risk of cardiovascular death; abnormalities in bone metabolism and its micro-environment, associated with inflammation and deregulation of iron, FGF23, a hormone derived from bone metabolism, essential in the metabolism of vitamin D and homeostasis phosphate. In the initial stages of CKD its value is increased to 1000-fold above normal to maintain blood levels of phosphorus. Several studies have shown the association of high levels of FGF3 and cardiovascular death, in addition this hormone is associated with anemia in CKD and inflammation. In this experimental study shows these associations of FGF23, anemia, chronic inflammation in CKD (24). The regulation of FGF23, is complex is not limited alone, as a classic factor at bone level; its regulation is complex and comprises a number of factors, such as erythropoietin, iron deficiency, and inflammation. This is a mechanism in the pathophysiologic of CKD, which presents itself as a new opportunity to evaluate in the treatment of CKD (25). The relationship between FGF23 and anemia in the CKD patient is very evident in a prospective study of 3869 pts evaluated between 2003 and 2008, which had an average e-GFR of 39 ml/min; 1872 were diagnosed with anemia, the prevalence of l-FGF23 was 1.39, 95% interval of 1.26-1.52; at 4 years l-FGF23 was 1.59, 95% with an interval 1.19-2.11 quartile 4, That value was independent of CKD etiology, cardiovascular disease, or mineral metabolism (26). 9. Do iron status, anemia, and/or iron supplementation affect the host immune response or host microbiome? In the context of the immune response due to iron-anemia deficiency, or overload of this nutrient is a little contradictory: it is widely known that the anemic patient with iron deficiency, has a high risk of mortality by infections: gastrointestinal, bacterial systemic, including chronic pathogens such as TB, fungi, others, etc.., by a poor immune response (altered macrophage activity, decreased interleukins, immune response factors such as: interferon α, necrosis factor tumoralβ, aggregating and cell growth factors, which amplify inflammation and response of macrophage/monocyte lines, lymphocytes). On the other hand, the mechanism of protection of the body, with respect to pathogens (bacteria, fungi, strange viruses), is to diminish the effective circulating iron, important for their growth; in this mechanism enters to mediate a protein produced by the liver the hepcidina which regulates the ferroportin, diminishing the absorption of iron by the gastrointestinal tract or the blood contribution of the iron to the reticulum endothelial system, for the erythropoiesis. Iron supplementation, above all venous iron, blood, is theoretically contraindicated, because we are providing IV iron to bacteria for their growth and therefore, aggravating the infectious picture suffered by the patient. (13) In several medical reviews show the importance of iron homeostasis, given that its two extremes: deficiency or overload, affect the course of an acute or chronic infection (27), for erythropoiesis. Iron supplementation, above all venous iron, is theoretically contraindicated, because we

are providing IV iron to bacteria for their growth and therefore, aggravating the infectious picture suffered by the patient. (13) References 1-Gafter- Gvili A Et al. Iron Deficiency Anemia in chronic kidney Disease. Acta Haematol 2019; 142: 44-.50 2-Thorp ML et al. Effect of anemia on mortality, cardiovascular hospitalizations and end – stage renal disease among patients with chronic kidney disease. Nephrology (carlton). 2009 Apr; 14(2):240-6 3-Kovesdy CP, et al. Association of anemia with out-comes in men with moderate and severe chronic kidney disease. Kidney Int. 2006 Feb; 69(3):560-4 4. Hayashi T, Tanaka Y, Iwasaki M, et al. Association of Circulatory Iron Deficiency With an Enlarged Heart in Patients with End-Stage Kidney Disease. J Ren Nutr 2019; 29: 39-47. 5. Eisenga MF, Nolte IM, van der Meer P, et al. Association of different iron deficiency cutoffs with adverse outcomes in chronic kidney disease. BMC Nephrol 2018; 19: 225. 6. Sato Y, Fujimoto S, Konta T, et al. Anemia as a risk factor for all-cause mortality: obscure synergic effect of chronic kidney disease. Clin Exp Nephrol 2018; 22: 388-394. 7- Yi SW, Moon SJ, Yi JJ. Low-normal hemoglobin levels and anemia are associatedwith increased risk of end-stage renal disease in general populations: A prospective cohort study. PLoS One 2019; 14: e0215920. 8. Eriksson D, Goldsmith D, Teitsson S, et al. Cross-sectional survey in CKD patients across Europe describing the association between quality of life and anaemia. BMC Nephrol 2016; 17: 97. 9. Pfeffer MA, Burdmann EA, Chen CY, et al. A trial of darbepoetin alfa in type 2 diabetes and chronic kidney disease. N Engl J Med 2009; 361: 2019-2032. 10. Drueke TB, Locatelli F, Clyne N, et al. Normalization of hemoglobin level in patients with chronic kidney disease and anemia. N Engl J Med 2006; 355: 2071- 2084. 11. Shaker AM, Mohamed OM, Mohamed MF, et al. Impact of correction of anemia in end-stage renal disease patients on cerebral circulation and cognitive functions. Saudi J Kidney Dis Transpl 2018; 29: 1333-1341. 12-Fukao W, et al. Oral Versus intravenous Iron Supplementation for the treatment of Iron Deficiency Anemia in Patients on Mantinance Hemodialysis- Effect on Fibroblast growth Factor -23 metabolism. Journal Renal Nutrition. January 2018 volume 28, issue 4, pages 270-277 13. Dev S, Babitt JL. Overview of iron metabolism in health and disease. Hemodial Int 2017; 21 Suppl 1: S6-S20. 14. Eisenga MF, Minovic I, Berger SP, et al. Iron deficiency, anemia, and mortality in renal transplant recipients. Transpl Int 2016; 29: 1176-1183. 15. Majernikova M, Rosenberger J, Prihodova L, et al. Posttransplant Anemia as a Prognostic Factor of Mortality in Kidney-Transplant Recipients. Biomed Res Int 2017; 2017: 6987240. 16. Lahera v. et al, oxidative Stress in uremia. the Role of anemia correction. J am Soc Nephrol 17: S174-S177, 2006 17- Garmeata Lliliana. Intra venous Iron, Inflammation, ad Oxidative Stress; is Iron a friend or an Enemy of Uremic Patients. J Ren Nutr. 2008 Jan 18(1):40-45 18- Mulder MB, et al. Comparison of hypersensitivity reactions of intravenous iron: iron isomaltoside -1000 (monofer®) versus ferric carboxy maltose (ferinject®). A simple center,

cohort study. BJCP, volume 85, issue 2, febr 2219- pages 385-392 19-Wang C. Et al. Comparative Risk of anaphylactic reactions associated with intravenous iron products. Jama 2015 Nov 17, 14(19):2062-8 20- Biggar P, et al. Importance of different IV iron generation for evereday medical practice. MMW Forschr Med, 2013 Mar 21, 155, Suppl 1:18-24 21- Cappelline MD. et al Iron core group. IRON CORE GROUP. Iron deficiency across chronic inflammatory conditions: international exp opinion on definition, diagnosis, and management, Am J Hematol, 2017 Oct 92(10)1068-78 22- Ratcliffe LE. et al. Diagnosis and management of Iron deficiency recommendations and their rationale .Am J kidney Dis. 2016, apr, 67(4).548-58 23-Yen Cl et al. intravenous iron supplementations does no increase infectious disease risk in hemodialysis patients: A nationwide cohort- base case – crossover study. BCM Nephrol 2019, aug 22, 20(1):327 24- Czaya b, Faul c. The role of fibroblast Growth factor 23 in inflammation and anemia.Crostalk betwen Int Mol Sci 2019 Aug 27;20(17) 4195 25-Babitt Jl, Sitara D, crosstalk between fibroblast growf factor 23 , iron, erytripoetin, and inflammation in kidney disease. Curr opin Nephrol hypertens 2019, Jul, 2884): 304-310 26-. Mehta R, Cai X, Hodakowski A, et al. Fibroblast Growth Factor 23 and Anemia inthe Chronic Renal Insufficiency Cohort Study. Clin J Am Soc Nephrol 2017; 12: 1795-1803. 27-Mariz M, Weiss G. Infection associated with iron administration. Met Lons life sci, 2019 Jan 14, 19.

Group 2: Pathogenesis and diagnosis of iron deficiency and anemia in CKD 1-What new insights in systemic iron homeostasis have been obtained in the last decade? What is their relevance for new diagnostic and treatment strategies for iron deficiency in the CKD setting? Is this different for inflamed and non-inflamed patients? Iron is a vital nutrient for human metabolism. This element has the property of giving (ferrous state) or accepting (ferric estate) electrons, imperative activity in cellular respiration; this property of iron of accepting or receiving electrons can cause severe oxidative stress and tissue damage, in a deregulation of its metabolism. It is also known that iron is important in energy metabolism; its daily absorption (1 to 2 mg/day in the duodenum), its transport and metabolism are strictly regulated, in its daily absorption in the duodenum, since the ability of the body to excrete iron is very limited At the intestinal level, several enzymatic systems and cellular transporters operate in this line: initially the iron in its ferrous state is reduced to ferric by the (cytochrome b ferri-reductase, from the enterocyte brush edge), and is carried inside the cell, by a divalent transporter (DMT-1), there it is stored in the enterocyte as Ferritin, which is excreted by digestive tract, or by action of hepcidin-ferportin, enters into circulation at blood level attached to the transferrin. Another source of plasma iron is recycled daily by the body from red blood cells, which are already senescent phagocytized by reticuloendothelial macrophages and their iron content is used in hematopoiesis or store for further

use. It is fully established that iron regulation is mediated through a small peptic hormone of 25 amino acids, synthesized and excreted by the liver whose function is to bind to ferroportin (cellular transporter of iron); its binding stimulates sequestration and cellular degradation of ferroportin (1, 2); with the presence of hepcidine iron is not absorbed or recycled by the endothelial retic system and its circulating levels are reduced. Hepcidine levels are controlled by multiple stimuli: iron stores, Hypoxia (3,), inflammation, and erythropoiesis (4). Hepcidine is filtered by the kidney and its levels are inversely related to renal depuration (5). In patients with CKD, renal clarence reduction increases their levels and therefore the presence of inflammatory cytokines, as well as reduced erythropoietin levels (6, 7). In the medical literature, the criteria for iron deficiency are fully defined by guidelines such as: WHO, KDIGO, NICE-(EBRP); they establish that absolute iron deficiency occurs when TSAT is ≤ 20% and serum ferritin ≤ is 100 ng/ml in pre-dialysis or peritoneal dialysis (PD); and in hemodialysis (HD) patients the % TSAT is ≤ 20% and ferritin is ≤ 200 ng/ml. In functional iron deficiency, or in inflammatory/infectious states: the diagnosis is made when TSAT is ≤ 20%, despite having an elevated ferritin 800 ng/ml (8, 9). 2. What is the best definition of iron deficiency and anemia in the CKD setting? Is the definition/diagnosis of iron deficiency still relevant considering the large iron use? Iron deficiency in patients with CKD is usually divided into 2 large groups, absolute or functional, these criteria apply to patients with an e-GFR < 60 ml/min/71.73mts^2. Initially anemia was defined by the WHO as Hb < 13 g/dl in men and < 12 g/dl in women (10), these criteria were accepted by KDIGO 2012 (11); The European Renal Best practice: defined it as Hb < 13.5 g/dl, in adult men, 13, 2 in men > 70 years and Hb < 12 g/dl in adult women (12). Iron deficiency takes into account several parameters for its diagnosis: serum iron, ferritin, transferrin and transferrin saturation percentage (TSAT = plasma iron divide by the total iron-binding capacity X100). The diagnostic criteria are different for the two modalities of iron deficiency: Absolute deficiency occurs when TSAT ≤ 20% and serum ferritin ≤ 100 ng/ml in pre-dialysis or peritoneal dialysis (PD); in hemodialysis (HD) patients the % TSAT is equal but ferritin is ≤ 200 ng/ml. It should be noted that in normal patients the diagnosis is made when the ferritin concentration is ≤ 30 ng/ml. Functional deficiency may occur with the use of ESAs, by a rapid erythropoiesis or (iron- restricted erythropoiesis) by an inflammatory state: it is determined with a TSAT ≤ 20% and a high ferritin 800 ng/ml (13). According to these parameters of classification of anemia, a series of guidelines have been made, which show not only the route of administration of iron: oral (ov), or intravenous (iv), but determine the dose of iron and time in which it is expected, to correct this deficiency and medical problems that lead to anemia. 3. What is the prevalence of iron deficiency and anemia in CKD? Is this different for various parts of the world? Data from the National Health

and Nutrition Examination Survey (NHANES) - 2007-2010, determined the prevalence of anemia in patients with CKD in the USA; according to Hb levels ≤ 12 g/dl in women and ≤ 13 g/dl in men, found that 14% of patients in the USA > 18 years of age between 2007-2010 had CKD; and that the prevalence of anemia was 15.4%, 2 times greater than the general population (7, 6%). The prevalence of anemia also varies according to the stages of CKD, 8.4% in stage 1, and 53.4% in stage 5, (14). Absolute or relative iron deficiency in patients who are not on dialysis (CKD ND) can be seen in a study of 933 463 patients in the US Veterans Administration with CKD; of this population 20.6% presented anemia, 23% of them were measured ferritin and TSAT; 30% of patients presented absolute anemia and 19% functional anemia. The following parameters were taken into account for classification: absolute anemia (TSAT ≤ 20%, ferritin < 100 ng/ml) and functional anemia (TSAT≤ 20% and ferritin 100-500 ng/ml); as noted, anemia was associated with increased mortality, dialysis, and hospitalizations due to cardiovascular events (15). In other parts of the world the prevalence of anemia is different: for example: in Korea in a study conducted in 2,198 non dialysis- CKD patients in stages 1 to 5, a high prevalence of 45% was found. Another striking data according to the agreement of Korean National health insurance system, only 7.9% were managed with iron, but 42.75% received ESAs as part of the treatment (16); likewise the incidence of anemia in Japan is slightly lower than in Korea according to an analysis of 1151 references of studies conducted in Japan on anemia and CKD which yields an incidence of 32% in stages 2 to 5 (17). 4. How can iron deficiency and anemia be diagnosed? What laboratory parameters should be used and what are their limitations? Is there a role for functional tests? Is there a clinical relevance for distinguishing absolute iron deficiency from functional iron deficiency and how should they be defined? Is there a role for novel diagnostic tests? The clinical picture of anaemia in patients with CKD is very bizarre and can be very symptomatic (asthenia, adinamia, lack of strength, inability to work, cardiovascular disease, sexual dysfunction, paleness of the skin, etc.).), or go unnoticed and only be noticed for cardiovascular complications, or serious infections: The KDIGO guidelines recommend not measuring Hb routinely, but only when the clinical picture warrants it; or annually in stage 3 of CKD, or 2 times per year in stage 4- 5 ND, or every 3 months in stage 5 on dialysis (5D). In patients who are not receiving ESAs, the recommendation is every 3 months in CKD 3-5 ND, and in CKD stage 5PD, and finally Hb should be determined monthly in stage 5D. (18). In addition to the measurement of Hb, plasma ferritin and TSAT are measured periodically; the diagnosis of anemia has already been determined by WHO, KDIGO, NICE-(EBRP); absolute deficiency when TSAT is ≤ 20% and serum ferritin ≤ of 100 ng/ml in pre-dialysis or peritoneal dialysis (PD); in haemodialysis(HD) patients TSAT≤ 20% and ferritin ≤ 200 ng/ml. Functional impairment may occur with the use of ASDs, by a

rapid erythropoiesis or (airon-restricted erytropoiesis) or by an inflammatory state: which is determined with a TSAT ≤ 20% and an elevated ferritin 800 ng/ml (13). The percentage of hypochromic red cell (%HYPO) and reticulocyte Hb (CHr) are more sensitive functional iron deficiency tests than ferritin and TSAT. A meta-analysis performed for the 2016 UK-based National institute for Health and Care excellence (NICE), guidelines showed that HRC > 6% and CHr < 29 pg is predictive of iron levels (TSAT < 20% and ferritin > 100 ng/ml), it is important to note that this method does not make the difference between absolute iron deficiency and relative deficiency (19), The usefulness of reticulocyte haemoglobin content (CHr) and hypocromic red cell (%HYPO), as markers of iron deficiency and anemia can be seen in another study of 258 black patients vs 141 patients of their staff as controls in the Charlote maxeke Johannesburg Academic hospital, South Africa, from January to December 2016. Where it could be demonstrated that the sensitivity and specificity in the diagnosis of anemia in CKD is high when used for diagnosis a CHr < 28pg and % HYPO> of 6% (20). Another study evaluating these dynamic tests was conducted in China in 150 patients, where reticulocyte hemoglobin content was used as a marker in the diagnosis of anemia in 140 patients; initially a bone marrow aspiration was taken in the department of hematology Peking Union Medical Colle Hospital, also was measured in the blood complete count, including Hb, mean cell volume, corpuscular hemoglobin free erytrocyte protoprphyrin concentration, reticulocyte hemoglobin content, ferritin, serum transferrin receptor, TSAT, and as inflammatory markers Protein C reactive and α-acid-glycoprotein. The final abstract the cut-off value of reticulocyte hemoglobin content in the diagnosis of anemia was 27.2 pg with a sensitivity of 87.5% and a specificity of 92.9%; the cut-off for cell volume, serum ferritin, and serum transferrin receptor were 76.6, 12.9 and 4, 89 mg/L, respectively. The Reticulocyte Hemoglobin content has a high sensitivity and specificity in diagnosis of iron anemia. (21). 5. What are the criteria to initiate therapy with ESA/iron? Should we use clinical or laboratory based criteria or both? The treatment of anemia in CKD patients requires a balance between erythropoiesis stimulating factors (ESAs) and how to maintain sufficient iron stores to ensure optimal production of Hb and other iron functions in the body; Finding new, efficient markers has been a very important task, in the field of nephrology, in May 2012, a record was made of all controlled trials (MEDLINE and Cochare), in which e compared, classical iron deficiency markers and new markers (CHr and %HYPO) vs. erythrocyte Zinc protoporfy ran (ZPP) soluble transferrin receptor (sTfR), Hepcidine, superconducting quantum interference devices (SQUID) and classical iron deficiency markers: serum iron, ferritin, TSAT; all this to establish defined criteria for the initiation of iron supplementation or ESAs. The final conclusion of this review was: that the CHr (with cutoff values of < 27 or < 28 pg) and the % HYPO (with cutoff values of >6% , o <10%), have a greater sensitivity and specified than all

the others studied, including the classics such as (TSAT < 20 or ferritin < 100 ng/ml); in the diagnosis of iron deficiency in CKD, in the different stages (3 to 5) and even in dialysis patients (22). Iron supplementation, especially form iv, reduces the number of transfusions and the need or dose of ASDs, as well as the patient's quality of life (12, 23). KDIGO, the national Kidney Foundation-Kidney Disease Outcomes Quality Initiative (KDOQUI), and the anemia working group of the ERBP, recommends iron supplementation in patients with absolute or relative anemia deficiency. KDIGOs recommend that adults with anemia, without ASDs and not on dialysis (CKD ND) initiate an iron iv trial or vo supply (for 1 to 3 months, in clinically stable patients, without dialysis requirements and without established infection); with the following blood parameters a TSAT < 30% and ferritin <500 ngs /ml, with improvement of symptoms, correction of anemia, Hb, and without requirement of transfusions, during the treatment with iron vo, if it does not tolerate it or it is not fulfilled the programmed one should change to iron iv. In patients with ESAs, who are not receiving iron, an iv iron trial is recommended or oral supply for (1 to 3 months), if the Hb and ASD requirements are on schedule and the TSAT is < 30% and the ferritin is ≤500 ng/ml, sustained and without trasfusional requirement, it should be continued if the above is not met passing to iv iron (11). The EBRP recommends in Europe that patients with CKD and iron deficiency, who are not on dialysis (stages 2-3) or receiving ESAs, or on PD, initiate treatment with oral iron but tolerate doing the switch to iron IV. Determine absolute iron deficiency (TSAT < 20% and ferritin < 100 ng/ml); This guide also suggests oral iron in patients without ESAs and with a TSAT < 25% and > 200 ngs/dl in NKD-ND and ferritin < 300 ng/dl in dialysis (12)- The NICE guidelines suggest oral iron in patients without ESAs, for 3 months, if it does not tolerate them to pass intravenously; and in patients with iron ESAs iv. (19) 6. Are there differences in prevalence, pathophysiology, diagnosis, treatment initiation criteria for iron deficiency and anemia between patients with CKD (non-dialysis) vs on hemodialysis vs on peritoneal dialysis vs pediatric patient vs kidney transplant recipients? The prevalence of anemia is different and depends on several conditions: stages of CKD: in the NHNES study (USA), carried out between 2007-2010 in over 18 years, a prevalence of CKD of 15, 4% was found, of which 7.6 presented anemia, 2 times greater than the general population (7.6%); its distribution according to the stage of CKD was: 8.4% stage 1, and 53.4% for stage 5, it should be noted that only 28% of the anemic started treatment of anemia in the following 3 months (14). In another work on the same subject, the prevalence of anemia was also different according to the GFR of Cr, in the initial stages of CKD 3-4, was calculated at 933, 463 in US Veterans Administration, in those with an e-GFR < 60 ml/min/73mts^2, the following results were obtained: 20.6% presented anemia: 13% absolute iron deficiency and 19% deficiency functional iron. (15) The degree of anemia, symptoms and complications,

can be taken into account to initiate treatment of anemia in CKD: In a study conducted in Northern Denmark from 2000- 2016- anemia was classified according to Hb levels: Grade 1 (10-12/13 gr/dl women/man), grade 2 (8-10g/dl Hb) and grade 3+ (hb < 8 gr/dl), N= 28, 510 patients with anemia, 16,972 CKD patients, 3594 on dialysis (DD) and 24,916 without dialysis (NDD): in the study145 there was no anemia, 35% had G1 anemia, 44% G2 anemia, and 17% G3+ anemia. When purchasing G3 with the other grades of anemia; G3+ had a high HR for dialysis incidents (1.91, 95% CI, 1.61-2.26), for emergency hospitalization (1.74, 95% CI, 1.57-1.93) and for all causes of death (1.82, 95% 1.7-1.94) and MACE (1.14 95% CI, 1.02-1.26). A similar risk (HR) was observed in dialysis patients (DD)-(24). The etiology of anemia in the CKD patient may be different, but in all stages of CKD iron is widely implicated in it; that is why one should have a broad knowledge of its metabolism, in everything concerning its absorption by TGI, transport, daily recycling by the endothelial reticulum system, and of the proteins that control its metabolism: (Hepcytine-ferroportine). In the initial stages of the CKD, in the deficiency of the iron it prevails the lack of the nutritional supply or lost, greater to its income/day by the gastro intestinal or menstrual tract among others etc.; as CKD progresses, other factors intervene such as short half-life of erythrocyte, oxidative stress, chronic inflammation, and relative or absolute deficiency of erythropoietin, high levels of hepcidine, uremic toxins, inflammatory interleukins such as IL-6, necrosis factor tumoralβ, etc. Finally, the KDIGO, NICE- (EBRP); Canadian, Japanese, guidelines have fully established the levels of ferritin and %TSAT, in the diagnosis of absolute or relative anemia according to the stages e-GFR, inflammation and RRT (peritoneal dialysis or Hemodialysis). The new criteria, for anemia and iron deficiency using CHr or %HYPO does not take into account the RRT or CKD stage. It is very little referred to in the literature referred to anemia in the post renal transplant, has as parameters wait the first 3 months after the post-operative period, induction, and prescribe a scheme of immunosuppresses to avoid rejection.

References 1- Nemeth e, et al. Hepcidin regulates cellular iron afflux by binding to ferroportin and inducing its internalization. Scince. 2004 Dec, 306(5704):2093 2. Dev S, Babitt JL. Overview of iron metabolism in health and disease. Hemodial Int 2017; 21 Suppl 1: S6-S20. 3- Shah YM et al. Hypoxia- inducible factores link iron hemeostasi and erythropoiesis. Gastroenterolgy 2014 Mar, 146 (3).630-42 4- Nicolas G et al. The gene encoding the iron regulatory peptide hepcidin is regulated by anemia, hypoxia, and inflammation. J Clin Invest. 2002 Oct, 110 879.1037-44 5- Zaritsky J et al. Hepcidin-a potential novel biomarker for iron status in chronic kidney disease. Clin J Am Soc Nephrol .2009 Jun, 4(6).1051-6 6- Babitt JL et al. Molecular mechanisms of hepcidin regulation: implications for the anemia of CKD. Am J Kidney Dis. 2010 Apr, 55 (4). 726-41 7. Stefanova D, Raychev A, Arezes J, et al. Endogenous hepcidin and its agonist mediate resistance to selected infections by

clearing non-transferrin-bound iron. Blood 2017; 130: 245-257. 8. Michels KR, Zhang Z, Bettina AM, et al. Hepcidin-mediated iron sequestration protects against bacterial dissemination during pneumonia. JCI Insight 2017; 2: e92002. 9. Schwartz AJ, Das NK, Ramakrishnan SK, et al. Hepatic hepcidin/intestinal HIF- 2alpha axis maintains iron absorption during iron deficiency and overload. J Clin Invest 2019; 129: 336-348. 10- Word Health Organization technical report series No 405: nutritional anemias-report of a WHOS scientific group. Geneva . WHO; 1968 11- Kidney Disease: Improving Global Outcomes (KDIGO), Anemia Work Group-KDIGO clinical practice guideline for anemia in chronic kidney disease. Kidney Int Suppl. 2012; 281):279-335 12- Locatelli F et al. ERA-EDTA ERBP Advisory Board. Kidney disease: Improvining global Outcomes guidelines on anemia management in Chronic kidney disease: A European Renal Best Practice position statement. Nephrol Dial transplant. 2013 Jun, 28(6):1346-59 13-Cappelline MD et al. Iron deficiency cross chronic inflammatory conditions: international expert opinion on definition, diagnosis, and management. Am J heamatol, 2017 Oct, 92(109):1068-78 14-Stauffer ME, Fan T. Prevalence of anemia in chronic kidney disease in the United States. PLoS One 2014; 9: e84943. 15-Awan AA, et al. Prevalence, correlates and outcomes of absolute and functional iron deficiency anemia in no dialysis –depend chronic kidney disease. Nephrol Dial Transplant 2019 Oct 22- Doi:10.1093/ndt/gfz192 16. Ryu SR, Park SK, Jung JY, et al. The Prevalence and Management of Anemia in Chronic Kidney Disease Patients: Result from the Korean Cohort Study for Outcomes in Patients with Chronic Kidney Disease (KNOW-CKD). J Korean Med Sci 2017; 32: 249-256. 17. Akizawa T, Okumura H, Alexandre AF, et al. Burden of Anemia in Chronic Kidney Disease Patients in Japan: A Literature Review. Ther Apher Dial 2018; 22: 444-456. 18-Kidey Disease: KDIGO, anemia work Group- KDIGO clinical practice guidelines for anemia in chronic kidney disease. Kidney Int Suppl.2012,2(1):279-335 19-Ratcliffe LE, et al. Diagnosis and management of iron deficiency in CKD: A summary of the NICE guidelines recommendations and the rationale. Am J kidney Dis. 2016 Apr 67(4).548-58 20-Nalado AM et al. Utility of reticulocyte haemoglobin content and percentage hypochromic red cell as markers of iron deficiency anaemia among black CKD patients in south Africa. PloS One, 2018 Oct 3, 13(10): e0204899 21-Cai J. et al. Evaluation of the efficiency of the reticulocyte Hemoglobin cont. on diagnosis for iron deficiency anemia in Chinese adults. Nutrients 2017 may 2;9(5). pii: E450. 22-Chung M, et alt. Biomarkers for Assessing and managing iron deficiency anemia in late-stage chronic kidney disease: AHRQ Publication No. 12(13)-EHC140-EF Oct 2012. 23- Kliger AS et al. KDOQI Us commentary on 2012 KDIGO clinical practice Guideline for anemia end CKD. Am J Kidney Dis .2013 Nov 62(59):849-59, 24-Toft G, et al Anemia and clinical outcomes in patients with non-dialysis dependent or dialysis dependent severe chronic kidney

disease: A Danish population –base study. J Neprhol Oct 5. Doi 10.1007/s40620-019-00652-9

Dwarakanathan Ranganathan - RBWH

Is it safe to continue Iron therapy in dialysis patients in the presence of infection
What is the upper limit of ferritin in a patient receiving Iron therapy in the presence of systemic infection.

Sandra Waechter - Vifor Pharma

Since iron has a role beyond erythropoiesis, we believe that iron deficiency should be discussed as a separate condition to anemia and the physiological consequences of iron deficiency as well as the benefits of iron repletion in iron-deficient individuals with and without anemia should be reviewed during this KDIGO Controversies Conference. Please consider the following clinical evidence and corresponding references for inclusion in your discussion: In recent years, evidence has been published about the deteriorating effects of iron deficiency in patients with CKD and iron deficiency has been identified as an independent risk factor for worse outcome [1-4]. On the other hand, there is a growing body of evidence in patients with HF showing that correction of iron deficiency with IV ferric carboxymaltose (FCM) improves symptoms, exercise capacity and quality of life [5,6]. In the CONFIRM-HF trial, treatment with IV ferric carboxymaltose for > 1 year, was even associated with a significant reduction in the risk of hospitalization for worsening HF [6]. Further, a recent patient data meta-analysis investigating the effect of FCM vs. placebo in HF patients with ID showed that hospitalizations for HF and mortality were significantly decreased in the iron-treated group. More than 40% of subjects in this analysis had eGFR < 60 ml/min per 1.73 m2 [7]. The assumption from all of these studies is that, since these effects were seen independently of changes in hemoglobin, they are mediated via improvements in cardiac and/or skeletal muscle function following FCM therapy. This assumption is further substantiated by the RED-HF trial in which treatment with darbepoetin alfa did not improve clinical outcomes in patients with systolic heart failure and mild-to-moderate anemia [8]. Interestingly, approximately 50% of the subjects enrolled in the RED-HF trial were iron deficient at baseline when applying the definitions given for iron deficiency in the ESC guideline [9,10]. In patients with ND-CKD, the TREAT trial also showed no mortality or cardiovascular benefits in the group treated with darbepoetin [11]. While both these trials were designed to measure a treatment effect related to hematologic improvements, the iron status was not taken into account when discussing the observed outcome. Hence, post-hoc analyses to evaluate the impact of iron deficiency on the observed outcome would be most desirable for both these

trials since these analyses could help to further enhance the understanding of the condition to be targeted, i.e. iron deficiency or anemia, to improve outcome in this population. Finally, in the context of the recently published PIVOTAL trial [12], we believe it would be interesting to understand why we did see the results we did. Whether more iron is protective – or more ESA is harmful; or whether the subjects in the low-dose arm were iron deficient and experienced worse outcome due to this uncorrected condition. It should be noted as well that while substantial evidence exists to support the use of FCM to improve exercise capacity and functional outcome, these benefits could not be shown with oral iron in patients with heart failure [13}. On the other hand, there is evidence that oral iron, although to a lesser extent than IV iron, can support erythropoiesis when combined with ESA therapy in patients with CKD [14]. In addition, emerging evidence is arising that oral iron has a negative impact at microbiome triggering inflammatory response in the intestines, which can lead to or aggravate IBD or colorectal cancer [15]. Oral iron adversely changes gut microbiota composition, the gut and systemic metabolome, and host immunity and infection in iron deficient predialysis CKD patients [16]. Both, the PIVOTAL trial and FIND-CKD [17], inform about the benefits when treating to higher ferritin levels and/or transferrin saturation: while in FIND-CKD targeting a higher ferritin level of 400 to 600 ng/mL with IV ferric carboxymaltose delayed or reduced the need for alternative anemia treatment in patients with ND-CKD, subjects being treated with a pro-active IV iron dosing regimen in the PIVOTAL trial had a lower risk of dying and/or experiencing a major cardiovascular event. Additionally, in PIVOTAL, the ESA consumption and transfusion rate could be reduced in the group of subjects being treated with higher doses of IV iron. On the other hand, no safety signal could be identified that was associated with the exposure to higher amounts of IV iron in neither one of the two trials. In fact, in PIVOTAL, the incidence of infection and hospitalization for any cause did not differ between the two treatment arms and a post-hoc safety analysis of FIND-CKD revealed no different safety profile for subjects who reached a ferritin greater than 800 ng/ml at least once in the course of this trial [18]. We believe that the results of these two trials have to be taken into account when reviewing best treatment practice in terms of ferritin and TSAT levels. While we believe the evidence listed above should support the use of IV iron in patients with CKD to improve their outcome, we are concerned by recent prevalence data published from the CKDOPPS cohort: including data from 2013 to 2018, this analysis showed that 50 % of patients with CKD stage 3 to 5ND were iron deficient. The analysis further identified that a high proportion (46%) of patients with anaemia and either ferritin < 100 ng/mL or TSAT < 20% were not treated with iron, even among those with persistent hemoglobin <10 g/dL on two consecutive measurements [19]. Practice pattern data (data on file Arbor Research for CKDOPPS cohort) as well as Market Research data show that anemia treatment, and there

included iron therapy, is only initiated once the Hb drops below 10 g/dL, although the 2012 KDIGO guideline on renal anaemia management recommends iron treatment initiation in all patients with an Hb below 12 or 13 (if female or male, respectively) and ferritin levels < 500 and TSAT < 30%. This deserves attention, especially in light of an analysis with DOPPS data showing that 53% of patients in this cohort had an Hb < 10g/dL at dialysis initiation and that low Hb at initiation of dialysis was associated with a higher first year HD-mortality [20]. When it comes to the differentiation of the different IV iron products available on the market, we would like to highlight that all IV iron complexes consist of a polynuclear Fe(III)-oxyhydroxide/oxide core that is stabilized with a compound-specific carbohydrate, which strongly influences their physico-chemical properties (e.g. molecular weight distribution, complex stability, and labile iron content). Thus, the carbohydrate determines the metabolic fate of the complex, affecting its pharmacokinetic/pharmacodynamic profile and interactions with the innate immune system. Accordingly, IV iron products belong to the new class of non-biological complex drugs for which regulatory authorities recognized the need for more detailed characterization by different methods, particularly when assessing generic/follow-on products. Evaluation of published clinical and non-clinical studies with different IV iron products in this review suggests that study results obtained with one IV iron product should not be assumed to be equivalent to other IV iron products that lack comparable study data. Without head-to-head clinical studies proving the therapeutic equivalence of one versus another IV iron product extrapolation of results and substitution with a different IV iron product is not recommended [21].

Reference list 1. Klip IT et al. Eur J Heart Fail 2014 2. Eisenga M et al. BMC Nephrol 2018;19:225 3. Cho ME et al. Kidney Int. 2019;96 4. Dziegala M. et al. J Cachexia Sarcopenia Muscle. 2018;9(5):802-815 5. Anker SD et al. N Engl J Med. 2009;361(25):2436-48 6. Ponikowski P et al, Eur Heart J. 2015;36(11):657-68 7. Anker SD, et al. Eur J Heart Failure. 2018;20, 125–133 8. Swedberg K. et al. N Engl J Med.2013;368(13):1210-9 9. McMurray JJV et al. European Journal of Heart Failure. 2013;15: 334–341 10. Ponikowski P. et al. European Heart Journal. 2016 ;37:2129–2200 11. Pfeffer MA et al. N Engl J Med. 2009;361:2019–32 12. Macdougall IC et al. N Engl J Med. 2019;380(5):447-458 13. Lewis GD et al. JAMA. 2017; 317(19): 1958–1966 14. Quinibi WY et al. Nephrol Dial Transplant. 2011;26(5):1599-607 15. Yilmaz et al. Pharmaceuticals (Basel). 2018;11(4) 16. Kortman GAM et al. Hemodial Int. 2017 Jun;21 Suppl 1:S28-S36 17. Macdougall IC, et al. Nephrol Dial Transplant.;29(11):2075-84 18. Roger SD, et al. Nephrol Dial Transplant. 2017;32: 1530–1539 19. Wong MMY, et al. Clin Kid J 2019:1-12 20. Karaboyas A. et al. Clin Kid J 2019; 1-9 21. Martin-Malo et al. ESC Heart Fail. 2019 Apr;6(2):241-253

Marta Christov - Westchester Medical Center

An area where clinical direction is lacking is anemia management in the AKI or AKI on CKD setting, especially in hospitalized patients. Such patients frequently become anemic while in the hospital and may initiate dialysis temporarily. Key practice questions become: should patients be treated with iron? Should such patients be started on ESAs and when? Is there efficacy or safety information about the use of iron preparations or ESAs in the AKI population? Are there downsides? I am interested in helping pursue some of these questions and would love to work with others on the topic.

Best, Marta Christov

Adriana Penalba - Hospital Angel C Padilla

1.what are the upper limits of ferritin that contraindicate intravenous iron treatment in patients with stage 5D chronic kidney disease

Adebowale Adekoya - Nephrology Association of Nigeria

Some Sickle Cell Disease patients present with features of nephropathy and often require management of their worsening anaemia. However, there is no consensus on the target haemoglobin in this group of chronic kidney disease patients. Let us have a robust expert opinion on this.

Kind Regards, Dr Adebowale Adekoya ISN/KRUK Fellow Consultant Physician/Nephrologist MBBS,MSc,FMCP (Nephrology), DTM&H(London),PhD(Sheffield),PGCE (Liverpool)

Richard Watt - Brigham Young University/Revitale Pharma

Inflammation causes elevated hepcidin which prevents intestinal iron absorption and blocks iron redistribution from the liver, spleen and macrophages causing Anemia of Chronic inflammation (ACI). Hepcidin inhibitors are essential CKD treatments to stabilize ferroportin and restore iron redistribution to the bone marrow. Previous targets include blocking HAMP mRNA production by inhibiting inflammatory pathways, however these targets fail because multiple inflammatory pathways are co-activated and inhibiting only one pathway cannot stop HAMP transcription. Hepcidin binding molecules such as antibodies, anticalins and RNA aptamers have struggled in trials due to the quantity of hepcidin present and its rapid rebound once hepcidin is cleared. The Hif-alpha prolyl hydroxylase drugs show promise but concerns relating to the potential activation of angiogenesis, MMP activation, and tumorigenesis have the dialysis field wary about their use. The Watt Lab at Brigham Young University and Revitale Pharma have taken a new approach to block the activation of prohepcidin to hepcidin by inhibiting the protease Furin. This method is independent of inflammatory pathway activation and prevents

active hepcidin from being secreted. The novel furin inhibitors being advanced by Revitale Pharma are drugs currently marketed and can be repurposed so a treatment can be available in 3-5 years due to existing safety data. Only mild side effects of diarrhea in 10-14% of patients exist. The potential to use Furin inhibitors to block hepcidin formation as a treatment for ACI is a new and promising approach.

Ray Pratt -Rockwell Medical

Dear Drs. Drueke and Babbitt, Thank you for seeking comments for the upcoming KDIGO meeting at Barcelona. In this regard we would like to share information on TRIFERIC (ferric pyrophosphate citrate) and request that it be included in the program for discussion. In this submission we have provided the following: 1. Introduction to TRIFERIC as outlined below 2. A list of key publications for reference 3. Answers to specific questions from the KDIGO working group sessions 4. An attached Word document with a more comprehensive overview of TRIFERIC Thank you for your consideration. Very truly yours, Dr. Ray Pratt 1. Introduction: TRIFERIC was approved by the FDA in 2015 as the first iron product indicated to be administered with each hemodialysis treatment to replace ongoing iron losses and maintain hemoglobin concentrations. TRIFERIC is also the first carbohydrate-free iron salt deemed suitable for parenteral administration. TRIFERIC is approved as a proactive iron therapy in adult patients with hemodialysis-dependent chronic kidney disease (HDD-CKD) that is delivered via the dialysate to replace the ongoing iron losses. IV iron, on the other hand, is only approved for intermittent administration to treat established iron deficiency anemia in adult hemodialysis patients. Though PIVOTAL trial provides evidence of safety of a proactive approach to intravenous iron the safety and efficacy of proactive IV iron therapy has yet to be established in prevalent hemodialysis patients. TRIFERIC is administered via the dialysate at every hemodialysis treatment, crosses the dialyzer membrane, directly donates iron to transferrin in blood compartment and delivers iron to the erythroid precursors in bone marrow. As would be clear from the publications listed below, TRIFERIC: • Effectively treats functional iron deficiency in CKD-HD by donating iron directly to transferrin, • Maintains hemoglobin without increasing serum ferritin and does not overload tissue iron stores. Hence TRIFERIC can be administered to patients with high serum ferritin levels, • Can be administered to patients with allergy to IV iron (since TRIFERIC is devoid of any carbohydrate moiety). There has been no cases of anaphylactic reactions to TRIFERIC in over 1 million doses administered to date. Therefore TRIFERIC holds great potential for use during home HD, • Does not generate NTBI in the therapeutic dose of 2 µM iron per liter of final dialysate; and does not induce oxidative stress or inflammation despite 3 times a week administration over 9 months (Gupta eta l, PRIME study, KI 2015), • TRIFERIC does not affect hepcidin-ferroportin axis (as compared to IV

iron), • TRIFERIC not associated with increased risk of infections. 2. Key publications for Reference Full prescribing information is available on www.TRIFERIC.com under the PI tab. The major original studies on TRIFERIC are listed below (see URLs below). • The PRIME study (a Phase 2 study) demonstrating that TRIFERIC reduces the requirements for ESAs needed to maintain hemoglobin in hemodialysis patients (http://www.kidney-international.org/article/S0085-2538(15)60995-4/pdf) • The CRUISE studies (Phase-3 studies) demonstrating maintenance of hemoglobin in hemodialysis patients by TRIFERIC (http://ndt.oxfordjournals.org/content/early/2015/07/13/ndt.gfv277.full.pdf+html) • "Physicochemical characterization of ferric pyrophosphate citrate" in BioMetals. (https://rdcu.be/9lIG) • “Ferric pyrophosphate citrate: interactions with transferrin” in Biometals (http://link.springer.com/article/10.1007/s10534-018-0142-2) 3. Answers to specific questions from the KDIGO working group sessions. GROUP #1. 2. What are the known or expected benefits from iron administration (e.g., reduction in mortality and/or morbidity, such as heart failure, cardiovascular disease, hospitalizations, exposure to ESAs, quality of life, fatigue, cognitive function)? Anemia is an inevitable complication in patients with chronic kidney disease who receive maintenance hemodialysis (CKD 5HD). This is primarily because of loss of renal erythropoietin production, chronic inflammation, and increased blood losses related to uremia and hemodialysis. The result is an iron loss of ~5–7mg per dialysis session. (Babitt 2012, Sargent 2004) Although erythropoietin deficiency and inflammatory suppression of erythropoiesis can be partly counteracted by erythropoiesis-stimulating agents (ESAs), increased erythroid iron requirements because of ESAs, together with ongoing blood losses, exceed the amount of iron that can be provided from adequate marrow iron stores. Furthermore, chronic inflammation suppresses the iron supply that is available from stores by stimulating hepatic production of hepcidin. (Goodnough 2010, Zubrennen-Bullough 2014). Hepcidin prevents efflux of iron from stores into plasma. (Ganz 2011) Iron is then retained within reticuloendothelial (RE) macrophages in bone marrow, liver, and spleen. Plasma transferrin saturation (TSAT) falls, making iron inaccessible for red blood cell production. (Thomas 2013) Intravenous (i.v.) iron is commonly administered in hemodialysis patients to replace dialysis-related blood losses and to overcome inflammatory sequestration of iron. A novel iron therapy, Ferric pyrophosphate citrate, (FPC) delivered via dialysate during hemodialysis replaces iron losses, maintains Hgb concentrations, does not increase iron stores and exhibits a safety profile similar to placebo. (Fishbane 2015). FPC administered by hemodialysis via dialysate represents a paradigm shift in delivering maintenance iron therapy to hemodialysis patients. 6. What is the differential risk of anaphylaxis for the currently available iron formulations? Can we develop a table of reported anaphylactic risk for all available iron formulations to

help guide selection? Anaphylaxis has not been observed in over 1,500,000 doses of FPC administered during clinical trials and post-marketing (Data on file, Rockwell Medical Inc, Wixom MI, USA); while other iron compounds have been reported to cause anaphylaxis at a rate of 20 or more cases per million doses administered (Wang et al. 2015). Whether the unique chemical structure of FPC, including lack of a carbohydrate moiety, are responsible for the growing evidence of enhanced safety remains to be determined. (Gupta 2018) Patients with iron allergy were included in the clinical studies of ferric pyrophosphate citrate (Gupta 2015, Fishbane 2015). Two patients with documented allergy to iron dextran (Dexferrum™) received FPC for 248 treatments without hypersensitivity reactions. One patient with documented allergy to sodium ferric gluconate (Ferrlecit™) received FPC for 237 treatments without hypersensitivity reactions. One patient with documented allergy to ferumoxytol (Feraheme™) received FPC for 130 treatments without hypersensitivity reactions. (Data on file, Rockwell Medical Inc, Wixom MI, USA). Among the 588 patients who received FPC in the CRUISE study, there was one potential reaction: a patient on placebo who transitioned over to open label (OL) and developed mild flushing and tingling when FPC was started. This resolved with Benadryl and FPC was discontinued. The patient declined work up or referral to a dermatologist for further evaluation. The study investigators suspected this was not a true reaction but since the patient declined a re-challenge or skin testing, it was reported as a mild reaction. (Data on file, Rockwell Medical Inc, Wixom MI, USA). Since then, with over 1,500,000 doses of FPC administered, there have been no reports of any reactions or any cases of anaphylaxis.

GROUP #2. 5. What are the criteria to initiate therapy with ESA/iron? Should we use clinical or laboratory based criteria or both? Both clinical and laboratory-based criteria fall short of identifying iron therapy requirements. Anemia is an inevitable complication in patients with chronic kidney disease who receive maintenance hemodialysis (CKD 5HD). This is primarily because of loss of renal erythropoietin production, chronic inflammation, and increased blood losses related to uremia and hemodialysis. The result is an iron loss of ~5–7mg per dialysis session. (Babitt 2012, Sargent 2004) In Fishbane's study, regular administration of FPC during hemodialysis by addition to the hemodialysis solution was shown to be well tolerated, effectively replacing ongoing dialytic and uremic iron losses, thereby maintaining iron balance and Hgb concentration. Maintenance iron therapy using FPC represents a paradigm shift in management of anemia in chronic hemodialysis patients.

GROUP #3. 2. What are the properties, efficacy (e.g., hemoglobin, iron status, functional, and clinical endpoints), and safety profiles (occurrence of hypersensitivity reactions; occurrence of interaction with CKD-MBD parameters [FGF23]) of currently available intravenous iron preparations to be used in anemia

of CKD? What is the evidence-based data directly comparing efficacy and/or safety among different intravenous iron preparations (e.g., modern versus classic iron preparations and their stability and ligand properties)? Administration of ferric pyrophosphate citrate (FPC) via hemodialysate allows replacement of ongoing uremic and hemodialysis-related iron losses. FPC donates iron directly to transferrin, bypassing the reticuloendothelial system and avoiding iron sequestration. In a study published by Fishbane et al in 2015, two identical Phase 3, randomized, placebo-controlled trials (CRUISE 1 and 2) were conducted in 599 iron-replete chronic hemodialysis patients. Patients were dialyzed with dialysate containing 2 μM FPC-iron or standard dialysate (placebo) for up to 48 weeks. Oral or intravenous iron supplementation was prohibited, and doses of erythropoiesis-stimulating agents were held constant. The primary efficacy end point was the change in hemoglobin (Hgb) concentration from baseline to end of treatment (EoT). Secondary end points included reticulocyte hemoglobin content (CHr) and serum ferritin. In both trials, Hgb concentration was maintained from baseline to EoT in the FPC group but decreased by 0.4 g/dL in the placebo group ($P < 0.001$, combined results; 95% confidence interval [CI] 0.2–0.6). Placebo treatment resulted in significantly larger mean decreases from baseline in CHr (−0.9 pg versus−0.4 pg, $P < 0.001$) and serum ferritin (−133.1 μg/L versus−69.7 μg/L, $P < 0.001$) than FPC treatment. The proportions of patients with adverse and serious adverse events were similar in both treatment groups. FPC delivered via dialysate during hemodialysis replaces iron losses, maintains Hgb concentrations, does not increase iron stores and exhibits a safety profile similar to placebo. 3. What should be the optimal treatment strategy with iron supplementation (e.g., how do we define different dosing regimens/strategies: high dose, low dose, maintenance, bolus, reactive versus proactive)? What are the optimal doses, frequency of administration, dosing strategies? Is there a maximal allowable dose? An optimal treatment strategy for iron supplementation in hemodialysis patients who are iron replete should be to replace iron in a method that most closely mimics natural, physiologic iron absorption and metabolism. FPC administered every session as per package insert (2 uM/L of dialysate), delivers 5-7 mg of iron to replace current iron losses. Pharmacokinetics analysis of FPC showed that all the administered iron complexes with transferrin and that it is rapidly cleared from the plasma, with a mean apparent terminal-phase half-life of 1.2 h. (Pratt et al. 2017) Therefore, FPC is not expected to accumulate or lead to iron overload. This represents a concurrent or proactive approach to replacing the small amounts of iron loss at every hemodialysis session. The only approved dose is to deliver FPC in the prescribed manner to deliver approximately 5-7 mg of iron each HD session to provide maintenance replacement of iron. This represents the approved maximum allowable dose in the maintenance of iron stores in hemodialysis patients. Other iron products should be used for iron

repletion and FPC utilized to physiologically replace ongoing losses in iron replete patients. 6. How to use iron supplementation in various patient populations? Should the choice of iron preparation, dosing strategy, treatment targets, or other parameters be modified (and how so) in different patient populations (e.g., patients with CKD [nondialysis] vs on hemodialysis vs on peritoneal dialysis vs pediatric patients vs kidney transplant recipients; patients with an active infection; patients with liver disease; patients with heart failure; patients with calciphylaxis; other special circumstances)? Ferric pyrophosphate citrate (FPC) is an iron replacement product indicated for the replacement of iron to maintain hemoglobin in adult patients with hemodialysis-dependent chronic kidney disease (HDD-CKD). FPC is not intended for use in patients receiving peritoneal dialysis. FPC has not been studied in patients receiving home hemodialysis. (Triferic™ prescribing information 2018)
GROUP #4. 9. What is the evidence regarding cost-effectiveness of novel therapeutic agents for treating anemia of CKD? In a study published by Gupta et al, FPC delivered via dialysate reduces the prescribed ESA dose and the amount of intravenous iron needed to maintain hemoglobin in chronic hemodia

Ajay Gupta - UC Irvine
Dear Dr. Babitt and Dr. Drueke, Below, please find a brief literature review that highlights the emerging evidence in preclinical studies that iron oxide nanoparticles (IONPs) administered by the intravenous route have effects on macrophage phenotype and the HIF-erythropoietin axis, thereby blunting the therapeutic action of HIF-PHI. Kindly note that this is being submitted by myself as an individual and does not reflect the opinion of that of my employer, Rockwell Medical. Summary Iron oxide nanoparticles (IONPs) that are suitable for intravenous use (i.v. iron) are iron carbohydrate complexes with a molecular mass of 45,000 to 750,000 Dalton15. IONPs are internalized by cells of reticuloendothelial system (RES) including macrophages and fibroblasts Iron loading and prolonged exposure to iron in the tissue microenvironment leads to a macrophage polarization profile of a pro-inflammatory (M1-like) phenotype which associates with tissue damage during inflammatory disease 1-3. Conversely, acute iron deprivation has in vivo protective effects mediated by an anti-inflammatory immune-metabolic switch in macrophages4 (Pereira et al, 2019). RES is central to the biology of hypoxia-inducible factor (HIF), and the beneficial effects of inhibitors of prolyl hydroxylase domain (PHD) inhibitor (PHI) drugs. The primary beneficial effect of PHIs on erythropoiesis is increase in HIF generation in renal fibroblasts which promotes transcription and translation of erythropoietin by the kidney. IONPs are internalized by renal fibroblasts and intracellular iron accumulation leads to detrimental effects on the HIF-erythropoietin axis. IONPs stimulates PHDs, thereby degrading HIF and

inhibiting erythropoietin production by renal fibroblasts5. IONPs inhibit transcription of HIF and suppress erythropoietin expression by promoting oxidative stress in mice with unilateral urinary obstruction6. In a mouse model of phlebotomy induced anemia, IONPs inhibited HIF, erythropoietin transcription, and decreased erythropoietin levels by about 25%6 . PHIs have beneficial effects on iron metabolism. PHIs decrease plasma hepcidin levels, the major regulator of iron metabolism7, thereby promoting gastrointestinal iron absorption and ferroportin mediated release of iron from RES. IONPs, on the other hand, markedly increases hepcidin levels and would antagonize beneficial effects of PHI on iron metabolism. PHIs have potential cardioprotective effects. PHI suppress hepcidin, and hepcidin retards macrophage-induced cardiac repair and regeneration through modulation of IL-4/IL-13 pathway, while macrophages lacking hepcidin have been shown to promote cardiomyocyte proliferation and robust reduction in both myocardial infarct size and tissue fibrosis8. By increasing hepcidin levels, IONPs could potentially antagonize the cardioprotective effects of PHI. PHIs have reno-protective effects in models of acute ischemic and inflammatory kidney diseases and chronic kidney disease. In adenine induced model of experimental tubulointerstitial chronic kidney disease, roxadustat, a PHI, reduced inflammatory infiltration by macrophages by shifting macrophages from an inflammatory phenotype to a regulatory, anti-inflammatory phenotype thereby attenuating renal dysfunction and tubulointerstitial damage9. In fact, acute iron deprivation reduces the severity of macrophage-dependent crescentic glomerulonephritis by limiting glomerular cellular proliferation and has in vivo protective effects mediated by an anti-inflammatory immunometabolic switch in macrophages10. Conversely, IONPs promote the inflammatory M1 phenotype thereby antagonizing PHI action. PHI including roxadastat promote vascular calcification11. As a marker of hypoxia, serum HIF-1alpha level may be an independent risk factor for the presence of coronary artery calcification in diabetic patients12. In ESRD, elevated inorganic phosphate is a potent stimulator of vascular calcification (VC), an effect that is mediated by HIF-1μ subunit stabilization and promoted by PHI. Intravenous iron also promotes vascular calcification and consequently is contraindicated in patients with calciphylaxis. Therefore, there is a possibility that use of PHI in CKD-HDD patients in conjunction with i.v. iron could lead to an additive or synergistic effect on VC13. The pathogenesis of enhanced vascular calcification with i.v. iron may be related to Fe(III) induced depletion of pyrophosphate, a potent anti-calcific molecule . Triferic is unlikely to promote vascular calcification since it replenishes the limited pyrophosphate pool in CKD-HDD patients and has negligible free Fe(III)14. Conclusions In summary, therapeutic actions of PHI on erythropoiesis, renal protection and cardiovascular protection are mediated by the reticuloendothelial system including renal fibroblasts, renal macrophages and

cardiac macrophages. PHI promote an anti-inflammatory immune-metabolic switch in macrophages. IONPs administered parenterally are internalized by RES and promote a macrophage polarization profile of a pro-inflammatory (M1-like) phenotype, thereby antagonizing PHI action. Reference List 1. Kroner A, Greenhalgh AD, Zarruk JG, Passos Dos Santos R, Gaestel M, David S. TNF and increased intracellular iron alter macrophage polarization to a detrimental M1 phenotype in the injured spinal cord. Neuron. 2014;83(5): 1098-1116. 2. Sindrilaru A, Peters T, Wieschalka S, et al. An unrestrained proinflammatory M1 macrophage population induced by iron impairs wound healing in humans and mice. J Clin Invest. 2011;121(3): 985-997. 3. Vinchi F, Costa da Silva M, Ingoglia G, et al. Hemopexin therapy reverts heme-induced proinflammatory phenotypic switching of macrophages in a mouse model of sickle cell disease. Blood. 2016;127(4): 473-486. 4. Pereira A, Alvares-Saraiva AM, Konno FTC, et al. B-1 cell-mediated modulation of M1 macrophage profile ameliorates microbicidal functions and disrupt the evasion mechanisms of Encephalitozoon cuniculi. PLoS Negl Trop Dis. 2019;13(9): e0007674. 5. Suzuki N, Vojnovic N, Lee KL, Yang H, Gradin K, Poellinger L. HIF-dependent and reversible nucleosome disassembly in hypoxia-inducible gene promoters. Exp Cell Res. 2018;366(2): 181-191. 6. Oshima K, Ikeda Y, Horinouchi Y, et al. Iron suppresses erythropoietin expression via oxidative stress-dependent hypoxia-inducible factor-2 alpha inactivation. Lab Invest. 2017;97(5): 555-566. 7. Chen N, Hao C, Liu BC, et al. Roxadustat Treatment for Anemia in Patients Undergoing Long-Term Dialysis. N Engl J Med. 2019;381(11): 1011-1022. 8. Zlatanova I, Pinto C, Bonnin P, et al. Iron Regulator Hepcidin Impairs Macrophage-Dependent Cardiac Repair After Injury. Circulation. 2019;139(12): 1530-1547. 9. Schley G, Klanke B, Kalucka J, et al. Mononuclear phagocytes orchestrate prolyl hydroxylase inhibition-mediated renoprotection in chronic tubulointerstitial nephritis. Kidney Int. 2019;96(2): 378-396. 10. Pereira M, Chen TD, Buang N, et al. Acute Iron Deprivation Reprograms Human Macrophage Metabolism and Reduces Inflammation In Vivo. Cell Rep. 2019;28(2): 498-511 e495. 11. Mokas S, Lariviere R, Lamalice L, et al. Hypoxia-inducible factor-1 plays a role in phosphate-induced vascular smooth muscle cell calcification. Kidney Int. 2016;90(3): 598-609. 12. Agharazii M, St-Louis R, Gautier-Bastien A, et al. Inflammatory cytokines and reactive oxygen species as mediators of chronic kidney disease-related vascular calcification. Am J Hypertens. 2015;28(6): 746-755. 26: 1137–1145 13. Neven,E. et al.: Iron and vascular calcification. Is there a link? NDT 2011; 14. Gupta, A Crumbliss, AL: Treatment of iron deficiency anemia: Are monomeric iron compounds suitable for parenteral administration 15. Crisponi et al: Toxicity of Nanoparticles: Etiology and Mechanisms Ch 18 Antimicrobial Nanoarchitectonics 2017.

Amy Pai - University of Michigan College of Pharmacy

IV iron formulations are defined by the FDA to be complex drugs and are suspensions of nanoparticles ranging from 1-40 nm in hydrodynamic radius. Because of their approval date they have not been adequately studied to determine biodistribution which will greatly vary based on particle size. Additionally, the US in particular will be facing challenges with the use of generic formulations as iron sucrose formulation. The global experience with generic formulations has demonstrated that there are significant challenges with bioequivalence evaluation that affect the efficacy and safety profiles of these agents. The following details the Scope of Work, Specific Aims and Areas for Further Study identified through my work as Principal Investigator on request for application from the FDA (1U01FD004892-01) to address labile iron release from IV iron and bioequivalence of generic formulations of IV iron. Notably, I was also requested to speak at the Generic Drug User Fee Act Public Forums at the FDA in 2016 and 2017. I also attended the first iron controversies conference in 2014. Achievement of Scope of Work by Specific Aims In vitro studies Specific Aim 1. Formulation study – Commercially available IV iron complex formulations including Venofer®, Ferrlecit®, generic sodium ferric gluconate complex (Watson Laboratories, Inc), InFeD®,Feraheme®, as well as GEH121333 a novel iron formulation developed by GE Global Research were fully characterized in terms of molecular weight, particle size distribution and physicochemical characteristics (PCC). • Some differences in the PCC between the RLD Ferrlecit® and generic SFGC were noted, however, based on in vitro and in vivo analyses of labile iron release, evaluation of only PCC for candidate generic agents will not be sufficient to describe labile iron release profiles. Specific Aim 2. Evaluation of labile iron in vitro – Each IV iron complex formulation was evaluated for appearance of labile iron by assays that detected chelatable iron and assays that determine redox active iron in both saline and serum matrices. • The HPLC-DFO assay was optimal for analysis across in vitro and in vivo experiment conditions at doses of 40 mg/kg or equivalent estimated Cmax concentrations. • High concentrations of agent prohibitively interfered with the performance of the FL-DFO, the Rhodamine, and the BDI assays, with the BDI assay additionally limited by the assay's inherent complexity. In vivo studies Specific Aim 3. Pharmacokinetic study in preclinical species – Pharmacokinetic studies with each of the iron complex formulations were conducted in a rat model by measuring serum non-transferrin bound iron (labile iron) with an HPLC-DFO assay. • A 40 mg/kg was found to be optimal for detection of labile iron using the HPLC-DFO assay across time points measured. Specific Aim 4. Establish the relationship between in vitro labile iron data and in vivo NTBI data – A systems analysis approach was utilized to evaluate the potential for an IVIVC for each IV iron complex formulation. • The serum concentration-time profile of labile iron can be measured using an HPLC-DFO assay

and modeled using a 1-compartment model. • Pharmacokinetic profiles for release constant (Ka) showed distribution into two groups: Venofer®, Ferrlecit®, generic sodium ferric gluconate complex (Watson Laboratories, Inc) and InFeD®,Feraheme®, GEH121333 • The in vivo release rate constants are similar for Ferrlecit® and SFGC • Data on rate of elimination of complex-associated labile iron are not available but if studied and measured may improve the final model. Discussion: IV Iron Use is Increasing in A Capitated Payment Environment: Trials raising concerns about erythropoiesis-stimulating agents (ESA), revisions to ESA labeling, and changes to practice guidelines and dialysis payment systems have provided strong stimuli to decrease ESA use. These data coupled with the prospective payment system (PPS) which went into effect in January 2011 have influenced the rapid increase intravenous iron (IVI) administration in recent years.1 The financial impact of medications in the PPS is significant and the use of lower cost iron agents will be desired among dialysis providers. The number of published studies of intravenous iron in other disease states (e.g pregnancy, inflammatory bowel disease, post bariatric surgery, post gastrointestinal bleed etc) has risen in the last five years which suggests an overall trend towards increasing intravenous iron use across populations. The necessity for infusion clinic use will favor lower cost IV iron products (i.e. generics) to optimize reimbursement. Currently, in contrast to other countries, the US has only one FDA-approved generic IV iron product, however other companies have filed ANDAs for iron sucrose.2 Clinical Use of IV Iron Formulations-An Example of Putting the Cart Before the Horse: Clinical use of IV iron formulations entered clinical practice beginning in the late 1950's, which preceded the nanomedicine exploration frontier.3 Thus, these agents were approved without full exploration of labile iron release profiles or comprehensive biodistribution studies. Most studies have relied on plasma pharmacokinetic analyses that require many model assumptions to estimate contribution of the iron-carbohydrate complex to elevations in iron indices and hemoglobin.4 The only exception is ferumoxytol that is measurable by NMR, however, the plasma PK profile still represents only a small part of the disposition of IV iron agents.5 Current commercially available intravenous iron formulations consist of an iron oxyhydroxide core surrounded by carbohydrate shells of various sizes and polysaccharide branch characteristics.3The size of commercially available intravenous iron-carbohydrate complexes range from 5 to 100 nm, and thus meet the definition for nanoparticles.3 The manufacture of iron-carbohydrate complex formulations is highly sensitive to pH, temperature and other conditions in the manufacturing process. This presents significant challenges to reproducible manufacturing, characterization and safety of generic or "similar" intravenous iron product production.3 Iron oxide nanoparticles with magnetic particle cores are well-established MRI agents and have been used safely, however, different

carbohydrate shell structure determines the relative uptake by endothelial and lymphatic cells as well as the by the reticuloendothelial system.3 The clinical use of iron-carbohydrate nanoparticle formulations has not been well studied with regard to potential long-term toxicity beyond immediate labile iron appearance.3,6 Because commercially available intravenous iron formulations, inclusive of generics, used in chronic kidney disease meet the criteria for nanoparticles, their pharmacodynamic profile with regard to direct cell uptake and subsequent physiological effects needs to be better characterized.7 Labile Iron Release from IV Iron Formulations-Need for Biorelevant Analysis: The hypothesis for the pathogenesis of acute oxidative stress induced by intravenous iron formulations is the release of iron from the iron-carbohydrate structure resulting in transient concentrations of labile plasma iron and induction of the Fenton chemistry and the Haber-Weiss reaction promoting formation of highly reactive free radicals such as the hydroxyl radical.8 Among available IV iron formulations, products with smaller carbohydrate shells are more labile and more likely to release labile iron directly into the plasma (i.e. before metabolism by RES). The proposed biologic targets of labile-iron-induced oxidative stress include nearly all systemic cellular components including endothelial cells, myocardium, liver as well as low density lipoprotein and other plasma proteins. Because of the extremely short half-lives of free radicals and the rapidity of the ensuing oxidative stress reactions produced by labile iron appearance, in vivo evaluation of this toxicity profile can only reasonably be accomplished by using biomarkers as surrogates. Recently, a systematic review of widely used biomarkers to assess oxidative stress in chronic kidney disease was conducted. The authors applied scores for commonly used biomarkers for relationships to other biomarkers and clinical indicators, reliability and characterization in the CKD literature.9 Many of the identified “robust” biomarkers have been evaluated in the context of potential intravenous iron toxicity in CKD (e.g. malondialdehyde, protein carbonyl and F2-isoprostane), however, it should be noted that none of the identified biomarkers have specificity for iron-induced oxidative stress. An additional concern regarding appearance of labile plasma iron is the potential for easily accessible iron to augment bacterial growth and increase the risk of infection.10 As we have investigated and confirmed in our in vitro analyses in a biorelevant matrix (rat serum) and in vivo, labile iron release profiles differ among available formulations. It is critical to understand that comprehensive evaluation of PCC is not sufficient to predict labile iron release of these agents in biorelevant matrices and in vivo. Other methods of estimating the potential for formulation-based labile iron release have relied on mathematical modeling based on structural characterization but do not factor in disposition changes that occur in vivo.11 As we and other groups have observed, conditions the IV iron agents (e.g. dilution) are exposed in preparation for analysis may affect behavior of the

compounds or result in failed analysis attempts.11 Moreover, some compounds that have similar PCC and have met or nearly met USP criteria, still exhibit differential toxicity profiles in vivo.12,13 Thus, it is clear that PCC alone will not be sufficient to inform labile iron release and the existing data in the literature considered in tandem with data presented in this report suggest further validation of our model would be useful to further inform bioequivalence of IV iron agents filing ANDAs. More data, including lot-to lot variation and larger numbers of generic products with known deviations in USP criteria, will allow further validation of a Ka (in vitro) to Cmax (in vivo) correlation. Biodistribution of RLD and Generic Products: Commercially available intravenous iron formulations meet the criteria for nanomedicines. Their pharmacodynamic profile with regard to direct cell uptake and subsequent physiological effects needs to be better characterized. These agents have not been well studied with regard to comparative biodistribution, metabolic fate and potential extracellular and intracellular toxicity profiles and further evaluation of these agents is urgently needed. Additionally, there are no data evaluating the metabolic fate of the carbohydrate shell which may impact efficacy and toxicity profile. Elford et al. recently studied iron concentration profiles using ICP-MS following injection of Venofer and the generic iron sucrose product from Azad Pharmaceuticals.14 They examined iron concentration within target storage organs (liver, spleen, bone marrow) following a 15 mg/kg dose in rats. The mean weight of the rats was 159 grams which would equate to an average dose of 3.9 mg. Subtracting the observed measurements from rats receiving only vehicle (ng/g tissue) from the iron amount deposited after IV iron administration shows that these organs only account for approximately 0.6 mg of the administered dose. This suggests that there are other potential sites with avid iron uptake (e.g. endothelium, monocytes) that have not yet been evaluated. The European Medicines Agency's reflection paper articulating the agency's thoughts and evidence base for suggested enhanced data requirements for bioequivalence notes the importance of biodistribution data.15 Section 2.2.2 Table 1 of the document suggests the relevant compartments for the distribution of intravenous iron-based nanoparticles include plasma, the reticuloendothelial system and other target tissues which include but are limited to bone marrow, kidney liver (hepatocytes) lungs and heart. As discussed previously these organs may only represent a small portion of locations where iron is deposited. The panel also states "Development of additional and more accurate analyses of the degradation process of nanoparticles is encouraged." Ultimately, biodistribution studies are limited for commercially available agents and it is unknown but highly plausible, that the metabolic fate of intravenous iron agents may affect local labile iron release. Thus, biodistribution studies are relevant to both RLD and generic formulations Future Directions: The scope of work completed under this U01 funding confirms that PCC alone is insufficient to predict

formulation-based labile iron release, has successfully identified an optimal candidate assay to detect labile iron in vitro and in vivo and suggests the potential utility of correlation labile iron release profiles between in vitro and in vivo analyses. Further validation of the model needs to be performed with additional lots of products as well as with other generic iron products with documented differences in clinical toxicity profiles (i.e iron sucrose products in the international market). Additionally, biodistribution studies will help to substantiate RLD and generic bioequivalence as well as further the understanding of the complex pharmacokinetics and pharmacodynamics of these widely used agents. References Cited 1. Hirth, RA, Turenne, MN, Wheeler, JR, Nahra, TA, Sleeman, KK, Zhang, W, Messana, JA: The initial impact of Medicare's new prospective payment system for kidney dialysis. Am J Kidney Dis, 62: 662-669, 2013 2. http://investor.sagentpharma.com/releasedetail.cfm?ReleaseID=740303 3. Duncan R, Gaspar R. Nanomedicine(s) under the microscope. Mol Pharm. 2011;58(6):2101-41. 4. Charytan DM, Pai AB, Chan CT, Coyne DW, Hung AM, Kovesdy CP, Fishbane S. Considerations and Challenges in Defining Optimal Iron Utilization in Hemodialysis. J Am Soc Nephrol. 2014 Dec 26. 5. Pai AB. Evaluating Plasma Pharmacokinetics of Intravenous Iron Formulations: Judging Books by Their Covers? Clin Pharmacokinet. 2014 Dec 2. 6. Danielson BG. Structure, chemistry, and pharmacokinetics of intravenous iron agents. J Am Soc Nephrol. 2004;Dec;15 Suppl 2:S93-8. 7. Pai AB, Nielsen JC, Kausz A, Miller P, Owen JS. Plasma pharmacokinetics of two consecutive doses of ferumoxytol in healthy subjects. Clin Pharmacol Ther. 2010;88(2):237-42. 8. Cabantchik ZI. Labile iron in cells and body fluids: physiology, pathology, and pharmacology. Front Pharmacol. 2014 Mar 13;5:45 9. Tucker PS, Dalbo VJ, Han T, et al. Clinical and research markers of oxidative stress in chronic kidney disease. Biomarkers 2013; 18: 103-115 10. Fishbane S, Mathew AT, Wanchoo R. Intravenous iron exposure and outcomes in patients on hemodialysis. Clin J Am Soc Nephrol. 2014 Nov 7;9(11):1837-9. 11. Fütterer S, Andrusenko I, Kolb U, Hofmeister W, Langguth P. Structural characterization of iron oxide/hydroxide nanoparticles in nine different parenteral drugs for the treatment of iron deficiency anaemia by electron diffraction (ED) and X-ray powder diffraction (XRPD). J Pharm Biomed Anal. 2013 Dec;86:151-60. 12. Barot BS, Parejiya PB, Mehta DM, Shelat PK, Shah GB. Physicochemical and structural characterization of iron-sucrose formulations: a comparative study. Pharm Dev Technol. 2014 Aug;19(5):513-20. 13. Toblli JE, Cao G, Oliveri L, Angerosa M. Comparison of oxidative stress and inflammation induced by different intravenous iron sucrose similar preparations in a rat model. Inflamm Allergy Drug Targets 2012;11: 66-78. 14. Elford P, Bouchard J, Jaillet L, Pearson N, Rogue A, Sabadie C, Forster R. Biodistribution and predictive hepatic gene expression of intravenous iron sucrose. J Pharmacol Toxicol Methods. 2013 Nov-Dec;68(3):374-83

15.http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2013/09/WC500149496.pdf

Ioav Cabantchik - Hebrew University of Jerusalem

Opinion regarding the use of novel oral iron formulations for the treatment of refractory anemias and chronic iron deficiencies as in CKD

Oct 27- 2019

CKD patients that are chronically iron deficient (low ion stores, low TSAT) with or without anemia (Hb< 11.5 for women, 12.5 for men) are recommended (in Western medical centers) to undergo periodic intravenous iron supplementation (IVIS), generally covered (at different levels) by national or private insurance programs. The outcomes have been positive using different iron formulations, although with new generic ones the documentation on safety (chemical quality of the product and iatrogenicity) are scanty.

An issue that has been raised is the price of the treatment (the product and IV administration in medical centers and private clinics) and patient compliance due to logistic inconvenience (time at the expense of lost work hours and income) and price (where applicable).

All previous attempts to treat iron deficiency (ID) or iron deficiency anemia (IDA) with “classical” oral iron supplements (OIS) have shown poor outcomes (due to poor intestinal absorption associated with ferroportin down-regulation, generally by increased hepcidin levels). However, recent trials with new delivery systems for oral iron formulations have changed the prospects of using OIS for iron fortification to treat absolute ID and ID concomitant with chronic inflammation in various clinical settings of (IBD, RA), CKD, pregnancy, cancer and bariatric surgery (see Gomez-Ramirez for review).

The rationale behind some iron products like sucrosomial iron (a stabilized iron-pyrophosphate carried by a phospholipid and sucrester matrix formulation) is the uptake of the encapsulated metal by intestinal M-cells and its delivery to the hematopoietic machinery via the lymphatic system-thus bypassing/circumventing the hepcidin ferroportin block (Girelli et al, 2017; Ganz et al, 2019). Although that non-canonical uptake mode has still to be further investigated, it has already shown to provide a route for iron delivery of high efficacy and safety in thousands of patients in various European countries and Israel. The perplexing fact is a published report about the positive outcome of sucrosomial iron treatment on a young IRIDA patient refractory or poorly responsive to IVIS and other ones in progress and/or in press.

The cost-efficacy of IVIS vs OIS (with sucrosomial iron) calculated for CKD or other ID-conditions might vary considerably between countries depending, among others, also on the quality of the available commercial formulations and local facilities. The

average cost of a 9-month course of OIS treatment for Israeli CKD patients designed to attain comparable outcomes (Hb, TSAT and ferritin) to those obtained with IVIS (Venofer or Ferrinject), has been estimated to be substantially (~2/3rd) lower). Taken the long-term cost-efficacy and treatment convenience, the new OIS treatment looks promising and worth seriously considering it as a treatment option in ID conditions, as CKD.

Gómez-Ramírez, S., Brilli, E., Tarantino, G., & Muñoz, M. (2018). Sucrosomial® Iron: A New Generation Iron for Improving Oral Supplementation. Pharmaceuticals (Basel, Switzerland), 11(4), 97. doi:10.3390/ph11040097

Asperti, M., Gryzik, M., Brilli, E., Castagna, A., Corbella, M., Gottardo, R., … Poli, M. (2018). Sucrosomial® Iron Supplementation in Mice: Effects on Blood Parameters, Hepcidin, and Inflammation. Nutrients, 10(10), 1349. doi:10.3390/nu10101349

Pisani et al Nephrol Dial Transplant (2015) 30: 645–652 doi: 10.1093/ndt/gfu357

Duranti et al (2018) Clin Nephrol Res 2: 33-35

Girelli, D., Ugolini, S., Busti, F., Marchi, G., Castagna, A. (2017). Modern iron replacement therapy: clinical and pathophysiological insights. International Journal of Hematology. https://doi.org/10.1007/s12185-017-2373-3

Pergola, PE., Fishbane, S., and Ganz, T., (2019). Novel Oral Iron Therapies for Iron Deficiency Anemia in Chronic Kidney Disease. Adv Chronic Kidney Dis.;26(4):272-291

Katja Stassen - F. Hoffmann-La Roche Ltd.

You may remember our discussion at ERA-EDTA where we mentioned the upcoming publication of our PASS study. I think these data will be important to address the second question for Group 4:"Are there differences among ESA preparations relative to their impact on iron parameters/needs? ***Are there differences between short- or long-acting ESAs?***"

I have attached the publication as well as supporting materials to this mail. I am also aware the Prof. Drueke was very much involved in different discussions on this topic earlier this year and I have shared the PASS data already with him in summer . During ASN last week there was also another very interesting study from Japan presented (PARAMOUNT-HD). I have attached the abstract and may be able to share the congress presentation later this week.

A second point we want to propose for consideration is the topic of artificial intelligence- or model based personalized dosing schedules (Iron and ESA in combination) to improve outcomes. We discussed this already a few times during our past meetings and we believe this could be a very interesting addition to the working groups discussion.

See end PDFs.

Bruno Riccardi - NutraGeneTech

See following proposal

KDIGO Controversies Conference on Optimal Anemia Management in CKD

I submit to the attention of KINDIGO this clinical protocol related to the subject : "Controversies Conference on optimal anemia Management in CKD ", which involves the use of an innovative delivery system in liposomal form for the administration of iron in CKD patients in total safety.

An innovative approach with the liposomal iron carriers for anemia

By Bruno Riccardi - NutraGeneTech

Introduction

"*Chronic kidney disease (CKD) is an increasingly common clinical problem that raises a patient's risk for developing several life-threatening medical conditions, including end-stage renal disease (ESRD) and cardiovascular disease (CVD). Appropriate treatment can delay or prevent these adverse outcomes. However, CKD isn't often recognized by clinicians or patients and as a result isn't often optimally treated.*" **(1)**

"Anemia is a complication of CKD that is proportional to eGFR and is independently associated with morbidity and mortality. A significant drop in hemoglobin (Hgb) is typically seen among patients with CKD G3b stage or worse.

Based on 2013 KDIGO guidelines, anemia in CKD patients is defined whit Hgb value < 13 in men and Hgb < 12 in women. Evaluation should include CBC, reticulocyte count, serum ferritin, and transferrin saturation (TSAT) to assess for iron deficiency. **(1)**

"*Malnutrition is considered to be one of the late complications of chronic renal failure. A sub-analysis of the Modification of Diet in Renal Disease (MDRD) study, however, demonstrated that progressive renal insufficiency was associated with a spontaneous decline in protein intake. Predialysed patients appeared to have a spontaneous protein intake of <0.7 g/kg/day which is below the minimal recommended daily intake. Thus, malnutrition in haemodialysis patients may already originate during stage IV of chronic renal failure.*" **(2)**

Multiple pathogenic factors have been called into question as responsible for chronic renal failure CKD, vascular, metabolic , infectious , etc., so that we should speak of chronic renal insufficiencies in the plural meaning, since each researcher wanted to highlight "***its pathogenic cause"***, the one subject of its investigation.

On this study I simply propose the treatment of ***ANEMIA , which is the common denominator of complications of ALL FORMS of chronic renal failure CKD .***In addition, ***the treatment of anemia*** is the specific subject of the Controversies Conference on ***Optimal Anemia Management*** in chronic kidney disease.

So this is the subject on which I would like offer my own contribution, not specialistic , as I am a biologist who has dealed with delivery systems technologies for many years , with a good experience in martial therapy.

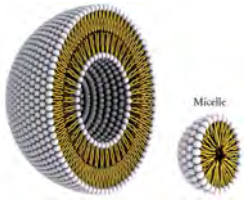
Iron supplementation is essential for the treatment of anaemia in patients with chronic renal failure (CKD). Therapeutic protocols include treatment in patients with intravenous iron CKD plus erythropoietin (EPO). This procedure, which is performed during the dialysis session in the dialysed patient, presents risks of side effects and organizational problems when administered in **the non-dialysed** CKD patient. **(3,4,5)** There is also a strong correlation between anemic status, progression of renal insufficiency and the onset of cardiovascular disease . **(6,7)**

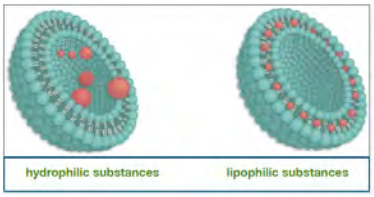
Another pathogenic factor increasingly frequently called in cause is the presence of hyperhomocysteinemia.**(8,9,10,11)**
Some authors propose homocysteine control as a prognostic factor of renal damage. **(12,13)**

Therefore an effective and complete treatment of the anaemic state in the patient with CKD renal failure not yet dialized is the one to correct the anemia and prevent the hyperhomocysteinemia , to counteract the progression of the disease. **(14)**

There are strong organizational limitations in the use of intravenous iron and important contraindications for risk of adverse effects in outpatient treatment of patients with non dialysed CKD **(3,4)**

Overcoming this problem, technological research has made available to medicine various transport systems (carriers) for drugs and active ingredients, to improve absorption and tolerability.
In the biomedical field in particular, delivery systems are intensively studied to optimize the results obtained in the diagnosis and treatment of the most common pathologies .
Among the various delivery systems used in the biomedical field are liposomes, nanospheres, nanocapsules, micelles, etc.





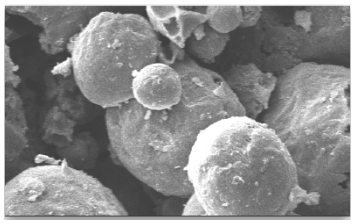
Liposomes and Micelles

A recent review of nanotechnologies and delivery systems used in the biomedical field, was the subject of a conference held in Rome in June 19th and 20th at the Istituto Superiore di Sanità and abstracs are published in "***European journal of histochemistry".*** **(15)**

Nowadays the most widespread and used technology is represented by liposomes, for the ease of their production and versatility in their use.

Liposomes are known to consist of a double layer of phospholipids with an internal cavity that can contain and transport various substances in solution such as medicines or active substances of various kinds. **(16,17,18,26)**

In 1993 was put on the market ***an innovative iron sulfate in liposomes*** , patented with the name of ***BIOFER*** by the Argentinian LIPOTECH which has a high bioavailability and overcomes all the side effects of traditional ionic iron. It is iron sulphate plus vitamin C enclosed in liposomes and dehydrated in powder.



BIOFER with SEM
Scanning Electron Microscope

The effectiveness and safety of BIOFER has been documented in numerous clinical studies and has been adopted for the fortification of various foods by major food Companies since 1994.**(19, 20,21,22,23,24)**
In 2016 the BIOFER associated with the B complex vitamins was introduced in the Italian market and *notified to the Health Italian Minister under the brand IRON-FOLIC : N° registration 87465.*
The association with BIOFER of three B-complex vitamins: Folic acid, Vitamin B6 and Vitamin B12, finds its rationale in the fact that these vitamins play an essential role in the synthesis of many substances essential for the well-being and in particular for erythropoiesis, and allows effective prevention of ***hyperhomocysteinemia*** as a recognized factor of nephropathic and cardiovascular risk.

The liposomal nature of the BIOFER and the capacity of assimilation on cell cultures in the absence of cytotoxicity has been verified by research carried out by the University of Urbino and the results have been subsequently published **(25)**

To test the effectiveness and safety of the product, we conducted a survey of Post Marketing Surveillance (PMS) on about 11,000 Italian patients who used it in the years 217 – 2018.

The results obtained in patients of different ages groups with different degrees of anaemia and for different diseases were excellent, without significant recorded side effects.
The report is being published with the title :" *IRON INTEGRATION IN ANAEMIAS AND NEW PHARMACEUTICAL TECHNOLOGIES"* in *Journal of Nutraceuticals and Food Science* .

<u>I propose testing the same product IRON-FOLIC in CKD non-dialysed patients with anemia</u> .

The advantages of this type of treatment compared to traditional ones are :

- ***Oral administration*** , which unlike that parenteral one, is easy for the patient , with home-basedtreatment and a better quality of life.
- ***The pharmaceutical form of iron sulphate in liposomes*** guarantees efficacy and safety of use;
- ***Does not require any commitment of hospital facilities for parenteral administration, as is the case for EV iron***, with the associated cost and management burden and, more importantly, does not expose the patient to the risks of intravenous iron therapy ;
- ***The treatment is more complete than martial therapy alone, as it provides , with a single administration,*** in addition to iron sulphate, also Folic acid, Vitamin B6 and Vitamin B12 essential for proper erythropoiesis and blood homocysteine control.

METHODS

Study design

The purpose of this study is to assess the effectiveness of liposomal iron treatment, *IRON-FOLIC* compared to intravenous iron (EV), in the anaemic patient with non-dialysis CKD in the presence of an iron deficiency, and at the same time demonstrate the utility in the prevention of hyperomocisteinemia.
Ours is a one-centre/multi-centre, prospective, randomised controlled, phase IV study, which may begin in 2020.

Type of Protocol : *Single or multicentre, prospective, randomised controlled, phase IV study, with informed consent of enrolled patients.*

Study intervention

A preliminary study will have to analyse 30 patients: 15 in the iron-liposomal group and 15 in the EV group. And evaluate the effectiveness and tolerability of treatment with liposomal iron os, compared to EV iron. Then the actual study will begin, as follows.
Patients with CKV in stages 3, 4 and 5 are enrolled and randomised in a 1:2 ratio **to iron EV** or iron **liposomial OS**. For Iron **EV** according to standard protocols , for liposomial Iron 30 mg die -1 capsule -for 3 months.
The primary end point is :

- **The increase in haemoglobin (Hb) from baseline**;(*)

The secondary end points are:

- **The normalisation of homocysteinemia if increased, and reduction of erythropoietin** dose by at least 25% in patients treated with erythropoiesis stimulating agents ;
- **The ferritin increase of 100 ng/ml from baseline.**

Baseline and Follow-up evaluations

<u>Criteria for inclusion in the study</u>
patients ≥ 18 years of age; with informed written consent; glomerular filtrate (GFR) ≤ 60ml/min (calculated according to MDRD 4 variables); haemoglobin 12 g/dl; ferritin 100 ng/ml or ferritin between 100 and 300 ng/ml with transferrin saturation (TSAT) ≤ 25%; if treated with epo, stable dose for at least three months. Omocisteina palasmatica > 10,5 micromol/liter .

<u>Criteria for exclusion from the study</u>
Patients with infectious diseases of any nature will be excluded; bleeding in the previous six months; history of malignancies in the last 3 years; anaemia from a cause other than that resulting from IRC; any type of surgery in the last three months; systemic haematological disease; haemorrhages, EV iron therapy or OS therapy in the last six months; severe liver disease/positivity for HCV and HBV; alcohol and drugs abuse in the previous six months; immunosuppressive therapy in progress; significant weight loss; pregnancy or lactation.

Then patients enrolled are going to be evaluated monthly for four months. No changes will be made during the study period to current therapy with Epo, ACE inhibitors, angiotensin II receptor antagonists, unless necessary. If ferritin exceeds 800 ng/ml or values are between 500 and 800 ng/ml with a 50% TSAT, martial

therapy will be discontinued for reintroduction if ferritin and TSAT values fall below 400 ng/ml and 45% respectively.

(*) The values of Hb below 12 and then also 11 g/dl represent the cut-off or threshold value, below which all national and international guidelines indicate EPO treatment.

REFERENCES

(1) Management of Chronic Kidney Disease , UMHS Chronic Kidney Disease Guideline, July 2019.

(2) Nephrol Dial Transplant (2007) 22 [Suppl 2]: ii45–ii87

(3) Acute Injury with Intravenous Iron and Concerns Regarding Long-Term Safety, Kalkidan Bishu and Rajiv Agarwal, Clin J Am Soc Nephrol 1: S19–S23, 2006

(4) Parenteral iron therapy: Beyond anaphylaxis, MUHAMMAD G. ALAM, MICHELLE W. KRAUSE,and SUDHIR V. SHAH, *Kidney International, EDITORIAL Vol. 66 (2004), pp. 457–458,*

(5) Parenteral iron nephrotoxicity: Potential mechanisms and consequences. RICHARD A. ZAGER, ALI C.M. JOHNSON, and SHERRY Y. HANSON; Kidney International, Vol. 66 (2004), pp. 144–156

(6) European Best practice Guidelines for Haemodialysis. Guideline VII.1: Assessment of Cardiovascular risk factor. Nephrol Dial Transplant 2002; 17(suppl 7): 88-109.

(7) Kidney Disease as a Risk Factor for Development of Cardiovascular Disease ; Mark J. Sarnak, MD, Cochair; Andrew S. Levey, MD, Cochair;and other ; Circulation October 28, 2003

(8) Effect of Homocysteine Lowering on Mortality and Vascular Disease in Advanced Chronic Kidney Disease and End-stage Renal Disease A Randomized Controlled Trial, Rex L. Jamison, MD and altri, JAMA, September 12, 2007—Vol 298, No. 10

(9) Homocysteine in Renal Injury; Yanjun Long, Jing Nie ; Kidney Dis 2016;2:80–87

(10) Homocysteine Studies Collaboration. Homocysteine and risk of ischemic heart disease and stroke: a meta-analysis. JAMA. 2002;288(16):2015-2022.

(11) Why is homocysteine elevated in renal failure and what can be expected from homocysteine-lowering?- Coen van Guldener , Nephrol Dial Transplant (2006) 21: 1161–1166

(12) Homocysteine as a predictive biomarker in early diagnosis of renal failure susceptibility and prognostic diagnosis for end stages renal disease , Hatem K. Amin, Mohamed-I Kotb El-Sayed & Ola F. Leheta, RENAL FAILURE, 2016 VOL. 38, NO. 8, 1267–1275

(13) The importance of homocysteine levels in the prognosis of patients with chronic renal disease and in hemodialysis patients ; Bruna M. Martella and other, J Bras Patol Med Lab. 2018 Jun; 54(3): 170-176.

(14) Wilcken DE, Wilcken B. The natural history of vascular disease in homocystinuria and the effects of treatment.; J Inherit Metab Dis. 1997;20(2):295-300.

(15) Therapeutic nanoproducts: from biology to innovative technology 19-20 June 2019 ; european journal of histochemistry a journal of functional cytology, ISSN 1121-760X volume 63/ supplement 1, 2019

(16) Liposome: classification, preparation, and applications: Abolfazl Akbarzadeh and other ; Nanoscale Research Letters 2013, 8:102

(17) LIPOSOMES – THE FUTURE OF FORMULATIONS ; Mohanty Sivasankar , and T. Katyayani, IJRPC 2011, 1(2)

(18) Liposomes ; Journal of Drug Delivery ; Copyright © 2011 Hindawi Publishing Corporation. All rights reserved.

(19) Comparison Study of Oral Iron Preparations Using a Human Intestinal Model ; Mohammed Gulrez ZARIWALA; Sci Pharm. 2013; 81: 1123–1139

(20) Bioavailability of microencapsulated ferrous-sulfate in fluid milk studies in human beings ; Gotelli CA,Gotelli MJ,Boccio JR.Zubillaga MB ,Caro RA,García del Río H,Weill R.

(21) A New Procedure to Fortify Fluid Milk and Dairy Products with High-Bioavailable Ferrous Sulfate , José R. Boccio, Marcela B. Zubillaga, Ricardo A. Caro, Ph.D., Carlos A. Gotelli, Mariano J. Gotelli, and Ricardo Weill ; Nutrition Reviews, Vol. 55, No. 6

(22) Alexis E. Lysionek et All. ; Bioavailability of Microencapsulated Ferrous Sulfate in Powdered Fortified Milk , Nutrition Volume 18, Number 3, 2002

(23) Iron availability of a fortified processed wheat cereal: a comparison of fourteen iron forms using an in vitro digestion/human colonic adenocarcinoma (CaCo-2) cell model ; Gary Wortley, Steven Leusner, Carolyn Good, Eric Gugger and Raymond Glahn , British Journal of Nutrition (2005), 93, 65–71

(24) Bioavailability study of dried microencapsulated ferrous sulfate — SFE 171 — by means of the prophylactic-preventive method *, • Alexis E. Lysionek1, Marcela B. Zubillaga1, María J. Salgueiro1, Ricardo A. Caro1,• Ricardo Weill2,José R. Boccio1 ; Journal of Trace Elements in Medicine and Biology Volume 15, Issue 4, 2001

(25) *Morphological evaluation of liposomal iron carriers ;Michela Battistelli , Sara Salucci , Elisabetta Falcieri ;Microsc Res Tech. 2018;1–6.",*

(26) Liposomes as nanomedical devices ; Giuseppina Bozzuto, Agnese Molinari, International Journal of Nanomedicine 2015:10 975–999

Abstract: TH-OR026

Prognostic Evaluation by Different Target Hemoglobin Levels During Treatment with Epoetin Beta Pegol in Hemodialysis Patients with ESA Hyporesponsiveness: PARAMOUNT-HD Study

Session Information

- Anemia and Iron Metabolism: Clinical Research
  November 07, 2019 | Location: 150, Walter E. Washington Convention Center
  Abstract Time: 05:30 PM - 05:42 PM

Category: Anemia and Iron Metabolism

- 202 Anemia and Iron Metabolism: Clinical

Authors

- Tsuchiya, Ken, Tokyo Women's Medical University, Shinjuku-ku, Japan
- Kuragano, Takahiro, Hyogo College of Medicine, Nishinomiya, Japan
- Joki, Nobuhiko, TOHO University Ohashi Medical Center, Tokyo, Japan
- Tsuruya, Kazuhiko, Nara Medical University, Kashihara, Japan
- Honda, Hirokazu, Showa University Koto Toyosu Hospital, Tokyo, Japan
- Hamano, Takayuki, Osaka University Graduate School of Medicine, Suita, OSAKA-FU, Japan
- Fujii, Hideki, Kobe University Graduate School of Medicine, Kobe, Japan
- Uemura, Yukari, Tokyo University Hospital, Tokyo, Japan
- Ohashi, Yasuo, Chuo University, Tokyo, Japan
- Nitta, Kosaku, Tokyo Women's Medical University, Shinjuku-ku, Japan

Background

The incidence of cardiovascular (CV) events is especially high in HD patients in association with hyporesponsive to erythropoiesis stimulating agents (ESAs). However, there is no recommended target ranges of hemoglobin for patients with ESA hyporesponsiveness.

Methods

We randomly assigned 304 HD, ESA-treated, patients with ESA hyporesponsiveness to a proactive treatment group (target hemoglobin [Hb] level; 11g/dL) and a maintenance treatment group (target Hb level; 9-10g/dL) by the use of epoetin beta pegol (CERA). The time from the

date of study treatment initiation to the earliest CV event was evaluated as the primary endpoint. The CV events included cardiac death, heart failure requiring hospitalization, and acute coronary syndrome requiring hospitalization. The patients were followed for 24 months.

Results

The proactive and maintenance groups had a mean baseline Hb level of 9.34 and 9.32g/dL, respectively. Mean Hb levels during the observation period were 10.58 and 10.26g/dL (p=0.001) and mean length of Hb level of over 10.5g/dL were 11.5 and 8.6 months (p=0.0002), respectively. Median doses of CERA for 6 months after study treatment were 166.7 and 150.0μg/4 weeks (p=0.298). However, there was a significant difference in frequency CERA administration (once every 4 weeks: 10.9% and 26.4%; once every 2 weeks: 86.5% and 72.3% [p=0.0006], respectively). Kaplan-Meier analysis showed a significant difference in the primary endpoint between the two groups (9 and 18 events; log-rank test, p=0.033). Cox proportional hazards analysis showed a significant lower risk of CV events in the proactive group (Hazard ratio [HR], 0.429; 95% CI; 0.193-0.955). Also, the longer length of Hb level of over 10.5g/dL was associated with lower risk of CV events (HR, 0.919 per month; 95% CI; 0.865-0.977).

Conclusion

Our results suggest that targeting Hb level of 11g/dL with CERA reduces the incidence of CV events in HD patients with ESA hyporesponsiveness. Twice-monthly administration of CERA can maintain adequate Hb levels in these patients.

Funding

- Commercial Support

Supplemental Material

Supplement to:
Locatelli F, Hannedouche T, Fishbane S, et al.
Cardiovascular Safety and All-Cause Mortality of Methoxy Polyethylene Glycol-Epoetin Beta Versus and Other Erythropoiesis-Stimulating Agents in Renal Anemia of Chronic Kidney Disease: A Randomized Trial

Contents

Supplemental Appendix 1: List of investigators

Argentina: Carlos Najun Zarazaga, Norma Garrote, Mariano Alpino, Jorge Emilio Lobo

Australia: Simon Roger, Mathew Mathew, Randall Faull, Peter Kerr, Ashley Irish, Johan Rosman, Balaji Hiremagalur, Murty Mantha, John Killen

Belgium: Kathleen Claes, Koenraad Stas, Annemie Dhondt, Raymond Vanholder, Patricia Van Der Niepen, Bart Maes

Brazil: Elias David Neto, Valter Garcia, Irene Noronha, Rachel Bregman, Roberto Flavio Pecoits Filho, Jose Luis Bevilacqua, Maria Da Graca Marabezi, Domingos Otavio D'avila, Maria Eugenia Canziani, Marcus Bastos.

Croatia: Petar Kes, Nikolina Basic Jukic, Marko Jakic, Dragan Ljutic, Sanjin Racki, Zarko Belavic, Vlasta Kupres, Dragan Klaric

Czech Republic: Sylvie Dusilova-Sulkova, Roman Safranek, Ivo Valkovsky, Vladimir Polakovic, Alena Parikova

France: Thierry Hannedouche, Pierre Bataille, Gabriel Choukroun, Christian Combe, Bruno Seigneuric, Pierre Bories, Dominique Durand, Celine Granolleras, Frederique Vecina, Bernadette Faller, Alexandre Klein, Dominique Fleury, Reynald Binaut, Vincent Lemaitre, Philippe Vigeral, Gérard Janin, Caroline Créput, Yannick Le Meur, Eric Legrand, Anne Paris, Laurent Juillard, Maurice Laville, Jean-Christophe Szelag, Walid Arkouche, Jean-Claude Aldigier, Vincent Allot

Germany: Vedat Schwenger, Wolfgang Backs, Kai Toussaint, Norbert Bockreiss, Martin Zschätzsch, Volker Wizemann, Johannes Mann, Michael Leidig, Kai-Uwe Eckardt, Joachim Leicht, Dominik Alscher, Stefan Zinn, Wolfgang Paetow, Roland Erwin Winkler, Werner Kleophas, Frank Dellanna, Markus Kroker, Peter Thon, Danilo Fliser, L. Christian Rump, Lutz Renders, Ulrich Kunzendorf, Rainer Woitas, Uwe Kraatz

Greece: Konstantinos Siamopoulos, Vassilios Vargemezis, Ploumis Passadakis, Ioannis Boletis, Dimitrios Memmos, Georgios Efstratiadis, Ioannis Stefanidis

Israel: Yehoshua Weissgarten, Leonid Feldman, Uzi Gafter, Avry Chagnac, Aaron Knecht, Nomy Levin-Iaina, Sharon Mini, Doron Schwartz.

Italy: Francesco Locatelli, Giuseppe Pontoriero, Giuseppe Villa, Ciro Esposito, Alberto Albertazzi, Gianni Cappelli, Alfredo Stefani, Sonia Pasquali, Giuseppe Cannella, Enzo Moriero, Salvatore Mandolfo, Attilio Elli, Marcello Amato, Gesualdo Campolo, Annamaria Bernardi, Fulvio Fiorini, Tecla Pati, Luigi Catizone, Alda Storari, Franco Malacarne, Luciano Cristinelli, Piergiorgio Poisetti, Roberto Scarpioni, Carlo Buzio, Salvatore David, Alessandro Zuccalà, Mario Renato Rapanà, Alberto Baraldi, Carmela Giovannone, Carmelo Cascone, Maria Cristina Maresca, Giuliano Martin, Alessandro Ciavatti, Piergianni Calzavara, Salvatore Cantaro, Agostino Naso, Claudio Minoretti, Ugo Teatini, Maurizio Gallieni, Domenico Di Landro, Massimo Matalone, Marina Di Luca, Nicola Di Daniele, Giacomo Colussi, Giorgio Giovanni Battaglia, Domenico Santoro

Republic of Korea: Yoon-Goo Kim, Kwon-Wook Joo

Lithuania: Vytautas Kuzminskis, Marius Miglinas

Malaysia: Chew Ming Wong, Soo Kun Lim, Ghazali Ahmad, Hin Seng Wong

Mexico: Tommaso Bochicchio

Panama: Rafael Perez

Philippines: Hazel Daphne Ninalga, Juliet Chua Chong-Noel

Poland: Jacek Manitius, Krzysztof Marczewski, Wieslaw Klatko, Maciej Drozdz, Robert Witek, Ewa Smolen, Waldemar Zelias, Bogna Roszkowiak, Zdzislaw Gozdzik, Dorota Radziszewska, Wiesława Kiszka, Marzena Janas

Russian Federation: Elena Kolmakova, Alexey Smirnov, Konstantin Gurevich, Olga Solovyeva, Alexander Zemtchenkov

Serbia: Nada Dimkovic, Steva Pljesa, Gordana Pekovic, Rodoljub Markovic, Milan Radovic, Vidosava Nesic, Vidojko Djordjevic, Miomir Stojanovic, Igor Mitic

Singapore: Hui Lin Choong, Titus Lau, Adrian Liew, Tsun Gun Ng

Spain: Javier Arrieta Lezama, Jesus Montenegro Martinez, Jose Ignacio Cornago, Saioa Bilbao, Ramon Ruiz De Gauna Lopez Heredia, Jesus Arteaga Coloma, Milagros Fernandez Lucas, Maria Dolores Del Pino Y Pino, M. Dolores Prados Garrido, M. José Garcia Cortes, Juan Carlos Herrero Berron, M. Luisa Muñiz Gomez, Fernando De Alvaro Moreno, Antonio Cirugeda

Sweden: Bjoern Wikstroem, Hans Furuland, Stefan Jacobson, Lilian Zezina

Taiwan: Jin-Bor Chen, Kuan-Yu Hung

Thailand: Dusit Lumlertgul, Worapon Buranachokpaisan, Kearkiat Praditpornsilpa

Turkey: Siren Sezer, Idris Sahin, Saime Paydas

United Kingdom: David Wheeler, Andrew Davenport, Sunil Bhandari, Richard D'souza, Kevin Harris, Jonathan Barratt, A. Peter Maxwell, Ashraf Mikhail, Steve Riley, Russell Roberts, Chris Winearls, Mona Wahba, David Makanjuola, Rebecca Suckling, Rob Parry, Jo Taylor, Alastair Woodman, Harish Shetty, Richard Baker, Neil Sheerin, Lui Forni, Patrick Carr, David Reaich, Andrew Paterson, Kevin Eardley, Philip Kalra, Daniel Zehnder, Waqar Ayub, Wai Tse, Gerald Glancey

MIRCERA PASS Data Safety Monitoring Committee

Diethelm Messenger, Ist Gmbh, Mannheim, Germany; Lars Weiss, Njurmedicinkliniken Centralsjukhuset, Karlstadt, Sweden; Nathan Levin, Renal Research Institute, New York, NY, USA; Tilman Drueke, INSERM, Paris, France.

MIRCERA PASS End Point Adjudication Committee

William B White (Chair), University of Connecticut School of Medicine, Farmington, CT, USA; Peter Kowey, Jefferson Medical College, Philadelphia, PA, USA; Philip Gorelick, University of Illinois College Of Medicine, Chicago, IL, USA; Steven Fishbane, Winthrop University Hospital, Mineola, NY, USA.

Supplemental Appendix 2: Cardiovascular end point definitions

A. Primary – APTC/MACE Fatal Events

i. Death due to myocardial infarction: the presence of two of the three following criteria: a) chest pain consistent with angina, b) any abnormal value of cardiac biomarkers (MB fraction of creatine phosphokinase and/or troponin I or T), c) myocardial injury current or the development of Q waves in two contiguous leads of the electrocardiogram. If the clinical diagnosis of myocardial infarction is not possible, autopsy findings with an unequivocal diagnosis of myocardial infarction may be used to confirm the diagnosis.

ii. Death due to stroke: ischemic or hemorrhagic stroke defined as an acute, focal neurological event that occurred within 30 days of death. Confirmation by imaging studies (magnetic resonance imaging or computerised tomography of the brain) or autopsy data will be sought in all cases, but will not be required for adjudication of the event.

iii. Cardiovascular Deaths: deaths that were sudden or unexplained without documentation of myocardial infarction or stroke as follows: Sudden arrhythmic death (observed to have had an arrhythmia)

    i. Sudden death (etiology unspecified)

    ii. Other cardiovascular death: death without documentation of myocardial infarction (A.i) or stroke (A.ii) or that is exclusive of the diagnoses listed in Sections A.iii.i. and A.iii.ii.

B. Noncardiovascular deaths

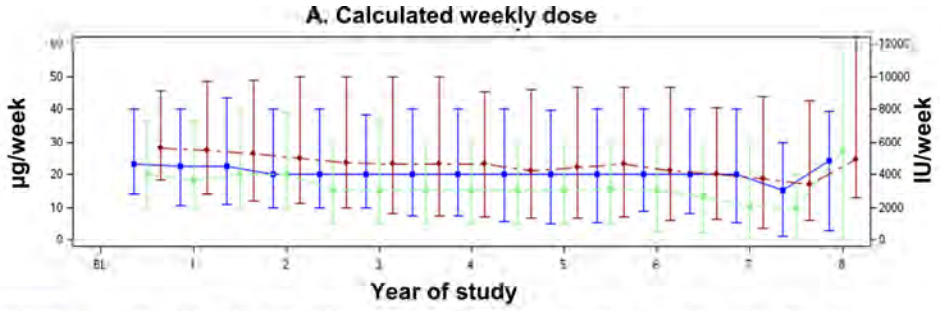
i. Deaths that are exclusive of the diagnoses listed in Section A

C. Death due to discontinuation of dialysis (voluntary)

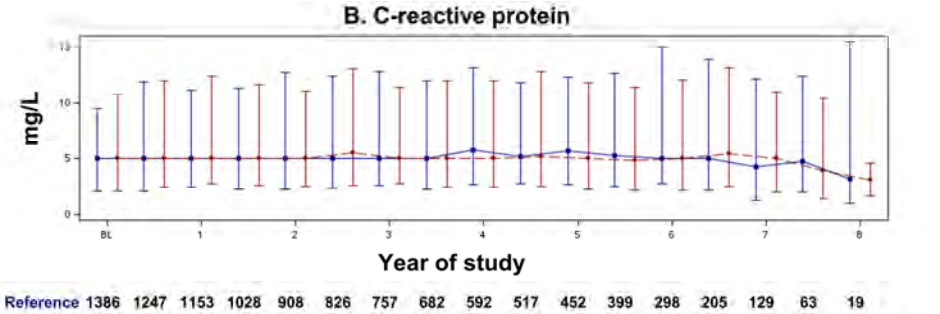
D. Nonfatal Stroke: ischemic or hemorrhagic stroke defined as an acute, focal neurological event that persisted for >24 h. Confirmation by imaging studies (magnetic resonance imaging or computerised tomography of the brain) will be sought in all cases, but will not be required for adjudication of the event. The diagnosis of stroke will be made when an imaging study clearly demonstrates brain injury (ischemic or non-ischemic), despite symptoms resolving in <24 h.

E. Nonfatal myocardial infarction: the presence of two of the three following criteria: a) chest pain consistent with angina, b) any abnormal value of cardiac biomarkers (MB fraction of creatine phosphokinase and/or troponin I or T), c) myocardial injury current or the development of Q waves in two contiguous leads of the electrocardiogram.

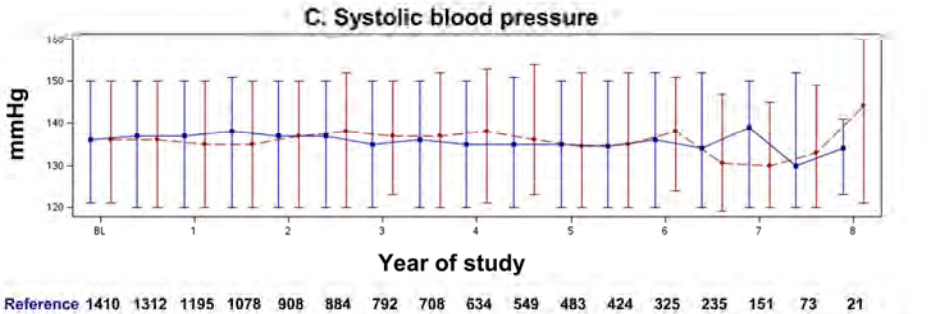
Supplemental Figure 1: **A. Doses of erythropoiesis-stimulating agents during the study. B. C-reactive protein levels during the study. C. Systolic blood pressure during the study. Values shown are median with interquartile ranges**



Epoetin alfa/beta	794	719	652	588	535	491	445	383	334	300	263	204	147	101	49	16
[illegible]	517	414	445	398	354	316	285	256	230	200	175	131	83	50	25	5
Methoxy polyethylene glycol-epoetin beta	1292	1145	1036	936	834	741	663	600	536	478	423	311	229	161	86	28



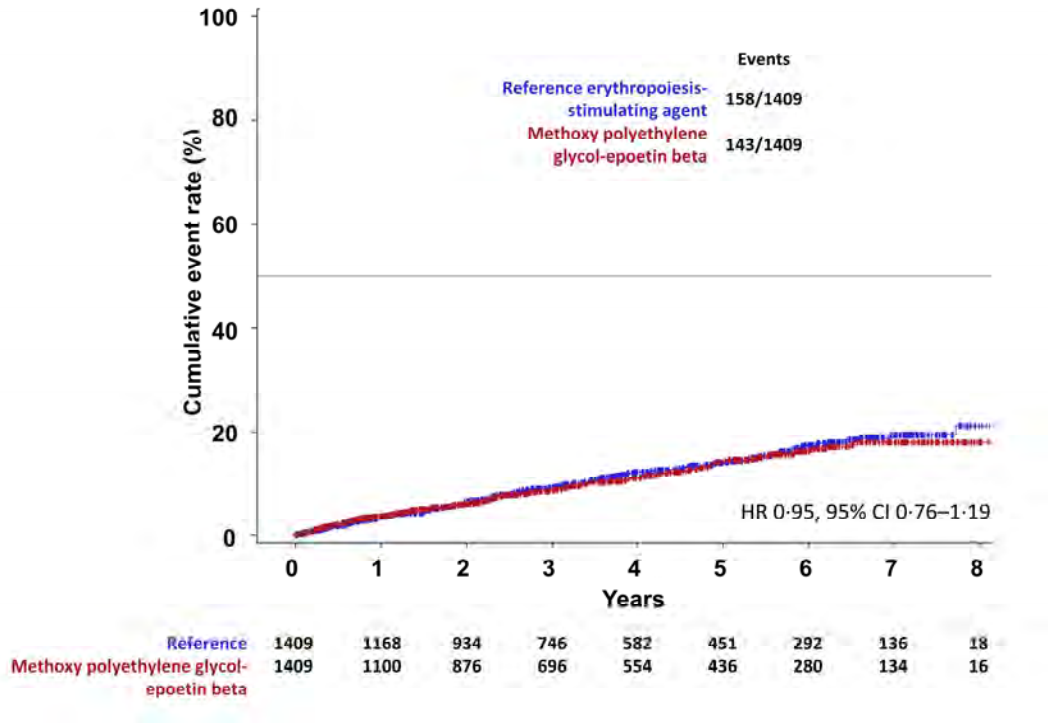
Reference	1386	1247	1153	1028	908	826	757	682	592	517	452	399	298	205	129	63	19
Methoxy polyethylene glycol-epoetin beta	1379	1190	1096	963	866	760	688	611	554	496	447	394	294	207	146	77	17



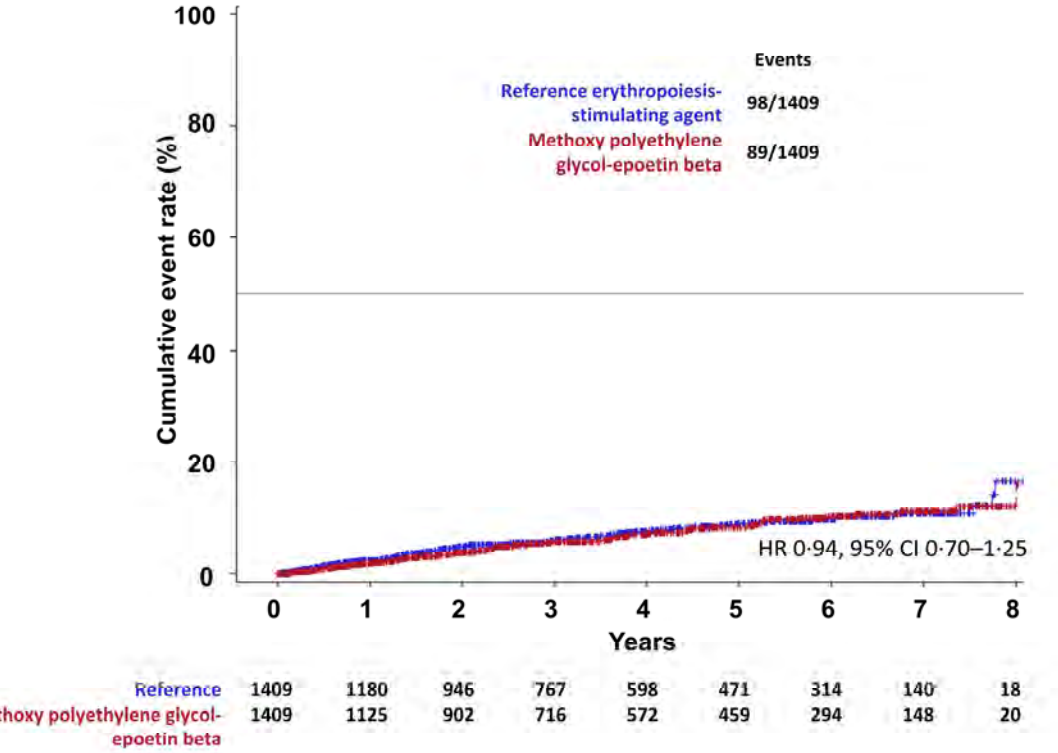
Reference	1410	1312	1195	1078	908	884	792	708	634	549	483	424	325	235	151	73	21
Methoxy polyethylene glycol-epoetin beta	1379	1190	1096	1026	924	819	727	644	590	525	466	407	301	226	154	89	26

Supplemental Figure 2: **Time-to-event curves for A. Time to myocardial infarction and B. Time to stroke**

A

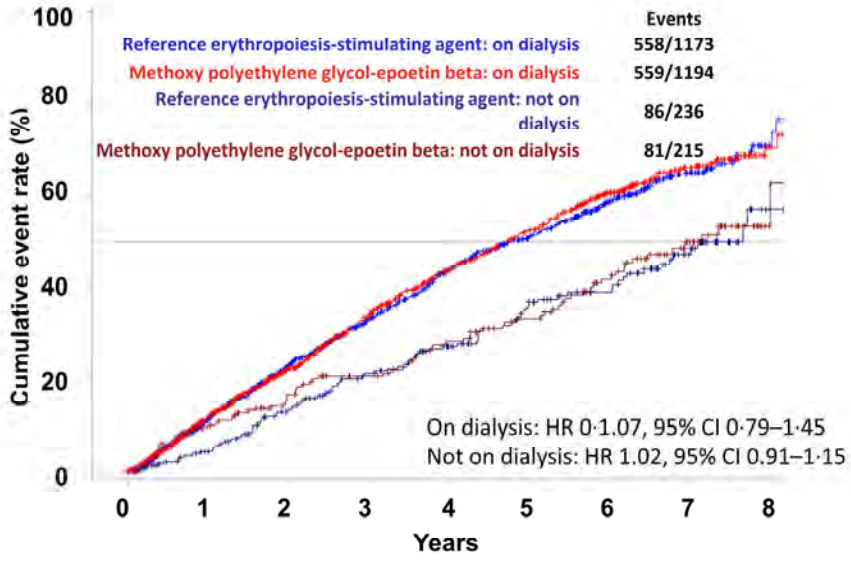


B



~`

***Supplemental Figure 3*: Time-to-event curves for death from any cause, nonfatal stroke or nonfatal myocardial infarction, for patients on dialysis or not on dialysis**



On dialysis									
Reference	1173	944	729	567	431	335	206	91	10
Methoxy polyethylene glycol-epoetin beta	1194	918	716	553	429	333	200	94	11
Not on dialysis									
Reference	236	207	175	147	121	94	76	39	7
Methoxy polyethylene glycol-epoetin beta	215	173	145	128	109	91	67	35	4

Supplemental Table 1: **Inclusion and exclusion criteria**

Inclusion criteria	Exclusion criteria
Written informed consent	Uncontrolled hypertension
Adult patients (≥18 years old) with symptomatic anemia associated with chronic kidney disease (renal anemia)	Hypersensitivity to the active substance or any of the excipients of methoxy polyethylene glycol-epoetin beta and other ESAs
Patients with renal anemia who are not treated with an ESA:	Any other contraindication to ESA therapy

Anemia defined as hemoglobin concentration <11 g/dL (mean of two screening values with at least 1 day and a maximum of 2 weeks between measurements) with a clinical indication for ESA treatment	Conditions known to cause inadequate response to ESA treatment or anemia other than symptomatic anemia associated with chronic kidney disease, including:
	• Hemoglobinopathies (e.g., homozygous sickle-cell disease, thalassemia of all types)
	• Anemia due to hemolysis
	• Pure red cell aplasia
	• Other:
	  • High likelihood of early withdrawal (e.g. within 1 year) or interruption of the study
	  • Pregnancy or breast-feeding
	Women of childbearing potential without effective contraception
	  • Administration of another investigational drug within 1 month before screening or planned during the study period

or	
Patients with renal anemia who are on maintenance ESA therapy	
If on dialysis: regular long-term hemodialysis or peritoneal dialysis therapy with the same mode of dialysis for at least 3 months before screening	
Continuous intravenous or subcutaneous maintenance ESA therapy: darbepoetin afa (Aranesp®, Nespo®, Aranest®), epoetin alfa (Eprex®, Epogen®, Epopen®, Erypo®) or epoetin beta (NeoRecormon®, Recormon®) administered according to approved label of the same agent and route of administration for at least 2 months before screening	
Hemoglobin concentration between 10 and 12 g/dL (mean of two screening values with at least 1 day and a maximum of 2 weeks between measurements)	
Patients with adequate iron status defined as: serum ferritin above or equal to 100 ng/mL or transferrin saturation above or equal to 20%	

ESA=erythropoiesis-stimulating agent.

Supplemental Table 2: **Starting dose of methoxy polyethylene glycol-epoetin beta according to previous ESA treatment when switching from another ESA**

Previous darbepoetin alfa dose (μg/week)	Previous epoetin dose (IU/week)	Methoxy polyethylene glycol-epoetin beta dose (μg/once monthly)
<40	<8000	120
40 –80	8000–16000	200
>80	>16000	360

ESA=erythropoiesis-stimulating agent; IU=international unit.

Supplemental Table 3: **Schedule of assessments**

The following laboratory assessments were to be performed during the randomized treatment period:

Test	Frequency
Hemoglobin	At study visit 1, at monthly visits and at the final visit. In correction patients' hemoglobin was assessed twice monthly until stabilized.
Platelet count	At study visit 1 and at monthly visits.
Total white blood cell count	At study visit 1 and every 3 months
C-reactive protein	Every 3 months
Serum albumin, calcium, phosphorus, potassium	At study visit 1 and every 3 months
Serum ferritin, serum iron, serum transferrin or total iron-binding capacity, transferrin or percentage of hypochromic red blood cells	Every 3 months
Cholesterol, triglycerides, HbA_{1c}, glucose	At study visit 1 and once a year
Anti-erythropoietin antibody determination	At study visit 1, once a year and at the final visit
Dialysis quantification indexes and renal function: • Patients on hemodialysis: Kt/V or urea reduction ratio • Patients on peritoneal dialysis: the weekly Kt/V • Patients not on dialysis: serum creatinine, creatinine clearance/estimated GFR	• At study visit 1 and every 6 months
12-lead ECG recording	At study visit 1 before dose administration.

Supplemental Table 4: **Additional baseline characteristics**

	Reference ESA	Methoxy polyethylene glycol-epoetin beta
Pre-dialysis systolic blood pressure (mmHg), n (%)	n=1091	n=1104
<140	569 (52)	597 (54)
140–160	359 (33)	384 (35)
>160	163 (15)	123 (11)
HbA_{1c} (%)	n=1221	n=1248
Mean±SD	5.8±3.3	5.9±4.1
Median (IQR)	5.6 (5.1–6.3)	5.6 (5.1 – 6.3)
Albumin (g/dL)	n=1308	n=1318
Categories, n (%)		
<3.5	261 (20)	261 (20)
3.5–4.0	583 (45)	607 (46)
>4.0	464 (35)	450 (34)
Triglycerides [mmol/L]	n=1291	n=1320
Mean ±SD	1.96±3.61	1.91±1.29
Median (IQR)	1.59 (1.10–2.30)	1.58 (1.10–2.33)
HD Vascular Access, n (%)	n=1092	n=1103
Arteriovenous fistula	889 (81)	900 (82)
Arteriovenous graft	61 (6)	56 (5)
Central venous catheter	142 (13)	147 (13)
eGFR (mL/min/1.73m^2)	n=229	n=207
Categories, n (%)		
<30	178 (78)	171 (83)
30–44	39 (17)	28 (14)
≥45	12 (5)	8 (4)
Smoking Status at Screening, n (%)	n=1406	n=1409
Smoker	156 (11)	137 (10)
Non-Smoker	1250 (89)	1272 (90)

ESA=erythropoiesis-stimulating agent; HbA_{1c}=hemoglobin A_{1c}; eGFR=estimated glomerular filtration rate; HD=hemodialysis; IQR=interquartile range; SD=standard deviation.

Supplemental Table 5. **Concomitant iron supplementation**

	Reference ESA	Methoxy polyethylene glycol-epoetin beta
Patients with at least 1 treatment during the study, n (%)	1315 (93)	1289 (92)
Top treatments:		
Iron sucrose	666 (47)	661(47)
Ferrous gluconate	428 (30)	390 (28)
Iron polymaltose	99 (7)	111 (8)
Ferrous sulfate	101 (7)	101 (7)
Iron dextran	85 (6)	102 (7)
Route		
IV or IV infusion	1244 (85)	1212 (85)
Oral	211 (14)	208 (15)

Supplemental Table 6: **Selected baseline parameters by withdrawal in the first year**

	Censored in first year		Remained on study past 1 year	
Parameter, mean (SD) unless stated	Reference ESA (*n*=129)	Methoxy polyethylene glycol-epoetin beta (*n*=172)	Reference ESA (*n*=1148)	Methoxy polyethylene glycol-epoetin beta (*n*=1090)
Hemoglobin, g/dL	10.8 (1.02)	10.88 (0.93)	10.76 (1.03)	10.75 (0.99)
Transferrin saturation, %	27 (13)	29 (15)	31 (23)	30 (20)
Ferritin, ng/mL	464 (561)	497 (353)	488 (391)	481 (415)
ESA type at screening, n (%)	*n* = 107	*n* = 148	*n* = 910	*n* = 865
Darbepoetin alfa	52 (49)	51 (34)	397 (44)	373 (43)
Epoetin alfa	23 (21)	46 (31)	182 (20)	193 (22)
Epoetin beta	32 (30)	51 (34)	331 (36)	299 (35)
Dialysis at screening	*n* = 129	*n* = 172	*n* = 1148	*n* = 1090
Hemodialysis	99 (77)	138 (80)	875 (76)	847 (78)
Peritoneal dialysis	9 (7)	12 (7)	67 (6)	70 (6)
None	21 (16)	22 (13)	206 (18)	173 (16)

Supplemental Table 7: **Adjudicated causes of death**

	Reference ESA n=1409	Methoxy polyethylene glycol-epoetin beta n=1409
All deaths	**557**	**558**
Death due to myocardial infarction	37	37
Death due to stroke	26	28
Other cardiovascular death	53	39
Death sudden due to arrhythmia	14	21
Death sudden etiology unknown	136	185
Noncardiovascular death	264	223
Death due to discontinuation of dialysis	27	25

ESA=erythropoiesis-stimulating agent.

Supplemental Table 8: **Time-dependent Cox regression models for hemoglobin level and dose prior to the event: Safety population, without baseline factors included (individual covariates Cox model) or with baseline factors included (multivariable Cox model)**

INDIVIDUAL COVARIATES COX MODEL*		
Mean hemoglobin concentration in 3 months before event (*vs* reference 10–11 g/dL)		
g/dL	HR (95% CI)	p value[†]
<10	2.76 (2.41–3.17)	<0.001
11–<12	0.72 (0.62–0.83)	<0.001
≥12	0.66 (0.53–0.81)	<0.001
Mean ESA dose quartile in 3 months prior to event (*vs* reference first quartile)		
	HR (95% CI)	p value
Second quartile	1.26 (1.06–1.48)	0.007
Third quartile	1.37 (1.16–1.62)	<0.001
Fourth quartile	2.44 (2.10–2.83)	<0.001
MULTIVARIABLE COX MODEL[‡]		
Mean hemoglobin concentration in 3 months before event (*vs* reference 10–11 g/dL)		
g/dL	HR (95% CI)	p value
<10	2.79 (2.43; 3.21)	<0.001
11–<12	0.71 (0.61; 0.82)	<0.001
≥12	0.68 (0.55; 0.83)	<0.001
Mean ESA dose quartile in 3 months prior to event (*vs* reference first quartile)		
	HR (95% CI)	p value
Second quartile	1.28 (1.08–1.51)	0.004
Third quartile	1.39 (1.17–1.65)	<0.001
Fourth quartile	2.52 (2.16–2.94)	<0.001

* Model containing on treatment hemoglobin or dose categories and treatment only.

[†] p value for a difference in the hazard ratio from 1; p values are exploratory and for illustration only.

[‡] Model containing hemoglobin or dose categories, treatment, and baseline factors: age, body mass index, sex, region, dialysis treatment, presence of risk factors and treatment setting.

Supplemental Table 9: **Adverse events**

MedDRA System Organ Class MedDRA Preferred Term Patients, n (%)	**Reference n=1409**	**Methoxy polyethylene glycol-epoetin beta n=1409**
Any adverse event		
Total number of patients with at least one adverse event	1340 (95)	1351 (96)
Overall total number of events	21270	20549
Adverse events experienced by ≥5% of patients		
Infections and infestations		
Pneumonia	239 (17)	243 (17)
Urinary tract infection	217 (15)	217 (15)
Bronchitis	208 (15)	201 (14)
Nasopharyngitis	171 (12)	187 (13)
Upper respiratory tract infection	160 (11)	138 (10)
Gastroenteritis	115 (8)	134 (10)
Sepsis	92 (7)	82 (6)
Respiratory tract infection	92 (7)	77 (5)
Influenza	87 (6)	75 (5)
Lower respiratory tract infection	70 (5)	84 (6)
Device related infection	82 (6)	62 (4)
Peritonitis	69 (5)	73 (5)
Gastrointestinal disorders		
Diarrhea	274 (19)	280 (20)
Constipation	179 (13)	149 (11)
Vomiting	149 (11)	124 (9)
Abdominal pain	102 (7)	111 (8)
Nausea	99 (7)	104 (7)
Abdominal pain upper	76 (5)	85 (6)
Dyspepsia	81 (6)	60 (4)
Vascular disorders		
Hypertension	446 (32)	462 (33)
Hypotension	156 (11)	136 (10)
Musculoskeletal and connective tissue disorders		
Muscle spasms	211 (15)	199 (14)
Back pain	166 (12)	120 (9)
Pain in extremity	133 (9)	125 (9)
Arthralgia	116 (8)	125 (9)
Osteoarthritis	119 (8)	99 (7)
Musculoskeletal pain	79 (6)	72 (5)
Injury, poisoning and procedural complications		
Procedural hypotension	231 (16)	236 (17)
Arteriovenous fistula site complication	209 (15)	187 (13)
Arteriovenous fistula thrombosis	178 (13)	151 (11)
Arteriovenous fistula site hemorrhage	79 (6)	88 (6)
Metabolism and nutrition disorders		
Fluid overload	168 (12)	144 (10)
Hyperkalemia	154 (11)	147 (10)
Hyperphosphatemia	142 (10)	136 (10)
Respiratory, thoracic and mediastinal disorders		
Cough	194 (14)	163 (12)
Dyspnea	120 (9)	122 (9)
General disorders and administration site conditions		
Pyrexia	119 (8)	111 (8)
Eedema due to renal disease	81 (6)	80 (6)
Asthenia	64 (5)	87 (6)
Cardiac disorders		
Atrial fibrillation	166 (12)	151 (11)
Angina pectoris	79 (6)	71 (5)
Nervous system disorders		
Headache	130 (9)	144 (10)
Dizziness	82 (6)	89 (6)
Blood and lymphatic system disorders		
Anemia	158 (11)	195 (14)
Psychiatric disorders		
Insomnia	116 (8)	104 (7)
Depression	67 (5)	77 (5)
Endocrine disorders		
Hyperparathyroidism secondary	167 (12)	169 (12)
Skin and subcutaneous tissue disorders		
Pruritus	118 (8)	114 (8)
Eye disorders		
Cataract	81 (6)	84 (6)

MedDRA, Medical Dictionary for Regulatory Activities.

Cardiovascular Safety and All-Cause Mortality of Methoxy Polyethylene Glycol-Epoetin Beta and Other Erythropoiesis-Stimulating Agents in Anemia of CKD: A Randomized Noninferiority Trial

Francesco Locatelli,[1] Thierry Hannedouche,[2] Steven Fishbane,[3] Zoe Morgan,[4] Delphine Oguey,[5] and William B. White[6]

[1]Department of Nephrology, Alessandro Manzoni Hospital, Azienda Socio Sanitaria Territoriale di Lecco, Lecco, Italy; [2]School of Medicine, University of Strasbourg, Strasbourg, France; [3]Department of Medicine, Zucker School of Medicine at Hofstra/Northwell, Great Neck, New York; [4]Biostatistics, F. Hoffmann-La Roche Ltd., Basel, Switzerland; [5]Clinical Science, F. Hoffmann-La Roche Ltd., Basel, Switzerland; and [6]Calhoun Cardiology Center, University of Connecticut School of Medicine, Farmington, Connecticut

Correspondence: Dr. Francesco Locatelli, Department of Nephrology, Alessandro Manzoni Hospital, Azienda Socio Sanitaria Territoriale di Lecco, via dell'Eremo 9/11, 23900 Lecco, Italy. Email: f.locatelli@asst-lecco.it

Abstract

Background and objectives Erythropoiesis-stimulating agents correct anemia of CKD but may increase cardiovascular risk. We compared cardiovascular outcomes and all-cause mortality associated with monthly methoxy polyethylene glycol-epoetin beta with those of the shorter-acting agents epoetin alfa/beta and darbepoetin alfa in patients with anemia of CKD.

Design, setting, participants, & measurements We conducted a multicenter, open-label, noninferiority trial in which patients were randomized to receive methoxy polyethylene glycol-epoetin beta or reference erythropoiesis-stimulating agents, stratified by maintenance or correction treatment status and C-reactive protein level. The trial had a prespecified noninferiority margin of 1.20 for the hazard ratio (HR) for the primary end point (a composite of all-cause mortality, nonfatal myocardial infarction or stroke, adjudicated by an independent blinded committee). This trial is registered with ClinicalTrials.gov, number NCT00773513.

Results In total, 2818 patients underwent randomization, received methoxy polyethylene glycol-epoetin beta or a reference agent, and were followed for a median of 3.4 years (maximum, 8.4 years). In the modified intention-to-treat analysis, a primary end point event occurred in 640 (45.4%) patients in the methoxy polyethylene glycol-epoetin beta arm, and 644 (45.7%) in the reference arm (HR 1.03; 95% confidence interval [95% CI], 0.93 to 1.15, P=0.004 for noninferiority). All-cause mortality was not different between treatment groups (HR 1.06; 95% CI, 0.94 to 1.19). Results in patient subgroups on dialysis or treated in the correction or maintenance settings were comparable to the primary analysis.

Conclusions In patients with anemia of CKD, once-monthly methoxy polyethylene glycol-epoetin beta was noninferior to conventional, shorter-acting erythropoiesis-stimulating agents with respect to rates of major adverse cardiovascular events or all-cause mortality.

CJASN 14: ●●●–●●●, 2019. doi: https://doi.org/10.2215/CJN.01380219

Introduction

Anemia is a common complication of advanced CKD (1,2). Up to 80% of patients with ESKD have significant anemia associated with relative erythropoietin insufficiency and/or absolute or functional iron deficiency (2). Severe anemia in CKD increases the need for transfusions, significantly impairs quality of life, and is associated with higher cardiovascular risk and shorter survival (1,3–5).

Recombinant human erythropoietins epoetin alfa and beta have been used since 1989 for treatment of anemia in patients with CKD, with dosing up to three times per week. Targeting hemoglobin levels of >13 g/dl does not improve outcomes in patients with anemia of CKD (6–9) and has been shown to correlate with increased cardiovascular and thrombosis risk (7–9), particularly in patients who require higher doses of erythropoiesis-stimulating agents (ESAs) (9,10). Hence, current guidelines do not recommend normalization of hemoglobin levels in patients with anemia of CKD (2,11).

The $t_{1/2}$ of epoetins has been extended through modification of the carbohydrate moiety (darbepoetin alfa) or attachment of a large methoxy polyethylene glycol polymer chain. These modifications permit less frequent administration schedules of every 1–2 weeks for darbepoetin alfa (or every 4 weeks in nondialysis patients) or monthly for methoxy polyethylene glycol-epoetin beta, reducing treatment burden for patients and health care providers.

In clinical development studies of methoxy polyethylene glycol-epoetin beta, no safety issues unexpected for ESAs were identified. In a pooled analysis of 11 studies, five related cardiac events were identified in 1789 patients treated with methoxy polyethylene glycol-epoetin beta compared with none in

948 patients treated with reference ESAs (12). In the context of uncertainty regarding the cardiovascular safety of ESAs, particularly when targeting higher hemoglobin levels, the US Food and Drug Administration and the European Medicines Agency mandated a postapproval safety study (MIRCERA PASS) of methoxy polyethylene glycol-epoetin beta as part of the risk management plan for the drug. A noninferiority design, used in many trials of this type, was agreed to be appropriate in this setting. The MIRCERA PASS trial was conducted to determine whether methoxy polyethylene glycol-epoetin beta was noninferior to the reference agents epoetin alfa/beta and darbepoetin alfa with regard to all-cause mortality and cardiovascular morbidity when targeting hemoglobin levels of 10–12 g/dl in patients with anemia and CKD.

Materials and Methods

Study Design

We conducted a multicenter, randomized, open-label, noninferiority trial. The funder (F. Hoffmann-La Roche Ltd.) was responsible for the trial design (in consultation with the regulatory authorities in the United States and Europe), conduct, monitoring, data collection, storage, and analysis of the final results. An independent data safety and monitoring committee monitored the trial and had access to the unblinded data. Statistical analyses were performed for the committee by an independent statistical group (International Drug Development Institute, Louvain-la-Neuve, Belgium).

The academic authors of the present article drafted the manuscript, had full access to the final trial data, and vouch for the accuracy and completeness of the data and the analyses, as well as for the fidelity of the trial to the protocol. The study was conducted in accordance with the Declaration of Helsinki and the appropriate national and institutional regulatory authorities and ethics committees approved the trial design.

Patients

Patients were eligible for enrollment in the trial if they had CKD (on dialysis or not on dialysis), with anemia defined according to guidelines at the time of study initiation in 2008 (13). Additional criteria for inclusion were a hemoglobin concentration $<$11.0 g/dl in the correction setting and between 10 and 12 g/dl for patients on maintenance treatment and adequate iron status (serum ferritin $\geq$100 ng/ml, or transferrin saturation $\geq$20%). Detailed inclusion and exclusion criteria are provided in Supplemental Table 1. All participants provided written informed consent before the initiation of any study-related procedures.

Randomization and Masking

Randomization was performed centrally by an independent Interactive voice/web Response System (IxRS) provider, following a pre-established randomization list of randomly permuted blocks of size 4 (balanced allocation: 1:1). Randomization was stratified by treatment setting (correction/maintenance) and baseline C-reactive protein category ($\leq$30 or $>$30 mg/L), with 20% of patients specified in the protocol to be in the correction setting at randomization. The study was open label, with both patients and physicians aware of treatment assignment. End points were assessed by an independent adjudication committee whose members were unaware of treatment assignment.

Procedures

Patients were randomly assigned to receive methoxy polyethylene glycol-epoetin beta or a reference agent (epoetin alfa/beta or darbepoetin).

The initial methoxy polyethylene glycol-epoetin beta dose was 0.6 μg/kg every 2 weeks in the correction setting. The dosing interval was changed to once monthly when the target hemoglobin concentration of 10–12 g/dl was achieved. For those in the maintenance setting, patients were switched to methoxy polyethylene glycol-epoetin beta administered at monthly intervals with a starting dose of 120, 200, or 360 μg, determined by the previous weekly ESA dose (see Supplemental Table 2). Patients randomized to the reference group started treatment according to the label if untreated or continued on the same regimen per the label if on a maintenance dose.

Hemoglobin concentrations were assessed monthly in patients on maintenance treatment, and every 2 weeks in previously untreated patients until target levels of 10–12 g/dl were reached, and monthly thereafter. Iron status was monitored every 3 months and supplementation was administered as required, orally or intravenously, to maintain serum ferritin $\geq$100 ng/ml and transferrin saturation $\geq$20%, according to the practice standard of the clinic investigator. Visits occurred monthly throughout the study after randomization and included vital signs, hemoglobin, and platelet counts; serum biochemical testing was conducted according to the schedule in Supplemental Table 3.

Outcomes

The primary composite end point was the time to the first occurrence of death, nonfatal myocardial infarction, or nonfatal stroke (definitions are provided in Supplemental Appendix 1). Secondary end points comprised the individual components of the primary end point. The consistency of effects on the primary end point was explored in a variety of subgroups. Additional safety end points were the occurrence of pure red cell aplasia, thromboembolic events, and gastrointestinal bleeding. The independent central end points committee, the members of which were unaware of the treatment assignment, adjudicated all suspected end point events.

Statistical Analyses

Cox proportional hazards models were used to analyze the time to the first occurrence of primary and secondary end points for all patients who underwent randomization and received treatment. Patients undergoing kidney transplantation were withdrawn from the study and censored at that time if no end points had occurred. A determination of noninferiority of methoxy polyethylene glycol-epoetin beta to the reference ESAs required that the upper bound of the two-sided 95% confidence interval (95% CI) of the

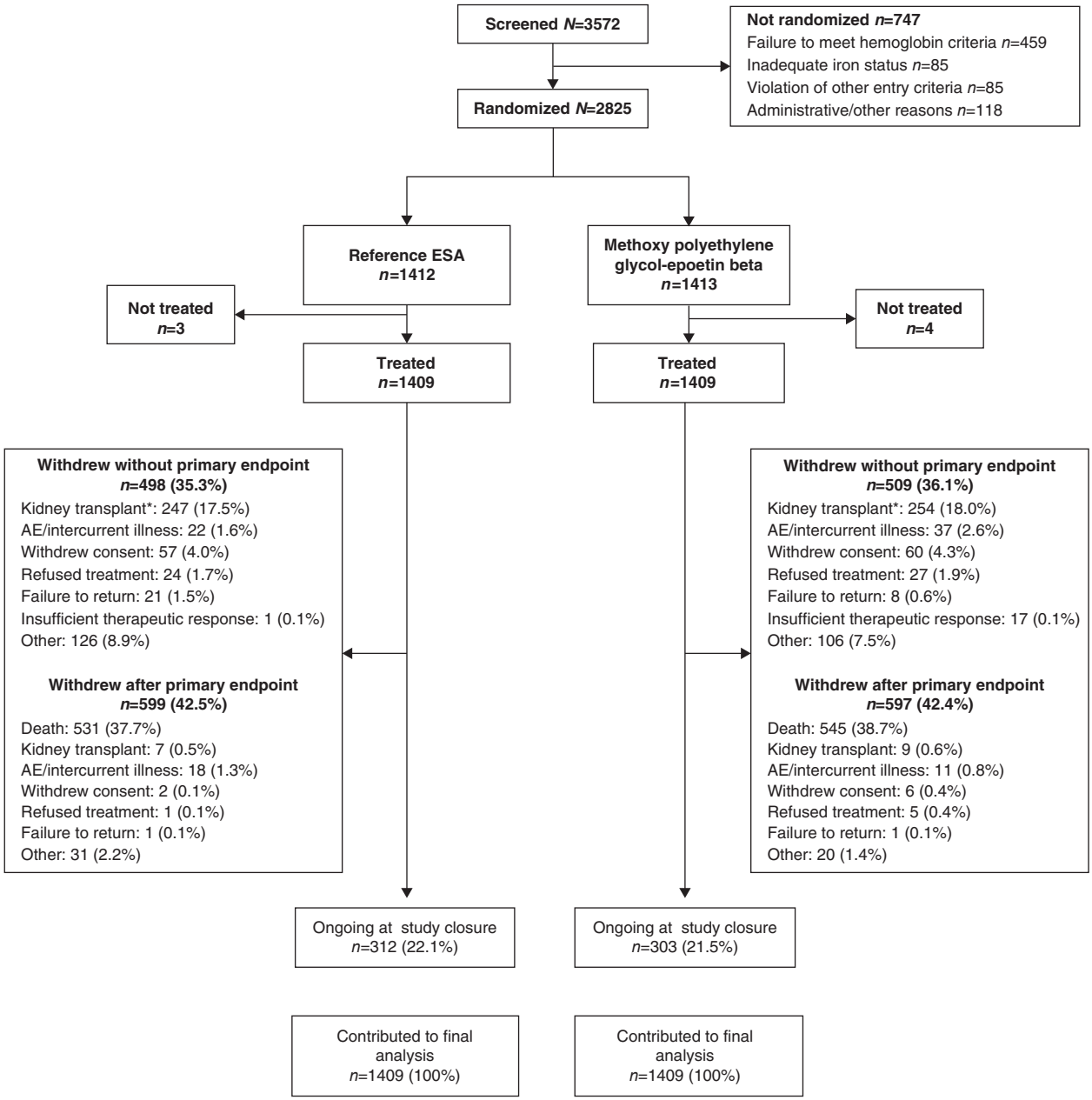


Figure 1. | **Patient disposition.** The most common reason for discontinuation in patients in the 'other' category was the patient moving away from participating dialysis units. *Kidney transplant was a prespecified reason for withdrawal from the trial.

hazard ratio (HR) for the primary end point be <1.20. The number and percentage of patients with a primary end point event were tabulated for predefined subgroups. The HR (methoxy polyethylene glycol-epoetin beta versus the reference ESAs) was calculated within each subgroup. Exploratory analyses using Cox models including time-dependent variables for hemoglobin and dose, averaged over the preceding 3 months before end point events, were performed. Analyses were performed using SAS version 9.4 (SAS Institute Inc., Cary, NC).

The trial was designed to accrue 1264 primary end point events for assessing the noninferiority criteria under the assumption of a true HR of 1.0 and 90% power. On the basis of a projected 20% composite event rate, assuming linear recruitment over 30 months and a follow-up time of 18 months after randomization of the last patient, a sample size of 2800 patients, representing approximately 7700 patient-years of exposure, was estimated to be adequate. A lower than expected event rate required a protocol amendment in 2015 to extend the anticipated study duration from an expected 4 years to 8–10 years (Amendment D to the protocol, available in Supplemental Appendix 1). This trial is registered with Clinicaltrials.gov under identifier NCT00773513 (date of registration October 16, 2008).

The data monitoring committee reviewed the safety data when approximately 25%, 50%, and 75% of the events had occurred, making recommendations concerning study

Table 1. Baseline characteristics of the patients

Characteristic, *n* (%)	Reference ESA (*n*=1409)	Methoxy Polyethylene Glycol-Epoetin Beta (*n*=1409)
Median age, yr (interquartile range)	65 (53–74)	64 (53–74)
Men	828 (59)	805 (57)
Region		
Europe	1038 (74)	1039 (74)
Asia	162 (11)	150 (11)
Australia	73 (5)	75 (5)
Latin America	136 (10)	145 (10)
Treatment condition		
Correction	273 (19)	273 (19)
Maintenance	1136 (81)	1136 (81)
On dialysis	1173 (83)	1194 (85)
Years receiving dialysis	*n*=1173	*n*=1194
<1	290 (25)	288 (24)
1–3	432 (37)	429 (36)
>3	451 (38)	477 (40)
Dialysis modality	*n*=1173	*n*=1194
Peritoneal dialysis	81 (7)	91 (8)
Hemodialysis	1092 (93)	1103 (92)
Cardiovascular risk factors and history		
Ischemic heart disease	420 (30)	376 (27)
Peripheral vascular disease	234 (17)	200 (14)
Cerebral vascular disease	137 (10)	131 (9)
Congestive heart failure	202 (14)	192 (14)
NYHA class I	54 (4)	47 (3)
NYHA class II	121 (9)	112 (8)
NYHA class III	30 (2)	41 (3)
NYHA class IV	10 (1)	15 (1)
Unknown	5 (0)	0 (0)
Venous thrombosis	165 (12)	164 (12)
Hypertension	1267 (90)	1268 (90)
Hyperlipidemia	776 (55)	759 (54)
Diabetes	495 (35)	496 (35)
C-reactive protein ≤30 mg/L	1288/1386 (93)	1283/1379 (93)

There were no significant differences between the two arms with regard to any baseline characteristics. ESA, erythropoiesis-stimulating agent; NYHA, New York Heart Association.

conduct or potential interruption or termination. Because of the length of the study, an additional meeting was convened 1 year after the 75% of events review.

Results

Patients

We enrolled 2825 patients from 186 sites in 27 countries between December 12, 2008 and November 9, 2011. Seven patients never received treatment, which left 2818 patients in a modified intention-to-treat analysis (Figure 1). The two treatment groups were well balanced with regard to all baseline characteristics (Table 1, Supplemental Table 4). The median doses of ESAs administered (calculated as equivalent weekly dose for each of the first 7 years of the study) were 18.8–28.0 μg methoxy polyethylene glycol-epoetin beta, 13.3–23.3 μg darbepoetin alfa, and 3604–5345 IU epoetin alfa/beta (Supplemental Figure 1A).

Over the 8.5-year study period, 1007 patients (36%) across both arms withdrew from the trial without experiencing a primary end point event, and approximately half of these were protocol-mandated withdrawals due to kidney transplantation (501 patients). The median time on treatment in the trial was 3.1 years (interquartile range [IQR], 1.3–5.7) in the methoxy polyethylene glycol-epoetin beta arm and 3.6 years (IQR, 1.6–5.9) in the reference arm. A higher proportion of withdrawals (without experiencing a primary end point) was observed in the first year in the methoxy polyethylene glycol-epoetin beta arm (19%) compared with the reference arm (12%). After the first year, rates of withdrawal during the trial were comparable. Considering separately patients who withdrew during the first year (without an event) or who remained on study after the first year, in each group baseline characteristics, including hemoglobin, transferrin saturation, and dialysis modality, were comparable between arms (Supplemental Table 5).

Hematologic and Biochemical Effects

Median hemoglobin concentration was maintained at 10–12 g/dl throughout the study and was similar between treatment arms (Figure 2A). Overall, the mean proportion of time that patients spent within the therapeutic range was 67% for methoxy polyethylene glycol-epoetin beta and 68% for the reference arm. Median serum ferritin levels were ≥100 ng/ml and were comparable for the treatment arms (Figure 2B), whereas median transferrin saturation levels remained ≥20% throughout the study in both arms (Figure 2C). Median transferrin saturation increased and remained approximately 5% higher in the methoxy

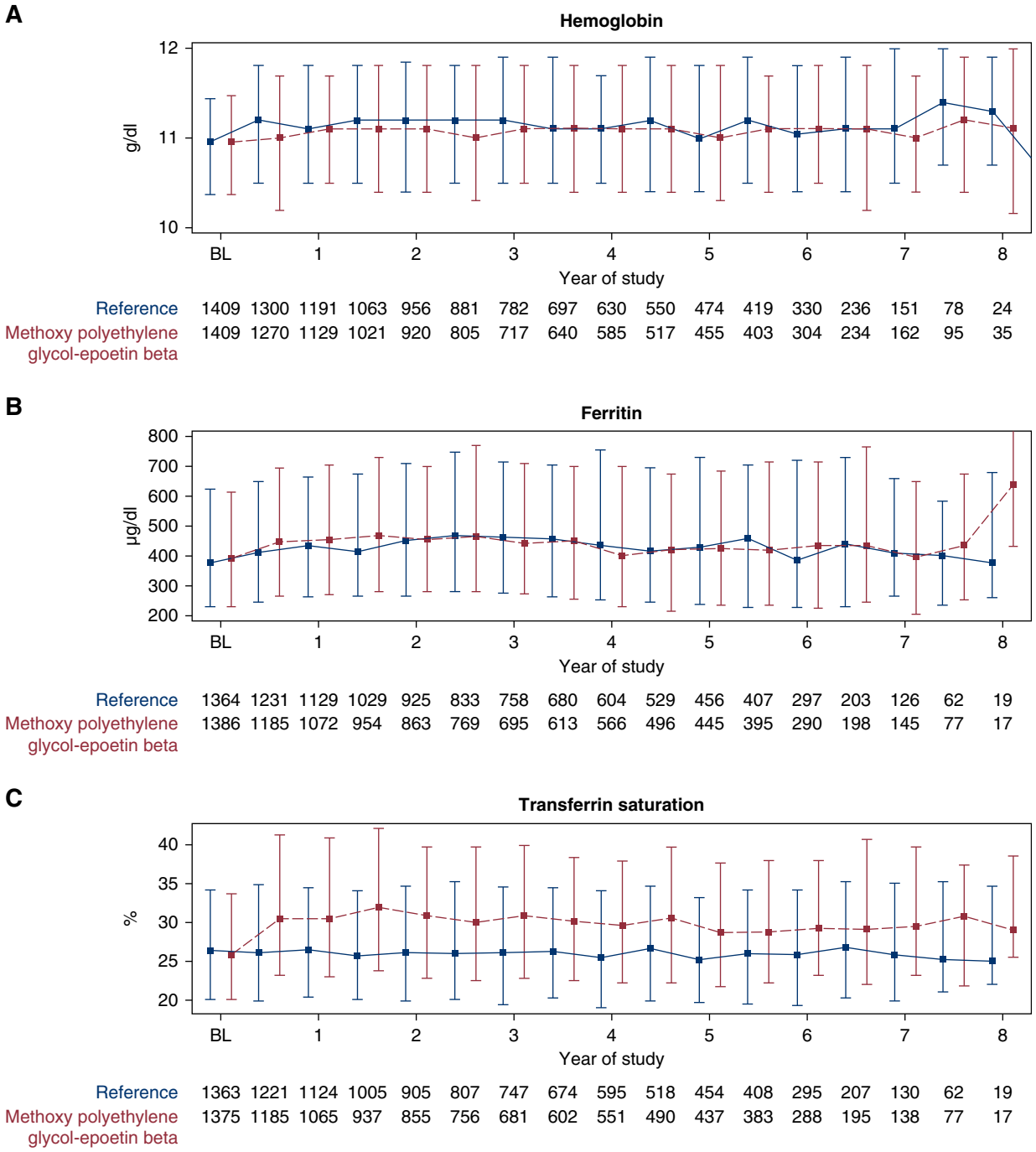


Figure 2. | **Hematologic and iron parameters during the study.** (A) Hemoglobin, (B) ferritin, and (C) transferrin saturation. Values shown are median with IQRs.

polyethylene glycol group than the reference group during the entire study. Iron supplementation (iron salt, route of administration) showed no notable differences between groups (Supplemental Table 6). Median C-reactive protein concentrations and median systolic BP were similar between arms (Supplemental Figure 1, B and C).

Safety

There were 1284 confirmed primary end point events at the study end. Of these, 925 (72%) were deaths (from any cause) and 359 (28%) were nonfatal myocardial infarction or stroke. Primary end point event rates were not different in the methoxy polyethylene glycol-epoetin beta and reference arms (45% and 46% of patients, respectively), with a median time to event of 5.1 years in the methoxy polyethylene glycol-epoetin beta arm (IQR, 2.3–not evaluable) and 5.1 years in the reference ESA arm (IQR, 2.4–not evaluable) (HR, 1.03; 95% CI, 0.93 to 1.15; P=0.004 for noninferiority) (Figure 3). The overall number of deaths during the study was 558 over a median duration of 5.9 years in the methoxy polyethylene

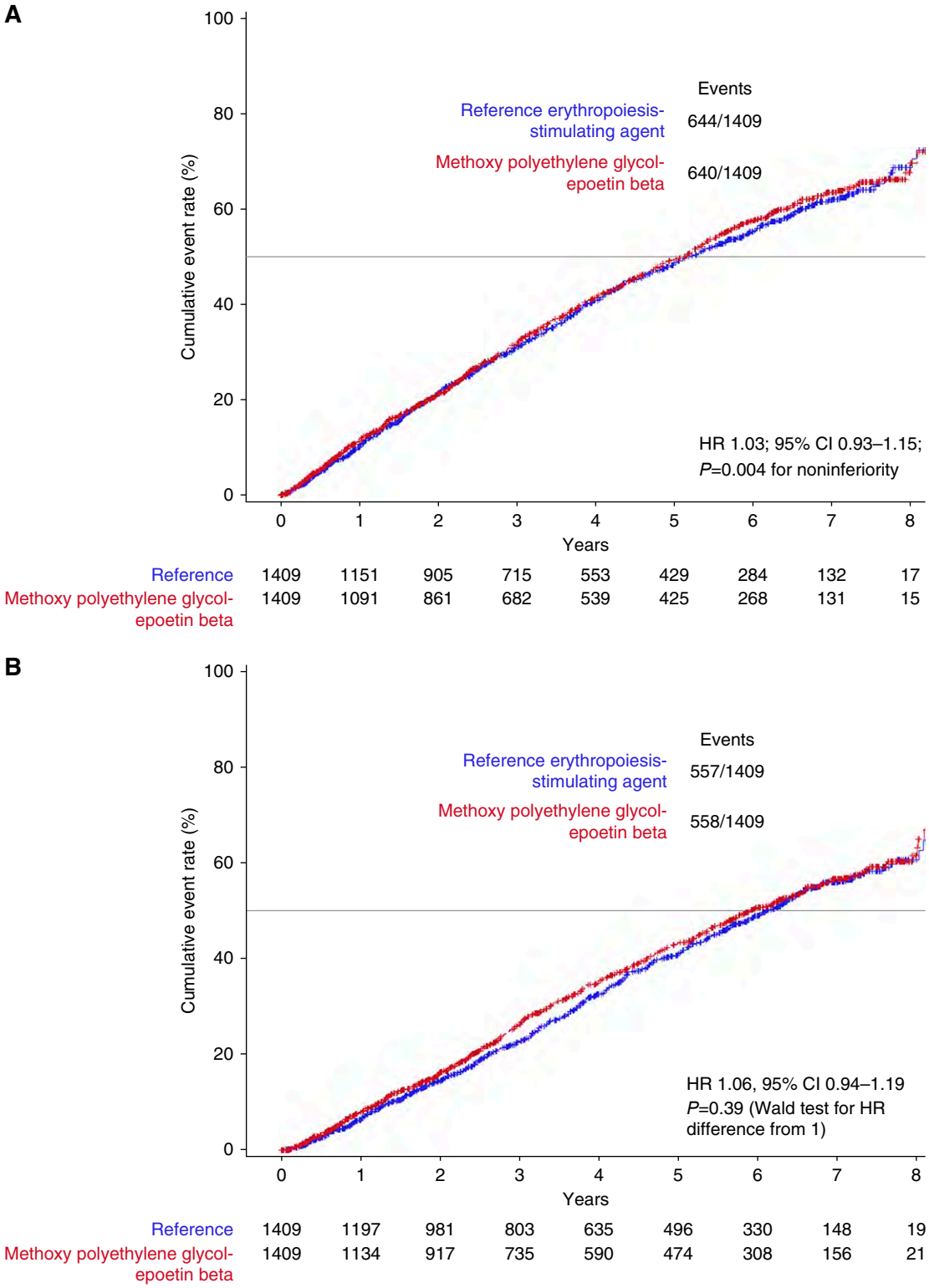


Figure 3. | **Time-to-event curves.** (A) Death from any cause, nonfatal stroke, or nonfatal myocardial infarction (primary end point) and (B) death from any cause.

glycol-epoetin beta arm and 557 over a median duration of 6.1 years in the reference ESA arm, with a HR for all-cause mortality of 1.06 (95% CI, 0.94 to 1.19) (Figure 3B). Rates of nonfatal events (myocardial infarction and stroke) were similar for the two groups (Table 2, Supplemental Figure 2).

In an analysis by subgroups, the results with regard to the primary end point showed an interaction only for patients

Table 2. Major safety end points

End Point	Reference ESA (n=1409)	Methoxy Polyethylene Glycol-Epoetin Beta (n=1409)	Hazard Ratio (95% CI)	P Value	
	No. of Patients (%)	No. of Patients (%)		For Noninferiority[a]	For Difference[b]
Primary end point (composite of all-cause death, nonfatal MI, and nonfatal stroke)	644 (46)	640 (45)	1.03 (0.93 to 1.15)	0.004	0.54
Secondary end point					
Death	557 (40)	558 (40)	1.06 (0.94 to 1.19)		0.36
Nonfatal MI or stroke	191 (14)	168 (12)	0.91 (0.74 to 1.12)		0.39
Fatal or nonfatal MI	158 (11)	143 (10)	0.95 (0.76 to 1.19)		0.66
Fatal or nonfatal stroke	98 (7)	89 (6)	0.94 (0.70 to 1.25)		0.66

ESA, erythropoiesis-stimulating agent; 95% CI, 95% confidence interval; MI, myocardial infarction.
[a]P value for the test for noninferiority at a hazard ratio of 1.20 (primary endpoint only).
[b]P value from the Wald test for hazard ratio difference from 1.

with a history of congestive heart failure (Figure 4). As anticipated, patients not receiving dialysis at the initiation of the trial had a longer time to a primary end point event than those on dialysis at the start of the trial (Supplemental Figure 3).

Other Analyses

Causes of Death. Among the adjudicated causes of death, noncardiovascular death, and death sudden etiology unknown were the most prevalent classifications (Supplemental Table 7). A greater number of deaths sudden etiology unknown occurred in the methoxy polyethylene glycol-epoetin beta arm (185 versus 136), whereas a greater number of noncardiovascular deaths occurred in the reference arm (264 versus 223). No association was found between causes of death and patient baseline characteristics.

Hemoglobin Concentrations and ESA Doses before an End Point Event. Exploratory analyses of the on-study 3-month average hemoglobin concentrations demonstrated that hemoglobin concentrations <10 g/dl were associated with an almost three-fold higher risk of experiencing a primary end point event (HR, 2.76; 95% CI, 2.41 to 3.17) compared with the reference category of 10–11 g/dl. In patients with hemoglobin 11 to ≤12 or ≥12 g/dl, the risk was significantly less than the reference category (Supplemental Table 8). The risk of a primary end point was also greater in patients receiving higher doses of ESAs (Supplemental Table 8). There was no association found between causes of death and hemoglobin levels or doses of ESAs received.

Other Safety Data

The percentage of patients with gastrointestinal bleeding (11.7% in the methoxy polyethylene glycol-epoetin beta group versus 11.1% in the reference group) and thromboembolic events (32.8% in the methoxy polyethylene glycol-epoetin beta group versus 34.5% in the reference group) were similar. No cases of antibody-mediated pure red cell aplasia occurred in the trial. Adverse events occurring in ≥5% of patients are shown in Supplemental Table 9.

Discussion

In the MIRCERA PASS trial, treatment with methoxy polyethylene glycol-epoetin beta resulted in rates of major cardiovascular events and mortality that were similar to rates with the reference ESAs among patients with anemia associated with CKD. The results of the analysis of the individual components of the primary end point (mortality, nonfatal myocardial infarction, and nonfatal stroke) and of analyses of deaths from any cause were consistent with those of the primary composite end point. The similar rates of the primary end point in the methoxy polyethylene glycol-epoetin beta and reference ESA groups were observed in the context of comparable hemoglobin levels.

The primary results of the MIRCERA PASS trial, which has the longest duration of follow-up of any study of anemia treatment in CKD, are consistent with the results from other pooled analyses of trials in the development program of methoxy polyethylene glycol-epoetin beta (14,15), as well as a Cochrane systematic review specifically analyzing the safety of methoxy polyethylene glycol-epoetin beta compared with the other available ESAs (16).

ESAs are effective in correcting anemia and maintaining hemoglobin concentrations in the majority of patients with CKD, although safety concerns have been raised about both higher cardiovascular and thrombotic risks when these agents are administered at higher doses or when the hemoglobin target is >13 g/dl (6–9). At the time the MIRCERA PASS study was initiated in December 2008, several studies had failed to demonstrate that correction of anemia to a target hemoglobin >13 g/dl in patients with CKD reduced the risk of cardiovascular events (6,7,9). MIRCERA PASS was not designed to look at different target hemoglobin levels, but exploratory analyses suggest that risk of cardiovascular events or all-cause death was highest in patients with low hemoglobin

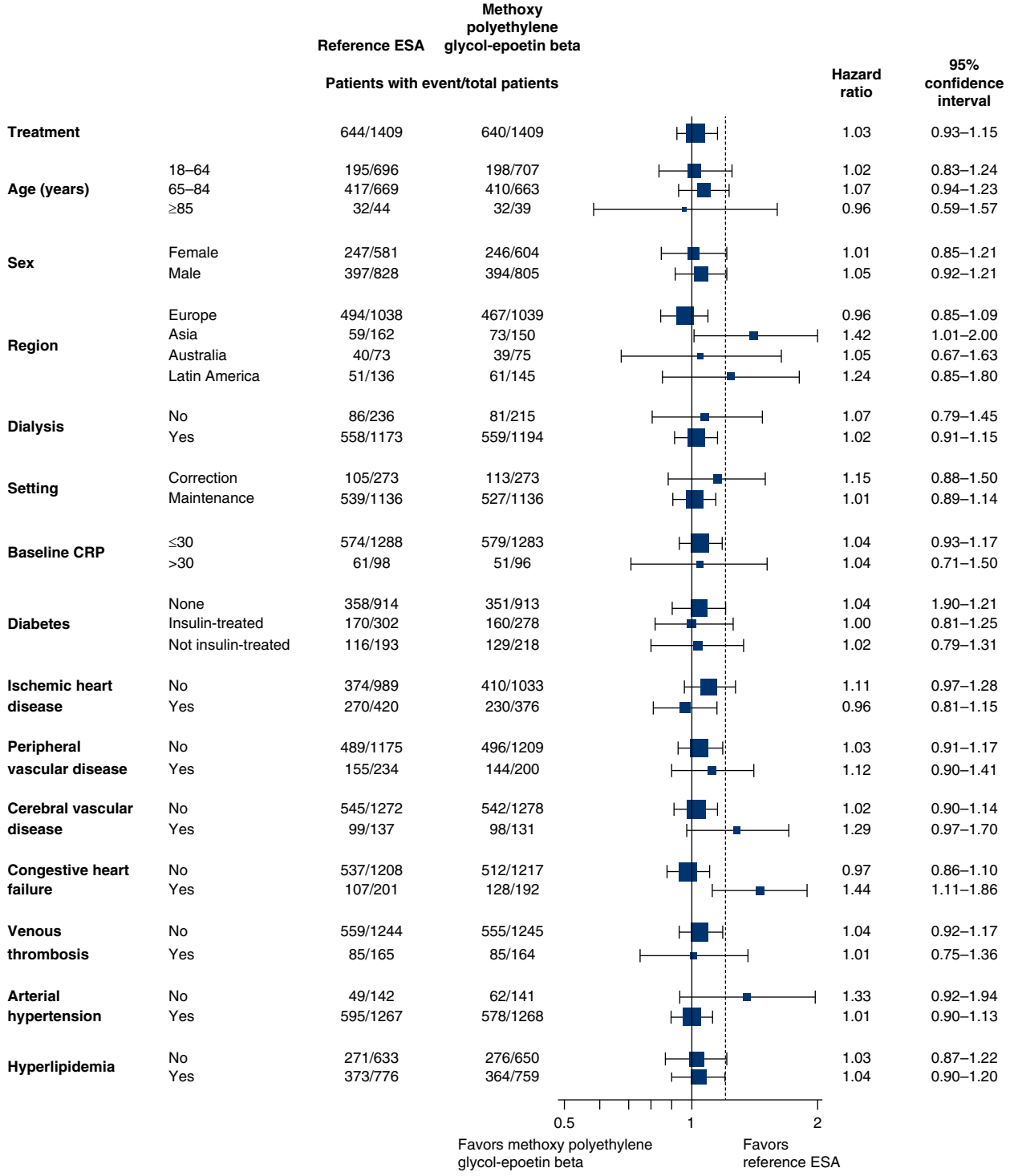


Figure 4. | Subgroup analysis of the primary end point by baseline patient and disease factors.

levels on treatment: no relative increases in cardiovascular events or death were observed in patients who had on-treatment values of ≥12 g/dl, whereas those with hemoglobin concentrations of ≤10 g/dl had increased rates of cardiovascular morbidity and all-cause mortality. Although these analyses were exploratory, and the association of low hemoglobin with cardiovascular morbidity and mortality may be confounded by disease factors or comorbidities, the results are consistent with a previous meta-analysis of the development trials of methoxy polyethylene glycol-epoetin beta (15). Higher doses of all ESAs were associated with increased rates of primary end point events. Analysis of prespecified subgroups supported the overall analyses.

More patients in the methoxy polyethylene glycol-epoetin beta arm than the reference arm withdrew during the first

year. This was not unexpected, given that the trial was open label and patients and physicians may have been more cautious when switching from one ESA to another compared with those in the reference arm who continued a treatment regimen with which they were already familiar. Most withdrawals were for nonsafety reasons, with no evidence to suggest that the higher rate of withdrawal during the first year in the methoxy polyethylene glycol-epoetin beta selected a healthier population in one arm versus the other.

Important limitations of the MIRCERA PASS trial include the open-label design, because both the investigator and patient were aware of the treatment randomization. However, patient and investigator blinding would have been unrealistic because of the differences in dosing schedules. The primary end point events (death, myocardial infarction, and stroke) were prospectively adjudicated by an independent end points committee who were blinded to study treatment throughout the trial. This process and the large number of events, long length of follow-up, and a contemporary, representative high-cardiovascular-risk population reflecting clinical practice are important study strengths. Iron supplementation was used as required throughout the study, to maintain iron parameters above the minimum threshold in both treatment groups (serum ferritin ≥100 ng/ml or transferrin saturation ≥20%), with no restriction on dose, formulation, or route of administration. This may be considered both a limitation, as different modalities may affect ESA use differently, and a strength, as it reflects clinical practice.

All-cause mortality was the largest component of the primary end point. Over half of all deaths were adjudicated as having a clear noncardiovascular cause, with the second largest category being "sudden death etiology unknown;" by definition, this category included all cases where the cause of death was unclear. Although there were some differences between treatment arms in the cause of death categorizations, this is not likely to be explained in terms of a differential effect of methoxy polyethylene glycol-epoetin beta, in the context of having a similar number of deaths overall.

Since completion of the MIRCERA PASS study, an observational cohort study from Japan has reported a 13% higher rate of 2-year mortality in patients on dialysis receiving long-acting ESAs compared with shorter-acting ESAs (17). The study population of MIRCERA PASS was diverse, including patients from several countries and both nondialysis and dialysis subgroups. Importantly, results obtained from a large randomized trial with several years of follow-up, with prospective adjudication of all cardiovascular events blinded to treatment assignment, remain the most robust form of evidence available for the safety of the ESAs used in patients with CKD. The data from the large observational study reported by Sakaguchi *et al.* (17), despite propensity matching and statistical adjustments, are hampered by indication bias and residual confounders that make direct comparisons between the two studies difficult.

In conclusion, among patients with anemia of CKD, treatment with monthly methoxy polyethylene glycol-epoetin beta resulted in overall rates of major cardiovascular events, all-cause mortality, and other serious adverse events that were similar to those associated with conventional reference ESAs administered more frequently.

Data-sharing statement

Qualified researchers may request access to individual patient-level data for use in further research studying the medicine or disease that was researched in the original studies through the clinical study data request platform (www.clinicalstudydatarequest.com). An independent panel reviews any research requests and allows access to the data through the platform where the proposal is appropriate. A signed Data Sharing Agreement is required before data access can be provided, as part of this the researchers commit to publish the results of the analysis in a scientific journal (or open-access journal/platform) within 1 year of analysis completion. Data will be available after the medicine studied has been approved by regulators for the indication in both the United States and European Union and at least 18 months after completion of the study report. Data can be shared where Roche have the legal authority to provide the data and consider it feasible to anonymize the data without compromising the privacy and confidentiality of research participants, amongst other criteria. Further details on Roche's criteria for eligible studies are available at https://clinicalstudydatarequest.com/Study-Sponsors/Study-Sponsors-Roche.aspx. Further details on Roche's Global Policy on the Sharing of Clinical Information and how to request access to related clinical study documents are available at https://www.roche.com/research_and_development/who_we_are_how_we_work/clinical_trials/our_commitment_to_data_sharing.htm.

Acknowledgments

We thank all the study investigators and patients who participated in the trial (for a full list of study investigators, please see Supplemental Appendix 2). All authors received medical writing support from F. Hoffmann-La Roche Ltd., furnished by Dr. John Carron of Health Interactions. The study was sponsored by F. Hoffmann-La Roche Ltd., Basel, Switzerland.

Dr. Locatelli and Dr. Oguey were involved in design of the study. Dr. Locatelli and Dr. Hannedouche collected data. Dr. Oguey collated, analyzed, and interpreted the data. Ms. Morgan was responsible for the statistical analysis. Dr. White was chair of the cardiovascular end points committee throughout the trial. Dr. Locatelli, Dr. Fishbane, Dr. Hannedouche, and Dr. White drafted and edited sections of the manuscript. All authors approved the final submitted manuscript.

Disclosures

Dr. Fishbane reports research support from AstraZeneca, Keryx, Corvidia, and Akebia and having been a consultant for AstraZeneca and Corvidia. Dr. Locatelli reports having received personal fees from Akebia, Astellas, and Astra Zeneca as a consultant and speaker and having received personal fees from Amgen, Roche, and Vifor-Fresenius Medical Care as a consultant. Dr. Locatelli also reports having served on advisory boards for Akebia, Amgen, Astellas, AstraZeneca, GSK, and Roche. Ms. Morgan and Dr. Oguey are employees of F. Hoffmann-La Roche Ltd. Dr. White reports personal fees from a position as member of steering committee of the trial at Relypsa Inc. Dr. White and Dr. Fishbane report having served on the endpoint adjudication committee in the current trial for F. Hoffmann-La Roche Ltd. All authors report medical writing support from F. Hoffmann-La Roche Ltd.

Supplemental Material

This article contains the following supplemental material online at http://cjasn.asnjournals.org/lookup/suppl/doi:10.2215/CJN.01380219/-/DCSupplemental.

Supplemental Appendix 1. Cardiovascular end point definitions.

Supplemental Appendix 2. List of investigators.

Supplemental Table 1. Inclusion and exclusion criteria.

Supplemental Table 2. Starting dose of methoxy polyethylene glycol-epoetin beta according to previous ESA treatment when switching from another ESA.

Supplemental Table 3. Schedule of assessments.

Supplemental Table 4. Additional baseline characteristics.

Supplemental Table 5. Concomitant iron supplementation.

Supplemental Table 6. Selected baseline parameters by withdrawal in the first year.

Supplemental Table 7. Adjudicated causes of death.

Supplemental Table 8. Time-dependent Cox regression models for hemoglobin level and dose prior to the event.

Supplemental Table 9. Adverse events experienced by ≥5% of patients.

Supplemental Figure 1. Doses of erythropoiesis-stimulating agents, C-reactive protein levels, and systolic BP during the study.

Supplemental Figure 2. Time-to-event curves for time to myocardial infarction and time to stroke.

Supplemental Figure 3. Time-to-event curves for death from any cause, nonfatal stroke, or nonfatal myocardial infarction, for patients on dialysis or not on dialysis.

References

1. Levin A, Thompson CR, Ethier J, Carlisle EJ, Tobe S, Mendelssohn D, Burgess E, Jindal K, Barrett B, Singer J, Djurdjev O: Left ventricular mass index increase in early renal disease: Impact of decline in hemoglobin. *Am J Kidney Dis* 34: 125–134, 1999
2. Kidney Disease: Improving Global Outcomes (KDIGO) Working Group: KDIGO clinical practice guideline for anemia in chronic kidney disease. 2012. Available at: https://kdigo.org/wp-content/uploads/2016/10/KDIGO-2012-Anemia-Guideline-English.pdf. Accessed August 5, 2019
3. Martinez-Vea A, Bardají A, García C, Ridao C, Richart C, Oliver JA: Long-term myocardial effects of correction of anemia with recombinant human erythropoietin in aged patients on hemodialysis. *Am J Kidney Dis* 19: 353–357, 1992
4. Fuller DS, Zepel L, Bieber BA, Robinson BM, Pisoni RL: Hemodialysis facility variation in hospitalization and transfusions using Medicare claims: The DOPPS Practice Monitor for US dialysis care. *Am J Kidney Dis* 67: 337–340, 2016
5. Cody JD, Hodson EM: Recombinant human erythropoietin versus placebo or no treatment for the anaemia of chronic kidney disease in people not requiring dialysis. *Cochrane Database Syst Rev* (1): CD003266, 2016
6. Drüeke TB, Locatelli F, Clyne N, Eckardt KU, Macdougall IC, Tsakiris D, Burger HU, Scherhag A; CREATE Investigators: Normalization of hemoglobin level in patients with chronic kidney disease and anemia. *N Engl J Med* 355: 2071–2084, 2006
7. Besarab A, Bolton WK, Browne JK, Egrie JC, Nissenson AR, Okamoto DM, Schwab SJ, Goodkin DA: The effects of normal as compared with low hematocrit values in patients with cardiac disease who are receiving hemodialysis and epoetin. *N Engl J Med* 339: 584–590, 1998
8. Pfeffer MA, Burdmann EA, Chen CY, Cooper ME, de Zeeuw D, Eckardt KU, Feyzi JM, Ivanovich P, Kewalramani R, Levey AS, Lewis EF, McGill JB, McMurray JJ, Parfrey P, Parving HH, Remuzzi G, Singh AK, Solomon SD, Toto R; TREAT Investigators: A trial of darbepoetin alfa in type 2 diabetes and chronic kidney disease. *N Engl J Med* 361: 2019–2032, 2009
9. Singh AK, Szczech L, Tang KL, Barnhart H, Sapp S, Wolfson M, Reddan D; CHOIR Investigators: Correction of anemia with epoetin alfa in chronic kidney disease. *N Engl J Med* 355: 2085–2098, 2006
10. Pérez-García R, Varas J, Cives A, Martín-Malo A, Aljama P, Ramos R, Pascual J, Stuard S, Canaud B, Merello JI; ORD Group: Increased mortality in haemodialysis patients administered high doses of erythropoiesis-stimulating agents: A propensity score-matched analysis. *Nephrol Dial Transplant* 33: 690–699, 2018
11. Locatelli F, Bárány P, Covic A, De Francisco A, Del Vecchio L, Goldsmith D, Hörl W, London G, Vanholder R, Van Biesen W; ERA-EDTA ERBP Advisory Board: Kidney disease: Improving Global Outcomes guidelines on anaemia management in chronic kidney disease: A European Renal Best Practice position statement. *Nephrol Dial Transplant* 28: 1346–1359, 2013
12. European Medicines Agency (EMA): Mircera. European Public Assessment Report (EPAR): Scientific Discussion, 2007. Available at: http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_-_Scientific_Discussion/human/000739/WC500033669.pdf. Accessed November 19, 2018
13. Locatelli F, Aljama P, Bárány P, Canaud B, Carrera F, Eckardt KU, Hörl WH, Macdougal IC, Macleod A, Wiecek A, Cameron S; European Best Practice Guidelines Working Group: Revised European best practice guidelines for the management of anaemia in patients with chronic renal failure. *Nephrol Dial Transplant* 19 [Suppl 2]: ii1–ii47, 2004
14. Locatelli F, Mann JF, Aldigier JC, Sanz Guajardo D, Schmidt R, Van Vlem B, Sulowicz W, Dougherty FC, Beyer U: C.E.R.A. Safety profile: A pooled analysis in patients with chronic kidney disease. *Clin Nephrol* 73: 94–103, 2010
15. Locatelli F, de Francisco A, Deray G, Fliser D, Armstrong G, Dougherty FC, Ehrhard P: Mortality and cardiovascular morbidity associated with haemoglobin levels: A pooled analysis of randomised controlled trials. *Nephron Clin Pract* 128: 323–332, 2014
16. Saglimbene VM, Palmer SC, Ruospo M, Natale P, Craig JC, Strippoli GF: Continuous erythropoiesis receptor activator (CERA) for the anaemia of chronic kidney disease. *Cochrane Database Syst Rev* 8: CD009904, 2017
17. Sakaguchi Y, Hamano T, Wada A, Masakane I: Types of erythropoietin-stimulating agents and mortality among patients undergoing hemodialysis. *J Am Soc Nephrol* 30: 1037–1048, 2019

Received: February 1, 2019 **Accepted:** July 10, 2019

Published online ahead of print. Publication date available at www.cjasn.org.

An international observational study suggests that artificial intelligence for clinical decision support optimizes anemia management in hemodialysis patients

see commentary on page 259

Carlo Barbieri[1], Manuel Molina[2], Pedro Ponce[3], Monika Tothova[4], Isabella Cattinelli[1], Jasmine Ion Titapiccolo[1], Flavio Mari[1], Claudia Amato[1], Frank Leipold[1], Wolfgang Wehmeyer[1], Stefano Stuard[1], Andrea Stopper[1] and Bernard Canaud[1,5]

[1]*Fresenius Medical Care, Bad Homburg, Germany;* [2]*Servicio de Nefrologia, Hospital Universitario Santa Lucía, Cartagena, Spain;* [3]*Fresenius Medical Care–Dialysis Center Lumiar, Lisbon, Portugal;* [4]*Fresenius Medical Care–Dialysis Center Motol, Prague, Czech Republic; and* [5]*Montpellier University I, UFR Medicine, Montpellier, France*

Managing anemia in hemodialysis patients can be challenging because of competing therapeutic targets and individual variability. Because therapy recommendations provided by a decision support system can benefit both patients and doctors, we evaluated the impact of an artificial intelligence decision support system, the Anemia Control Model (ACM), on anemia outcomes. Based on patient profiles, the ACM was built to recommend suitable erythropoietic-stimulating agent doses. Our retrospective study consisted of a 12-month control phase (standard anemia care), followed by a 12-month observation phase (ACM-guided care) encompassing 752 patients undergoing hemodialysis therapy in 3 NephroCare clinics located in separate countries. The percentage of hemoglobin values on target, the median darbepoetin dose, and individual hemoglobin fluctuation (estimated from the intrapatient hemoglobin standard deviation) were deemed primary outcomes. In the observation phase, median darbepoetin consumption significantly decreased from 0.63 to 0.46 μg/kg/month, whereas on-target hemoglobin values significantly increased from 70.6% to 76.6%, reaching 83.2% when the ACM suggestions were implemented. Moreover, ACM introduction led to a significant decrease in hemoglobin fluctuation (intrapatient standard deviation decreased from 0.95 g/dl to 0.83 g/dl). Thus, ACM support helped improve anemia outcomes of hemodialysis patients, minimizing erythropoietic-stimulating agent use with the potential to reduce the cost of treatment.

Kidney International (2016) **90,** 422–429; http://dx.doi.org/10.1016/j.kint.2016.03.036

KEYWORDS: anemia; chronic kidney disease; erythropoietin; hemodialysis

Correspondence: *Carlo Barbieri, Via Crema 8, 26020 Palazzo Pignano (CR), Italy. E-mail: Carlo.Barbieri@fmc-ag.com*

Received 1 December 2015; revised 21 March 2016; accepted 31 March 2016; published online 2 June 2016

Anemia management in end-stage kidney disease patients (ESKD) receiving hemodialysis (HD) and treated by erythropoietic-stimulating agents (ESAs) is an important task for nephrologists who are asked to achieve several objectives at the same time, at both the patient and facility levels.

Briefly, hemoglobin (Hb) values should be maintained in a quite narrow target window, in a stable manner, using the smallest possible ESA doses. These are all delicate objectives on their own because of the following:

(i) Hb targets have changed over time, as well as in special clinical situations[1–4]; target levels have been reduced and the window has been narrowed (10–12 g/dl), according to the results of recent randomized trials[5–7] and a large meta-analysis.[8] In clinical practice, it is difficult to maintain Hb levels within such narrow range due to substantial inter- and intrapatient variability.[9–11]

(ii) Hb variability needs to be minimized to prevent undesired effects in fragile patients,[11–13] but this is not a trivial issue considering that, as reported, for example, by Berns *et al.*,[10] 1-month Hb values exhibit the greatest degree of variability, with ~20% of the patients showing Hb variations >3.3 g/dl.

(iii) The ESA dose has to be reduced to mitigate ESA-related hazards.[14] The U.S. Food and Drug Administration recommends administering the lowest ESA dose needed to avoid recurrent blood transfusions.

(iv) Cost-related issues have also emerged as an additional hurdle, questioning the cost-benefit value of ESAs to treat anemia in dialysis patients.[15]

Successfully achieving all of these requirements can place an additional workload on nephrologists caring for a large number of patients; this is aggravated by the complexity and heterogeneity of the ESKD population, presenting with different medical profiles and diverse, possibly changing, sensitivity to ESAs, leading to the need for more precise, personalized dose adjustments. Given the importance of anemia management for the patient's well-being, developing interactive guided clinical tools to support the physician's work would be a favorable advancement.

In recent years, a variety of predictive algorithms based on sophisticated modeling approaches have been proposed to

predict Hb levels in ESKD patients and to offer a personalized treatment in line with the predicted Hb trend[16–19]; their promising results suggest that such approaches can be powerful tools for anemia management in dialysis patients. Some of these algorithms have also been tested in a clinical setting, albeit often in relatively small cohorts of patients.[20–23]

In a previous work, we built our anemia modeling approach and evaluated its reliability and predictive value in a retrospective study involving a large number of ESKD patients treated in the Fresenius Medical Care clinical network.[24] Motivated by our encouraging results, we decided to deploy our decision support system, the Anemia Control Model (ACM), in 3 pilot clinics as part of the daily care routine of a large population of unselected patients. Complementing the i.v. iron therapy based on internal protocols following best practice guidelines (see Supplementary Appendix), the ACM computes the ESA dose suggestions based on the following 2 components: (i) an artificial neural network model that uses patients' clinical data as input and predicts future Hb concentrations[24] and (ii) an algorithm that, simulating the effect of different ESA doses, determines the optimal prescription to achieve the desired Hb targets.

The purpose of this study was to determine how ACM support can affect outcomes of anemia management in daily clinical practice, with the aims of maintaining Hb targets and reducing Hb variability and ESA consumption in ESKD patients.

RESULTS

Outcome at the dialysis facility level

Baseline characteristics of ESKD patients participating in the study are presented in Table 1; 653 patients were included in the control phase and 640 in the observation phase, for a total of 752 patients participating in at least 1 phase. These 2 populations were quite similar in both clinical characteristics and lab data at baseline. In the control phase, therapy for anemia was devised by the attending physicians, following established best clinical practices and internal network Standard Operating Procedures (Supplementary Appendix), without ACM support; during the observation phase, physicians were provided with ACM recommendations.

Anemia outcomes are presented in Table 2. During ACM-guided care, darbepoetin consumption decreased by 25% (from 40 [interquartile range, 100] to 30 [interquartile range, 100] μg/month), whereas the percentage of Hb values within the target range increased by 6% (from 70.6% to 76.6%) in the entire population. It should be noted, however, that only a portion of Hb values in the observation phase resulted from *accepted* ACM suggestions; others were obtained after rejecting the suggestion or independently of the ACM (as the patient was ACM ineligible at the time). Therefore, to more closely evaluate ACM value, we consider anemia outcomes when suggestions were actually confirmed. Both figures show a more decisive improvement (83.2% Hb values on target; median darbepoetin = 20 [interquartile range, 80] μg/month). Between the study phases, the percentage of Hb values over target decreased (from 17% to 9.8%), whereas the percentage of Hb values below target range slightly increased (from 12.3% to 13.6%); however, when considering only lab tests resulting from accepted ACM suggestions, both percentages actually decreased (to 7.5% and 9.3%, respectively). Iron consumption also decreased across the 2 periods.

Adverse events, i.e., mortality, cardiovascular events, hospitalizations, and transfusions, were also extracted. All these events tended to decrease after ACM entrance (Table 2).

Table 1 | Patients characteristics in the 2 study periods in the facility level analysis

Characteristics	Control phase	Observation phase	*P*-value
Total no. of patients	653	640	
Age, yr, mean ± SD	63.65 ± 15.45	63.86 ± 15.46	0.81[b]
Male, no. (%)	409 (62.6)	397 (62.0)	0.86[a]
Patients initiating RRT, no. (%)	70 (10.7)	62 (9.7)	0.58[a]
Comorbidities, no. (%)			
Coronary artery disease	59 (9.0)	56 (8.8)	0.92[a]
Congestive heart failure	147 (22.5)	145 (22.7)	1.00[a]
Peripheral vascular disease	187 (28.6)	184 (28.8)	1.00[a]
Cerebrovascular disease	114 (17.5)	115 (18.0)	0.83[a]
Chronic pulmonary disease	96 (14.7)	92 (14.4)	0.87[a]
Diabetes	196 (30.0)	188 (29.4)	0.81[a]
Charlson Comorbidity Index, mean ± SD	5.98 ± 3.98	5.76 ± 3.86	0.28[b]
Causes of kidney disease, no. (%)			
Diabetes	141 (21.6)	140 (21.9)	0.95[a]
Hypertension	123 (18.8)	126 (19.7)	0.72[a]
Chronic glomerulonephritis	143 (21.9)	133 (20.8)	0.64[a]
Urinary obstruction/chronic interstitial nephritis	11 (1.7)	10 (1.6)	1.00[a]
Polycystic kidney disease	39 (6.0)	41 (6.4)	0.82[a]
Other	196 (30.0)	190 (29.7)	0.90[a]
Vascular access, no. (%)			
Fistula	427 (65.4)	418 (65.3)	1.00[a]
Catheter	130 (19.9)	121 (18.9)	0.67[a]
Graft	96 (14.7)	101 (15.8)	0.64[a]
Treatment modality, no. (%)			
HDF online	608 (93.1)	595 (93.0)	1.00[a]
High-flux HD	32 (4.9)	36 (5.6)	0.62[a]
Other	13 (2.0)	9 (1.4)	0.52[a]
Laboratory test value			
Hemoglobin, g/dl, mean ± SD	11.32 ± 1.08	11.19 ± 1.07	0.02[b]
Ferritin, ng/ml, median (IQR)	526.90 (365.88)	580.65 (325.10)	0.03[c]
TSAT, %, median (IQR)	29.77 (13.14)	30.50 (12.47)	0.21[c]
Albumin, g/dl, mean ± SD	3.90 ± 0.45	3.92 ± 0.38	0.43[b]
Calcium, mg/dl, mean ± SD	8.79 ± 0.60	8.92 ± 0.62	<0.001[b]
Phosphate, mg/dl, mean ± SD	4.37 ± 1.07	4.30 ± 1.02	0.21[b]
Potassium, mmol/l, mean ± SD	4.95 ± 0.65	4.92 ± 0.62	0.45[b]
PTH, ng/l, median (IQR)	276.45 (240.13)	271.70 (245.00)	0.85[c]
Overhydration, l, mean ± SD	1.83 ± 1.62	1.91 ± 1.40	0.35[b]
eKTV, mean ± SD	1.67 ± 0.42	1.70 ± 0.31	0.54[b]
spKTV, mean ± SD	1.90 ± 0.47	1.94 ± 0.36	0.45[b]

Overhydration was estimated by bioimpedance by means of the body composition monitor. For laboratory tests, mean/median values were computed for each patient and then averaged across all patients.
Ch Int, chronic interstitial; eKTV/spKTV, equilibrated/single-pool Kt/V; HD, hemodialysis; HDF, hemodiafiltration; PTH, parathyroid hormone; RRT, renal replacement therapy; TSAT, transferrin saturation index.
[a]Fisher exact test.
[b]Unpaired *t* test.
[c]Wilcoxon test.

Table 2 | Anemia outcomes and adverse events comparison before and after ACM introduction

	Control phase	Observation phase	*P* value
Hb control: Hb within target range, no. (%)			
All Hb measurements	4555 (70.6)	4946 (76.6)	<0.001[a]
Accepted suggestions[d]		3292 (83.2)	
Rejected suggestions[e]		432 (67.9)	
No suggestion[f]		1222 (65.6)	
Hb control: Hb above target range, no. (%)			
All Hb measurements	1099 (17.0)	630 (9.8)	<0.001[a]
Accepted suggestions		298 (7.5)	
Rejected suggestions		118 (18.6)	
No suggestion		214 (11.5)	
Hb control: Hb below target range, no. (%)			
All Hb measurements	796 (12.3)	880 (13.6)	0.03[a]
Accepted suggestions		367 (9.3)	
Rejected suggestions		86 (13.5)	
No suggestion		427 (22.9)	
Consumption, median (IQR)			
Darbepoetin per month per patient per kilogram (μg/kg/mo)	0.63 (1.51)	0.46 (1.44)	<0.001[b]
Iron per month per patient per kilogram (mg/kg/mo)	1.79 (4.05)	1.67 (3.45)	<0.001[b]
Darbepoetin per month per patient (μg/mo)	40.00 (100.00)	30.00 (100.00)	<0.001[b]
Accepted suggestions		20.00 (80.00)	
Rejected suggestions		60.00 (90.00)	
No suggestion		30.00 (120.00)	
Iron per month per patient (mg/mo)	100.00 (300.00)	100.00 (200.00)	<0.001[b]
Accepted suggestions		100.00 (200.00)	
Rejected suggestions		100.00 (200.00)	
No suggestion		100.00 (300.00)	
Adverse events			
Deaths, no. (%)	59 (9.0)	42 (6.6)	0.12[a]
Patients with cardiovascular events, no. (%)	179 (27.4)	157 (24.5)	0.25[a]
Cardiovascular events (incidence/1000 patient-years)	516.66	439.66	0.008[c]
Hospitalization days (incidence/1000 patient-years)	8104.92	6869.99	<0.001[c]
Patients with transfusion events, no. (%)	43 (6.6)	28 (4.4)	0.09[a]
Transfusion events (incidence/1000 patient-years)	152.4	91.67	<0.001[c]

ACM, Anemia Control Model; Hb, hemoglobin; IQR, interquartile range.
[a]Fisher exact test.
[b]Wilcoxon test.
[c]Poisson exact test.
[d]Hb measurements ($n = 3957$) following ACM suggestions that were accepted by the physician.
[e]Hb measurements ($n = 636$) following ACM suggestions that were rejected by the physician.
[f]Hb measurements ($n = 1863$) that were ACM ineligible.

Outcome at patient level

Table 3 shows the characteristics of the 2 cohorts considered for the patient-level analysis. Only patients with at least 6 Hb measurements in each study phase were included ($n = 383$); among these, we also isolated the subpopulation of patients for whom suggestions were accepted most of the time (ACM-compliant group; $n = 313$). Outcomes and adverse events are reported in Table 4.

Table 3 | Characteristics of the 2 cohorts of patients in the patient longitudinal analysis

Characteristics	All patients	ACM-compliant patients
No. of patients	383	313
Follow-up period, mo, mean ± SD	22.12 ± 2.40	22.06 ± 2.50
Age, yr, mean ± SD	65.18 ± 14.89	65.23 ± 14.83
Male, no. (%)	231 (60.3)	193 (61.7)
Comorbidities at ACM entrance, no. (%)		
Coronary artery disease	33 (8.6)	24 (7.7)
Congestive heart failure	82 (21.4)	69 (22.0)
Peripheral vascular disease	114 (29.8)	87 (27.8)
Cerebrovascular disease	71 (18.5)	56 (17.9)
Chronic pulmonary disease	58 (15.1)	49 (15.7)
Diabetes	87 (22.7)	83 (26.5)
Charlson Comorbidity Index, mean ± SD	6.98 ± 3.30	6.86 ± 3.26
Causes of kidney disease, no. (%)		
Diabetes	75 (19.6)	60 (19.2)
Hypertension	69 (18.0)	62 (19.8)
Chronic glomerulonephritis	88 (23.0)	63 (20.1)
Urinary obstruction/chronic interstitial nephritis	10 (2.6)	6 (1.9)
Polycystic kidney disease	25 (6.5)	23 (7.4)
Other	116 (30.3)	99 (31.6)
Vascular access, no. (%):		
Fistula	261 (68.1)	219 (70.0)
Catheter	59 (15.4)	48 (15.3)
Graft	63 (17.2)	46 (14.7)
Treatment modality, no. (%)		
HDF online	361 (94.3)	296 (94.6)
High-flux HD	14 (3.7)	9 (2.9)
Other	7 (1.8)	8 (2.6)

ACM, Anemia Control Model; HD, hemodialysis; HDF, hemodiafiltration.

After ACM entrance, a significant decrease in Hb fluctuation (from 0.95 ± 0.41 g/dl to 0.83 ± 0.33 g/dl; $P < 0.001$) can be observed, together with a significant increase in the percentage of patients having at least two-thirds of their Hb values within target range (from 64.5% to 84.1%; $P < 0.001$).

These results are emphasized when focusing on the ACM-compliant group. For these patients, the decrease in Hb fluctuation was even greater (from 0.97 ± 0.41 g/dl to 0.80 ± 0.29 g/dl; $P < 0.001$) as was the increase in the percentage of patients with most of their Hb values within target range (from 65.2% to 89.5%; $P < 0.001$). Interestingly, whereas the median monthly darbepoetin doses decreased in the observation phase, the dose variability (measured as the absolute difference between 2 subsequent monthly doses [median absolute delta darbepoetin doses in Table 4]) tended to increase.

Figure 1 shows the distribution of Hb SDs in the study phases in (i) all patients and (ii) ACM-compliant patients. The change in skewness and kurtosis and the distribution shift confirm that Hb variability decreased after ACM deployment.

Table 4 | Results of the patient longitudinal analysis

	Control phase	Observation phase	*P*-value
All patients (*N* = 383)			
Anemia outcomes			
Hb SD, g/dl, mean ± SD	0.95 ± 0.41	0.83 ± 0.33	<0.001[a]
Patients with >66.6% Hb within target range, no. (%)	247 (64.5)	322 (84.1)	<0.001[b]
Median darbepoetin doses, μg, median (IQR)	40.00 (68.75)	30.00 (70.00)	<0.001[c]
Median absolute delta darbepoetin doses,[e] μg, median (IQR)	10.00 (25.00)	20.00 (40.00)	0.03[c]
Adverse events			
Patients with cardiovascular events, no. (%)	82 (21.5)	54 (14.1)	0.01[b]
Cardiovascular events (incidence/ 1000 patient-years)	296.73	248.91	0.11[d]
Hospitalization days (incidence/ 1000 patient-years)	3488.63	3768.45	0.006[d]
Patients with transfusion events, no. (%)	9 (2.3)	3 (0.8)	0.14[b]
Transfusion events (incidence/ 1000 patient-years)	55.46	8.68	<0.001[d]
ACM-compliant patients (*n* = 313)			
Anemia outcomes			
Hb SD, g/dl, mean ± SD	0.97 ± 0.41	0.80 ± 0.29	<0.001[a]
Patients with >66.6% Hb within target range, no. (%)	204 (65.2)	280 (89.5)	<0.001[b]
Median darbepoetin dose, μg, median (IQR)	40.00 (80.00)	20.00 (70.00)	0.001[c]
Median absolute delta darbepoetin dose, μg, median (IQR)	10.00 (25.00)	10.00 (40.00)	0.24[c]
Adverse events			
Patients with cardiovascular events, no. (%)	64 (20.4)	39 (12.5)	0.009[b]
Cardiovascular events (incidence/ 1000 patient-years)	276.36	191.15	0.002[d]
Hospitalization days (incidence/ 1000 patient-years)	3319.69	3348.67	0.42[d]
Patients with transfusion events, no. (%)	7 (2.2)	0 (0)	0.02[b]
Transfusion events (incidence/ 1000 patient-years)	54.59	0	<0.001[d]

The top of the table shows results for the larger group of patients, whereas the bottom part shows results for patients having at least two-thirds of their ACM suggestions confirmed (ACM-compliant patients).
ACM, Anemia Control Model; Hb, hemoglobin; IQR, interquartile range.
[a]Paired *t* test.
[b]Fisher exact test.
[c]Wilcoxon test.
[d]Poisson exact test.
[e]Absolute delta darbepoetin doses: the absolute difference in 2 subsequent darbepoetin doses.

Figure 2 shows the clinical evolution for a sample patient, including Hb concentrations (top panel) and corresponding ESA interventions (bottom panel). The vertical dotted line represents ACM introduction; each Hb measurement is paired with the ESA dose suggested by the ACM at that point in time. Green circles represent Hb values resulting from a confirmed suggestion. It can be noted that, after ACM entrance, Hb values were stabilized, especially when suggestions were accepted.

The occurrence of adverse events also tended to decrease after ACM introduction, particularly in the ACM-compliant group; hospitalization days per 1000 patient-years increased in the observation phase, but not in the ACM-compliant group.

DISCUSSION

In this international retrospective analysis involving 3 countries, we performed a direct comparison between standard anemia management (by expert nephrologists following established best clinical practices) and ACM-supported anemia management. The purpose of the ACM is to facilitate the physicians' decision making in devising a personalized anemia therapy for their patients.

At the facility level, after ACM introduction, the percentage of Hb values within target range increased from 70.6% to 76.6%. As this figure was computed considering all Hb measurements for our population, including ACM-ineligible patients, it serves as a measure of the global impact of ACM introduction. Even if this 6% increase cannot be completely credited to ACM intervention, it still provides a first, rough indication of its positive effect. The ACM contribution can be assessed more closely considering only Hb values resulting from accepted recommendations; in this case, the on-target percentage increases to 83.2%, indeed suggesting a net benefit of ACM use for the improvement of Hb outcomes. When suggestions are accepted, both Hb values above and below target range are reduced (Table 2).

Our results show that the Hb outcome is consistently worse when ACM suggestions are rejected: the on-target percentage is 67.9%, with an Hb distribution comparable to that found in the control phase. In this study, physicians rejected a suggestion in 13.8% of the cases. Even though EuCliD, the clinical information system of the Fresenius Medical Care clinic network,[25] keeps track of the physicians' feedback whenever a recommendation is not accepted, it is virtually impossible to determine, case by case, whether the ACM or the physician identified the optimal ESA dose in case of disagreement. Still, the improvement on Hb outcome when considering accepted recommendations is large enough to support ACM effectiveness.

Table 2 also shows that, not surprisingly, outcomes are worse when no ACM suggestion could be made (28.9% of the data): this is the case for newly admitted patients (no past information), patients who had recently received transfusions, and patients who missed several dialysis treatments (e.g., because of hospitalization, vacation leave). For these patients, the lack of a complete clinical history in the preceding 3 months prevents the model from computing a reliable prediction. The particular conditions of these patients justify the observed pattern of outcomes. It should be noted that the overall figures include the contribution of this set of patients and therefore might mitigate the positive effect of the ACM.

The patient-level analysis (Table 4) confirmed, with stronger figures, the increase of on-target Hb values (89.5% of the ACM-compliant patients had at least two-thirds of their Hb on target in the observation phase vs. 65.2% in the control

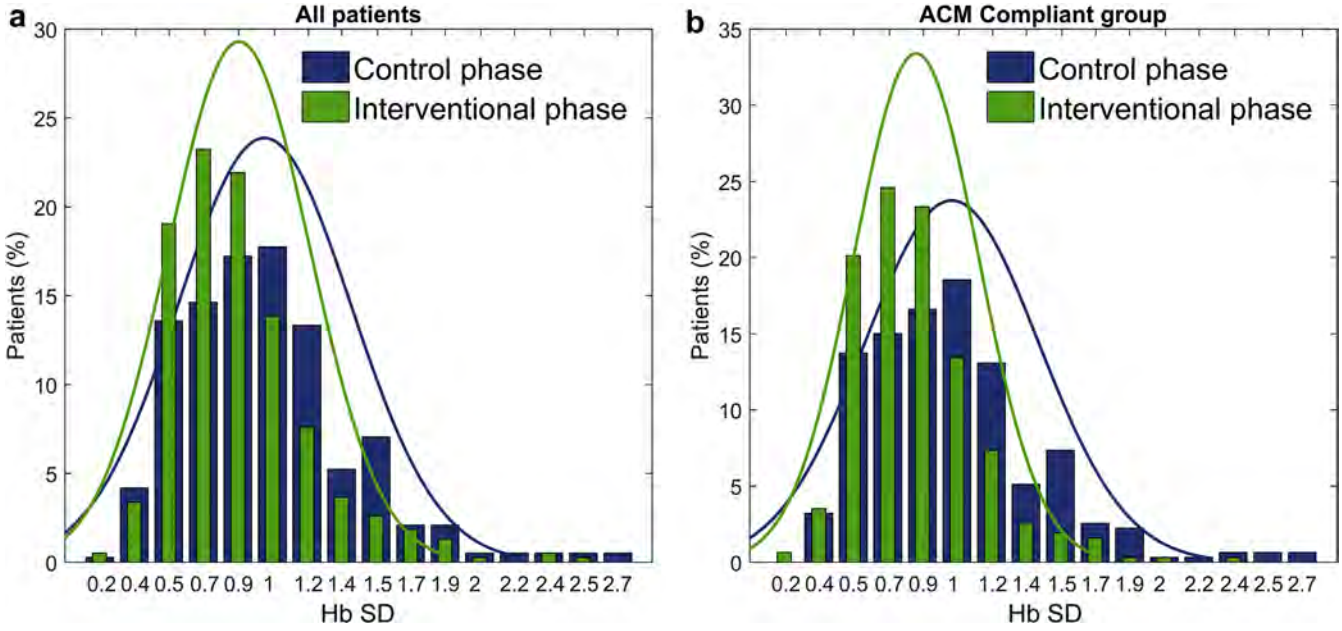


Figure 1 | Histograms of hemoglobin (Hb) SDs before and after Anemia Control Model (ACM) introduction. (**a**) Histograms related to all patient data. (**b**) Histograms related to the ACM-compliant subgroup.

phase) and allowed us to estimate the effect of the ACM on Hb variability as follows: the intrapatient Hb SD decreased after ACM introduction and even more so when considering the ACM-compliant group (from 0.97 ± 0.41 g/dl to 0.80 ± 0.29 g/dl; $P < 0.001$). These results suggest a positive impact of the ACM in reducing Hb fluctuation. Interestingly, a reduction in Hb fluctuation is associated with more frequent changes in ESA dosing, although this effect, in the

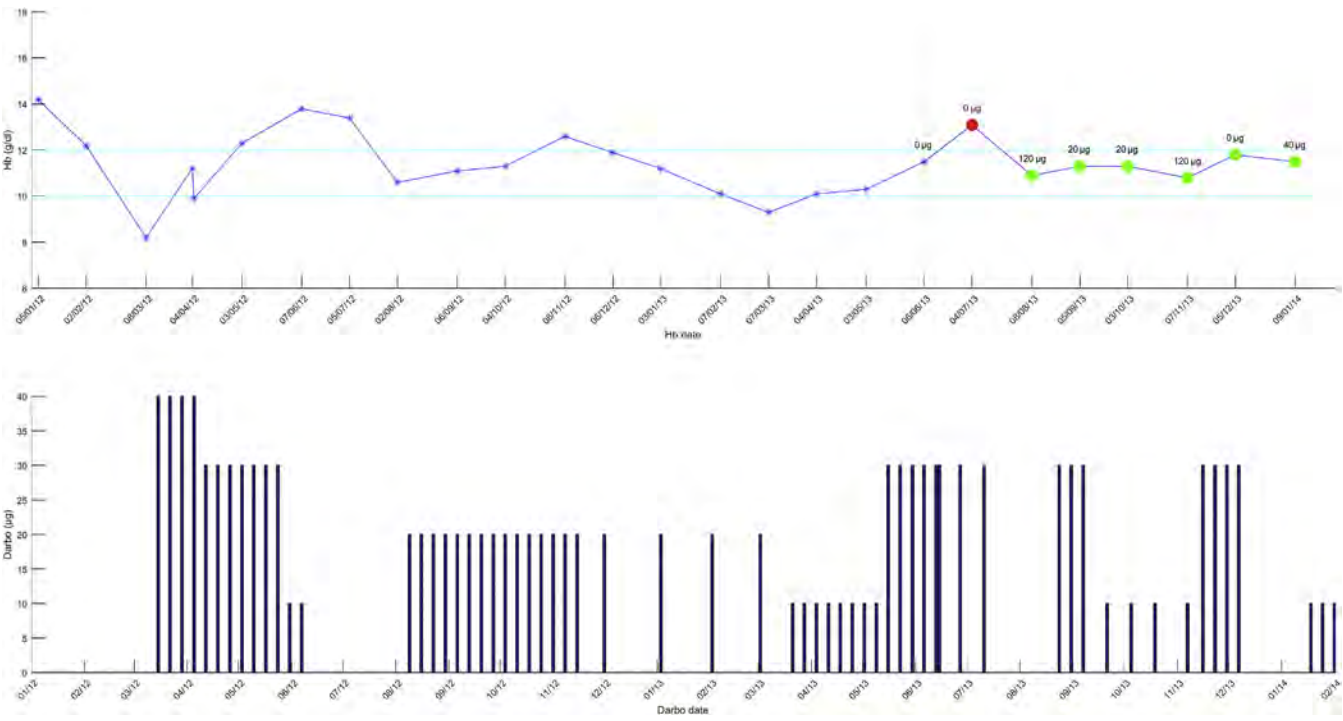


Figure 2 | Hemoglobin (Hb) series and erythropoietic-stimulating agent (ESA) administrations for a sample patient. (*Top*) Hb temporal evolution for a sample patient is plotted. The vertical dotted line represents the time of Anemia Control Model (ACM) introduction; green circles identify Hb values resulting from confirmed suggestions. (*Bottom*) ESA administrations. The first ACM suggestion was rejected (as indicated by the red circle on the resulting Hb value), whereas all subsequent suggestions were accepted. Correspondingly, a reduction in Hb cycling can be observed. Darbo, darbepoetin.

ACM-compliant group, is nonsignificant; however, it should be noted that, although frequently changing ESA doses as a reaction to new Hb tests may lead to cycling,[11] the dose variability induced by a predictive model that anticipates future changes in Hb level is intended to reduce it.

Darbepoetin consumption decreased in the observation phase at both levels of analysis; again, this decrease was larger when the ACM suggestions were accepted (Table 2). Importantly, the reduction in ESA consumption is not associated with higher iron dosing regimens. It appears that the ACM was able to propose effective ESA doses that could achieve the therapeutic goals (in terms of Hb target and variability) while optimizing consumption and, thus, possibly reducing both the potential hazards of high-dosing ESA and i.v. iron and their related costs.[26–28]

This is the first large-scale observational study conducted in an unselected population of prevalent HD patients comparing the standard care of anemia treatment with guided and supported care relying on a predictive model. The following other groups have reported on similar approaches: Brier *et al.*[20] performed a double-blind, randomized, controlled trial to compare anemia treatment based on standard protocols with recommendations provided by an artificial neural network–based predictive model; Gaweda *et al.*[22] described a similar clinical trial testing an extension of their previous model. In this latter study, the on-target percentage in the treatment group was found to be 72.5% (10.7% greater than in the control group). However, both studies were performed with a limited number of subjects (60–62 subjects), a tenth of the population considered in our study. A 10% increase (from 56% to 66%) in the on-target percentage was also obtained by Lines *et al.*[21] in a larger cohort of patients ($n = 214$), using a simple model relying on linear projection of Hb values; this study also reported a reduction in interpatient Hb variability from 1.46 g/dl to 1.25 g/dl. None of these studies resulted in decreased ESA consumption. To our knowledge, the largest study in terms of population size (300–342 patients) was the study by McCarthy *et al.*,[23] who tested a model of the dynamics of erythrocyte production based on coupled, nonlinear, first-order differential equations. When targeting the Hb range of 10.0 g/dl to 11.9 g/dl, the percentage of on-target patients reached 68% from a baseline of 57%; a significant reduction in monthly ESA doses was also reported (from 300 μg to 157 μg), although these consumption values are still much higher than those achieved in our study. McCarthy *et al.*[23] also analyzed Hb SD in a small selected subset of patients (iron replete, with no hospitalizations longer than 1 day; $n = 58$) and found a decrease in intrapatient variability (from 0.86 g/dl to 0.59 g/dl). Taken together, these studies show that the use of a predictive model for suggesting appropriate ESA doses can improve Hb outcomes by at least 10%.

Our work confirms that a decision support system based on predictive modeling can achieve similar improvements, even in a large, unselected population of patients. In addition, it provides clinical proof of the beneficial effects of deploying the ACM in everyday practice as part of the nephrological routine practice. From a clinician's perspective, our results also look more promising and appealing: when ACM recommendations were accepted, the on-target percentage at the facility level reached 83.2%, and the percentage of patients having most of their Hb values on target increased by 24.3%. These results show that the implementation of the ACM in a large dialysis network was feasible, beneficial for the patients, and helpful for the physicians; improved anemia outcomes; and optimized ESA consumption. The rate of recommendation acceptance in this pilot study was already satisfying (>86%), and it is expected to increase as clinical acceptance and trust in the ACM progress.

This study has some limitations. It is not a randomized or blinded controlled trial, and the evaluation of adverse events, although favorable, would need further, careful scrutiny. Now that the ACM has been implemented in clinical routine, we will extend the follow-up period to assess outcomes, and short-acting ESAs will be included in the model to generalize its use to our entire HD network, with a more specific cost-effectiveness focus. Another limitation is that the ACM could not provide a suggestion in 28.9% of the cases because its predictive model needs to be fed with a consistent 3-month clinical history, with coverage of previous ESA administrations as precise as possible. It should be noted that ACM ineligibility is indeed a temporary condition, and therefore a patient who is currently ineligible will be able to receive a suggestion later on. Nonetheless, we intend to improve the model for reducing such ACM-ineligibility conditions.

In summary, our findings confirm that tighter control of anemia can be achieved by means of ACM support. The ACM provides a feedback control loop closer to physiological

Table 5 | Parameters used by the hemoglobin predictive model

Parameter	Time
Sex	At admission
Height	At admission
Previous delta hemoglobin	Past month
Ferritin	Latest measurement
Transferrin saturation index	Latest measurement
Albumin	Latest measurement
Phosphate	Latest measurement
Leukocytes	Latest measurement
C-reactive protein	Latest measurement
Mean corpuscular volume	Latest measurement
Mean corpuscular hemoglobin	Latest measurement
Calcium	Latest measurement
Sodium	Latest measurement
Potassium	Latest measurement
Dry body weight	Latest measurement
Predialysis weight	Latest measurement
OCM Kt/V	Latest measurement
Darbepoetin doses	Past 90 days
Iron doses	Past 90 days

Sex and height are taken at admission; for laboratory tests, weight, and kt/V, the latest value is used. All erythropoietic-stimulating agent and iron doses in the past 90 days are considered. The change in hemoglobin with respect to the previous month is also computed.
OCM, online clearance monitoring.

regulation and is likely to provide a buffer period of ESA dose adjustment, reducing prescription patterns that may react too quickly to Hb concentration changes. The ACM may be potentially adapted and used in the management of other chronic diseases in which anemia requires an active treatment.

METHODS

Anemia Control Model

The ACM is a decision support system based on an artificial intelligence core intended to support nephrologists in making decisions related to anemia therapy in HD patients.

The predictive model at the ACM core is a machine learning model that was trained on a set of almost 170,000 clinical records and whose ability to generalize to unseen examples was tested on >40,000 records; records were extracted from the clinical information system EuCliD.[25] The artificial neural network was trained to predict the change in Hb value occurring during a period of a month, based on the parameters listed in Table 5. The mean absolute prediction error of the model on Hb concentration is 0.59 g/dl.

The dose-selection algorithm of the ACM has been designed in adherence to clinical guidelines for anemia treatment in HD patients; by simulating on the predictive model the effect of different drug doses on the predicted Hb, the algorithm selects the dose that would move Hb to the target interval while avoiding excessive Hb decreases or increases.

The ACM is triggered whenever a new Hb value is recorded, which is typically once per month. Patients are considered temporarily ineligible for a suggestion if, at ACM run time:

(i) They are younger than 18 years of age.
(ii) They were admitted to the facility in the previous 90 days.
(iii) They received a transfusion during the previous 90 days.
(iv) They did not receive a minimum number of dialysis treatments in the facility during the previous 90 days (at least 27 of the expected 39 treatments) due to hospitalization or vacation.

In fact, for the model to elaborate a reliable prediction, clinical data over the past 90 days are needed, and, in particular, information regarding drugs administered at treatment should be nearly complete. ESA therapy for ACM-ineligible patients needs to be closely assessed by their attending physician. It is important to stress that ACM only provides therapy *recommendations*: physicians are required to evaluate the validity of recommendations on an individual patient basis and to decide whether to accept them or to formulate a different drug prescription. In the NephroCare (NephroCare ensures care of CKD patients within the Fresenius Medical Care network) clinics where the ACM has been deployed, a dedicated module is available in EuCliD, where drug suggestions are presented to the physician and turned into actual prescriptions in case of acceptance; when a suggestion is rejected, the physician is asked to provide a reason for such a decision. Updated clinical data are fed to the ACM by means of an automatic interface module with EuCliD. Although the ACM is not publicly available, its distribution to third parties is currently under consideration.

Study design and statistical analyses

This 2-year study (June 2012–May 2014) consisted of 2 periods. The first period (June 2012–May 2013) was considered as the control phase (no ACM support); data were collected retrospectively. The second period (June 2013–May 2014) was considered the observation phase. The ACM was deployed in June 2013 in 3 NephroCare clinics: Motol Prague, Czech Republic; Cartagena and San Pedro del Pinatar, Spain; and Lumiar, Portugal.

The patients treated in the Fresenius Medical Care network are informed that their data, collected through EuCliD, might be used for scientific purposes and are asked to sign a consent form if they agree. All data were anonymized when transferred to the calculation center and were disclosed only to their attending physician. The use of the ACM in clinics was approved by the company's Medical Board as part of a quality improvement project.

The effects of the ACM were assessed both at the dialysis facility level and at the patient level, using traditional key indicators of anemia treatment as outcomes.

At the dialysis facility level, all patients having at least 1 HD treatment and 1 Hb measurement at 1 of the study clinics during the control or observation phase were selected, provided they were at least 18 years old. The percentage of Hb values on target and the median ESA administered dose (expressed as dose per patient per kilogram per month) were considered as primary outcomes. We consider a Hb value to be on target if it falls in the range of 10 to 12 g/dl or if it is >12 g/dl in the absence of any ESA therapy (that is, the patient did not receive an ESA in the 35 days before the Hb measurement). Outcomes were computed for the whole population as a measure of the global impact of the ACM. Furthermore, a subanalysis was performed focusing on confirmed recommendations; that is, we isolated Hb results and drug doses resulting from an accepted ACM suggestion. Adverse events (mortality, cardiovascular events, hospitalizations, and transfusions) were also analyzed. Cardiovascular events were identified by extracting the entries in EuCliD with an International Classification of Diseases code in the range of I00 to I99, except for I80 to I89 codes (diseases of veins, lymphatic vessels, and lymph nodes). A patient was categorized as experiencing a cardiovascular event in the considered period if he or she died of a cardiovascular cause, experienced the occurrence of a new pathologic cardiac condition, or was hospitalized for cardiovascular reasons.

For the patient-level analysis, among the cohort of patients defined previously, we selected only the HD patients who had at least 6 consecutive monthly Hb measurements at 1 of the study clinics in the control phase and another 6 in the observation phase (thus, at least 12 consecutive tests). This requirement is motivated by the need to have a relevant number of consecutive Hb measurements for each patient, so that the Hb fluctuation over time can be evaluated. For this analysis, therefore, each patient can contribute to the 2 phases with data series of unequal length. Individual Hb fluctuation was estimated from the Hb SD over the control/observation phase, and supplemented with the percentage of Hb values on target. The same parameters were evaluated also for the ACM-compliant subpopulation; we included a patient in this group if, given the set of recommendations produced for that patient, the proportion of accepted suggestions was at least two thirds (~66%).

A subanalysis of patients grouped according to their vascular access was also performed (see Supplementary Appendix).

Analyses were performed using MATLAB (MathWorks; Natick, Massachusetts) and R software (R Project, Vienna, Austria). The Student *t* test was performed to compare the values of normally distributed data, whereas the Wilcoxon-Mann-Whitney test was applied to compare the values of nonnormally distributed data. The Fisher exact test was used for proportions and the Poisson exact test for rate parameters. When applicable, data are expressed as mean ± SD or median (interquartile range). For all tests, a P value <0.05 was considered statistically significant.

DISCLOSURE

All authors, with the exception of MM, are Fresenius Medical Care employees and may have stock options. MM collaborates with Fresenius Medical Care.

SUPPLEMENTARY MATERIAL

Supplementary Appendix.

Supplementary material is linked to the online version of the paper at www.kidney-international.org.

REFERENCES

1. Barrett BJ, Fenton SS, Ferguson B, et al. Clinical practice guidelines for the management of anemia coexistent with chronic renal failure. Canadian Society of Nephrology. *J Am Soc Nephrol*. 1999;10(Suppl 13): S292–S296.
2. National Kidney Foundation. NKF-K/DOQI clinical practice guidelines for anemia of chronic kidney disease: update 2000. *Am J Kidney Dis*. 2001;37(Suppl 1):S182–S238.
3. Locatelli F, Aljama P, Bárány P, et al. Revised European best practice guidelines for the management of anaemia in patients with chronic renal failure. *Nephrol Dial Transplant*. 2004;19(Suppl 2):ii1–ii47.
4. National Kidney Foundation. KDOQI clinical practice guidelines and clinical practice recommendations for anemia in chronic kidney disease. *Am J Kidney Dis*. 2006;47(Suppl 3):S11–S145.
5. Drüeke TB, Locatelli F, Clyne N, et al. Normalization of hemoglobin level in patients with chronic kidney disease and anemia. *N Engl J Med*. 2006;355:2071–2084.
6. Singh AK, Szczech L, Tang KL, et al. Correction of anemia with epoetin alfa in chronic kidney disease. *N Engl J Med*. 2006;355:2085–2098.
7. Pfeffer MA, Burdmann EA, Chen CY, et al. A trial of darbepoetin alfa in type 2 diabetes and chronic kidney disease. *N Engl J Med*. 2009;361:2019–2032.
8. Phrommintikul A, Haas SJ, Elsik M, Krum H. Mortality and target haemoglobin concentrations in anaemic patients with chronic kidney disease treated with erythropoietin: a meta-analysis. *Lancet*. 2007;369:381–388.
9. Pisoni RL, Bragg-Gresham JL, Young EW, et al. Anemia management and outcomes from 12 countries in the Dialysis Outcomes and Practice Patterns Study (DOPPS). *Am J Kidney Dis*. 2004;44:94–111.
10. Berns JS, Elzein H, Lynn RI, et al. Hemoglobin variability in epoetin-treated hemodialysis patients. *Kidney Int*. 2003;64:1514–1521.
11. Lacson E Jr, Ofsthun N, Lazarus JM. Effect of variability in anemia management on hemoglobin outcomes in ESRD. *Am J Kidney Dis*. 2003;41:111–124.
12. Ebben JP, Gilbertson DT, Foley RN, Collins AJ. Hemoglobin level variability: associations with comorbidity, intercurrent events, and hospitalizations. *Clin J Am Soc Nephrol*. 2006;1:1205–1210.
13. Yang W, Israni RK, Brunelli SM, et al. Hemoglobin variability and mortality in ESRD. *J Am Soc Nephrol*. 2007;18:3164–3170.
14. Fishbane S, Besarab A. Mechanism of increased mortality risk with erythropoietin treatment to higher hemoglobin targets. *Clin J Am Soc Nephrol*. 2007;2:1274–1282.
15. Schiller B, Doss S, De Cock E, et al. Costs of managing anemia with erythropoiesis-stimulating agents during hemodialysis: a time and motion study. *Hemodialysis Int*. 2008;12:441–449.
16. Brier ME, Gaweda AE. Predictive modeling for improved anemia management in dialysis patients. *Curr Opin Nephrol Hypertens*. 2011;20: 573–576.
17. Nichols B, Shrestha RP, Horowitz J, et al. Simplification of an Erythropoiesis Model for Design of Anemia Management Protocols in End Stage Renal Disease. *Conf Proc IEEE Eng Med Biol Soc*. 2011;2011:83–86.
18. Martínez-Martínez JM, Escandell-Montero P, Barbieri C, et al. Prediction of the hemoglobin level in hemodialysis patients using machine learning techniques. *Comput Methods Programs Biomed*. 2014;117:208–217.
19. Escandell-Montero P, Chermisi M, Martínez-Martínez JM, et al. Optimization of anemia treatment in hemodialysis patients via reinforcement learning. *Artif Intell Med*. 2014;62:47–60.
20. Brier ME, Gaweda AE, Dailey A, et al. Randomized trial of model predictive control for improved anemia management. *Clin J Am Soc Nephrol*. 2010;5:814–820.
21. Lines SW, Lindley EJ, Tattersall JE, Wright MJ. A predictive algorithm for the management of anaemia in haemodialysis patients based on ESA pharmacodynamics: better results for less work. *Nephrol Dial Transplant*. 2012;27:2425–2429.
22. Gaweda AE, Aronoff GR, Jacobs AA, et al. Individualized anemia management reduces hemoglobin variability in hemodialysis patients. *J Am Soc Nephrol*. 2014;25:159–166.
23. McCarthy JT, Hocum CL, Albright RC, et al. Biomedical system dynamics to improve anemia control with darbepoetin alfa in long-term hemodialysis patients. *Mayo Clin Proc*. 2014;89:87–94.
24. Barbieri C, Mari F, Stopper A, et al. A new machine learning approach for predicting the response to anemia treatment in a large cohort of end stage renal disease patients undergoing dialysis. *Comput Biol Med*. 2015;61:56–61.
25. Marcelli D, Kirchgessner J, Amato C, et al. EuCliD (European Clinical Database): a database comparing different realities. *J Nephrol*. 2001;14(Suppl 4):S94–S100.
26. Rostoker G, Griuncelli M, Loridon C, et al. Hemodialysis-associated hemosiderosis in the era of erythropoiesis-stimulating agents: a MRI study. *Am J Med*. 2012;125:991–999.e1.
27. Fishbane S, Mathew A, Vaziri ND. Iron toxicity: relevance for dialysis patients. *Nephrol Dial Transplant*. 2014;29:255–259.
28. Rostoker G, Griuncelli M, Loridon C, et al. Reassessment of iron biomarkers for prediction of dialysis iron overload: An MRI Study. *PLoS One*. 2015;10:e0132006.