Effect of iron source on the alleviation of nutritional anaemia in common sole (*Solea solea*)

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Diapositiva 1

KJ21 abstract number 22866
Kals, Jeroen; 18/08/2016

Effect of iron source on the alleviation of nutritional anaemia in common sole

- Introduction
- Hypotheses
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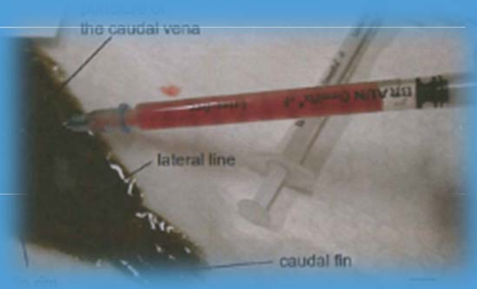
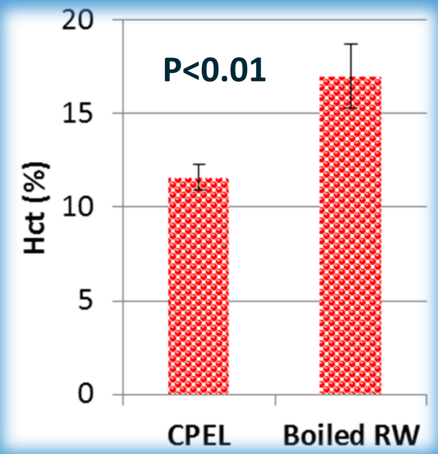
Introduction



- Sole fed commercial pellets (CPEL) suffer from a nutritional anaemia
- An iron (Fe) deficiency is a common cause

- Intake of Fe can be eliminated
- Intake Fe in sole fed boiled ragworm ≈ sole fed CPEL, but Hct sign. higher



- Yet, bioavailability of iron cannot be excluded

Kals et al. 2015

Introduction

- Bioavailability iron depends on form (e.g. heme *vs.* non heme)

- Absorption heme *vs.* non heme differs as heme
    - has its own pathway
    - independent of pH
    - immune for antagonists (e.g. Ca^{2+})

KJ22

- Fe in CPEL is mainly non heme from premix or damaged heme

- Alkaline character of sole's intestine might hamper absorption of non heme iron, leading to an iron deficiency?

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KJ22 non heme requires reduction of Fe3+ to Fe2+, and hence, an acidic environment
Kals, Jeroen; 19/08/2016

Hypotheses

1. nutritional anaemia in sole fed CPEL is due to an iron deficiency
2. assumed Fe deficiency is due to poor absorption of Fe
3. increase in absorption due to a higher bioavailability of heme or iron chelates will alleviate anaemia in sole
4. haematocrit (Hct) and haemoglobin (Hb) are expected to follow iron absorption patterns

- We also estimated absorption of Cu, Co, Cr, Mn, Mo & Zn to evaluate interaction between iron source and other minerals

Material and Methods

- Sole raised on CPEL and anaemic at start

- Dietary treatments:
    - Fe sulphate
    - Fe methionate
    - Fe proteinate
    - Haemoglobin

- Feeding: restricted, equal feeding levels for all diets
- Tank experimental unit (n=3, 10 fish.tank^{-1})
- Duration 23 days, sampling at start & day 23

Material and Methods

Formulations

Dietary Fe

sources 64%
basal diet 36%

Marker: Yttrium

Processing:

Diet Code	A	B	C	D
Iron source	*Iron sulphate*	*Heme*	*Iron proteinate*	*Iron methionate*
Basal ingredients (%)	57.2	57.2	57.2	57.2
Test ingredients (%):				
Caseine	16.38	9.00	16.35	16.35
Pea protein conc.	13.00	12.70	13.00	13.00
Corn gluten	12.50	13.00	12.50	12.50
L-threonine	0.35	0.40	0.35	0.35
DL-methionin	0.16	0.30	0.16	0.12
L-iso leucin	0.00	0.30	0.00	0.00
Corn Starch	0.27	0.86	0.28	0.34
Yttrium oxide	0.02	0.02	0.02	0.02
Iron sources				
Bovine hemoglobin	--	6.200	--	--
Iron sulfate hydrate (20%)	0.101	--	--	--
Iron proteinate	--	--	0.127	--
Iron methionate	--	--	--	0.106
Check	100.00	100.00	100.00	100.00

Cold extrusion was used to keep iron sources in their native state

Material and Methods

Formulations

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- isonitrogenous
- isoenergetic
- equal in

*AA comp. KJ12
*total iron KJ9
*calcium KJ11
*taurine
*vitamin B12

content.

Iron source		*Iron sulphate*	*Heme*	*Iron proteinate*	*Iron methionate*
DM	($g.kg^{-1}$)	909	913	920	917
ASH	($g.kg^{-1}.dm$)	94	92	94	94
CP	“”	661	655	669	661
EE	“”	130	122	130	127
CF	“”	6.6	6.6	5.4	5.5
NFE	“”	17.9	37.9	22.2	30.4
Check	----	909	913	920	917
GE	($MJ.kg^{-1}$)	21.2	21.1	21.5	21.3
CP/GE	----	31.1	31.0	31.1	31.0

Vit B12	($ug.kg^{-1}.dm$)	1254	1150	959	1075
Fe	($mg.kg^{-1}.dm$)	355	323	299	349
Ca	($g.kg^{-1}.dm$)	8	8	8	8
Na	“”	19	19	19	19
K	“”	6	6	6	6
Mg	“”	2	2	2	2
Cu	($mg.kg^{-1}.dm$)	12	12	12	12
Co	“”	2	2	2	2
Cr	“”	5	5	4	5
Mn	“”	36	35	38	38
Mo	“”	1	1	1	1
Zn	“”	119	115	121	120

[1]DM=Dry matter, ASH=Ash, CP=Crude Protein, EE=Ether extract, CF=Crude Fibre, NFE=Nitrogen Free Extract, GE=Gross Energy

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KJ9 iron content was formulated to be equal to the iron content in ragworm (352 mg.kg-1.dm).
Kals, Jeroen; 18/08/2016

KJ11 Taurine can affect haematological parameters
Kals, Jeroen; 18/08/2016

KJ12 Individual amino acids were added to compensate for the amino acid content of the iron sources, especially of the heme iron.
Kals, Jeroen; 18/08/2016

Analyses

- Hct: centrifuging blood (5 min, 5000g)
- Hb: colorimetric (van Kampen & Zijlstra 1961)
- Minerals:
  - ICP-AES (e.g. Cu, Fe, Mn, Zn) KJ25
  - ICP-MS (e.g. Co & Mo) KJ26

- Calculation apparent absorption coefficient (AAC)

$$AAC_{mineral\ (x)} = 100 - (100 * \left(\frac{Yttrium_{diet}}{Yttrium_{faeces}}\right) * \left(\frac{mineral\ (x)_{faeces}}{mineral\ (x)_{diet}}\right)$$

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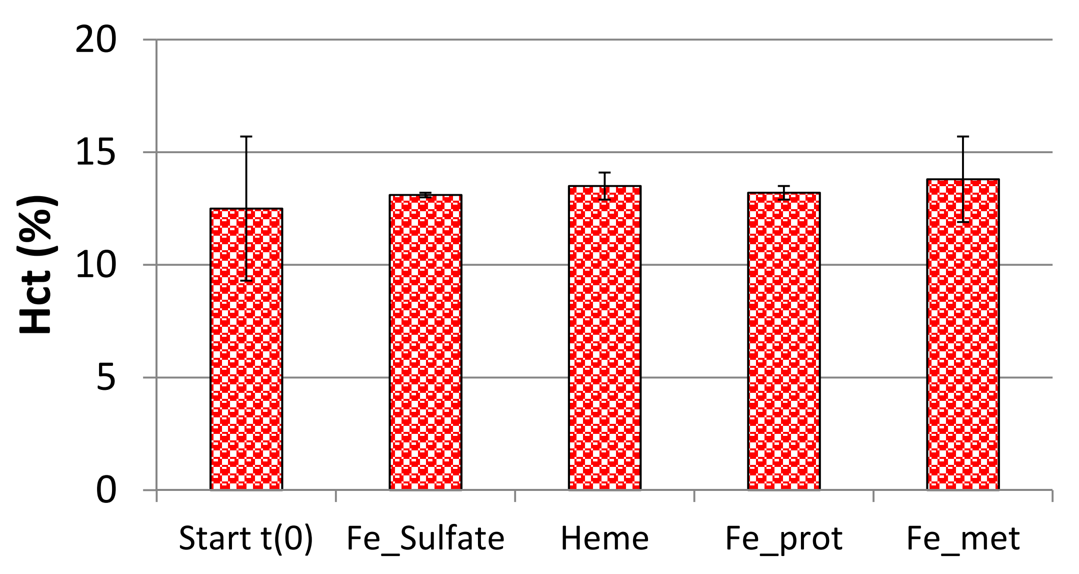
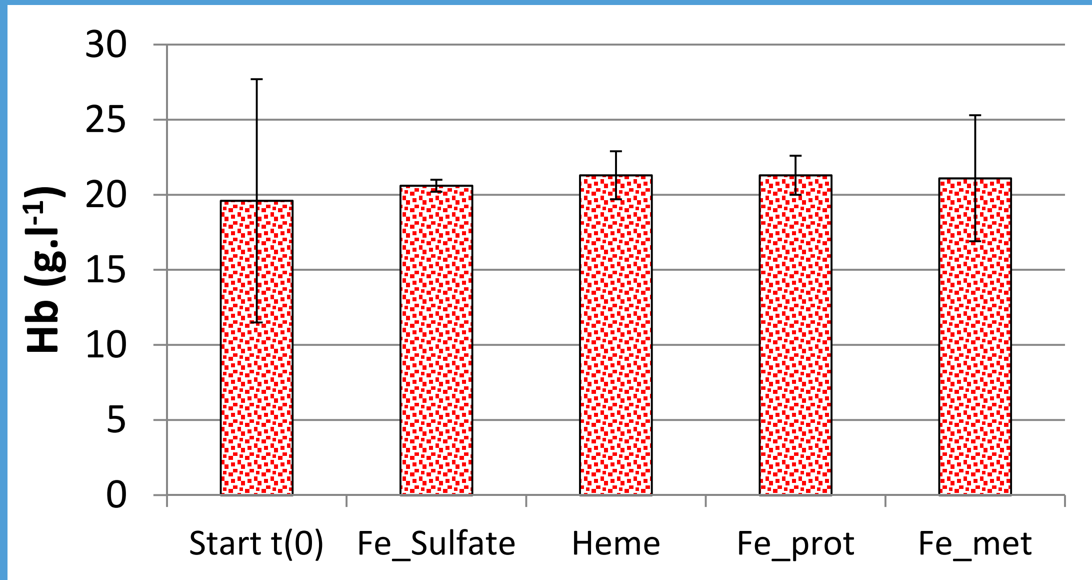
KJ25 inductively coupled plasma-atomic emission spectrometry
Kals, Jeroen; 18/08/2016

KJ26 Inductively Coupled Plasma Mass Spectrometry
Kals, Jeroen; 18/08/2016

Results Hct and Hb



- Hct & Hb levels not different from t(0)
- Hct & Hb levels not affected by source



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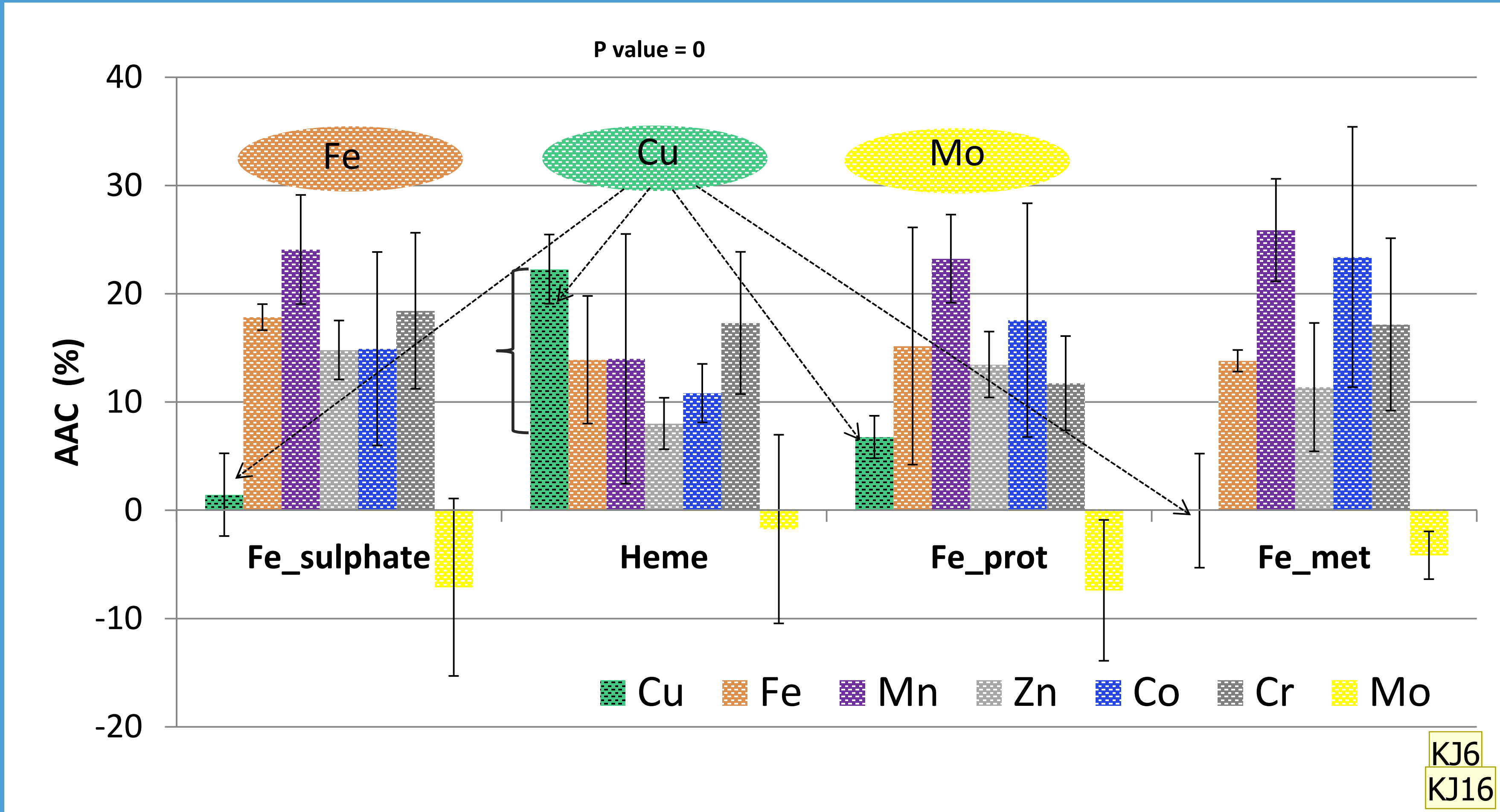
Results AAC's



- AAC Fe, Mn, Zn, Co, Cr, Mo, except Cu not affected by source

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KJ6 Yet, the iron absorption was high for all sources.

The AAC of Cu was 15-22% higher in sole fed the diet with heme.

Kals, Jeroen; 18/08/2016

KJ16 The negative ADC's of Mo are most likely a result from the relatively high presence of Mo in seawater and the fact that marine fish must drink to keep their water balance in order

Kals, Jeroen; 27/07/2016

Discussion

- Despite the high absorption of iron, fish stayed anaemic independent of iron source

  ->Implies nutritional anemia is not an iron deficiency anaemia

- The alkaline character of the sole's intestine would hamper the ability to absorb non heme iron

  ->Iron absorption in relation to iron source seems not a limiting factor

Discussion

- Hypotheses

  1) nutritional anaemia in sole fed CPEL is due to an iron deficiency anaemia,

  2) NA is caused by inadequate absorption of iron,

  3) Rise of iron absorption due to a higher bioavailability of heme and/or iron chelates alleviates anaemia in sole

  all have to be rejected!

  4) Hence, hypothesis 4: Hct & Hb levels, follow iron absorption patterns could not be tested

Discussion

- AAC of Cu was high using heme
- none heme Fe & Cu, need DMT1
- Heme is not claiming DMT1 capacity

-> Pos. effect of heme on Cu can be explained by a reduced competitive binding of Cu and non heme Fe claiming DMT1

- Cu is crucial for iron uptake
- AAC of Cu is high in sole fed heme, yet Hct & Hb did not respond

-> nutritional anaemia is not a Cu deficiency anaemia

Conclusions

- Iron absorption is high & independent of iron source

- Heme iron increases absorption of copper

- High absorption of Fe & Cu in sole fed heme did not affect Hct and Hb, suggesting the nutritional anaemia in sole is

  -> not an iron

  -> nor a Cu deficiency anaemia

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Thank you!

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Questions?

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