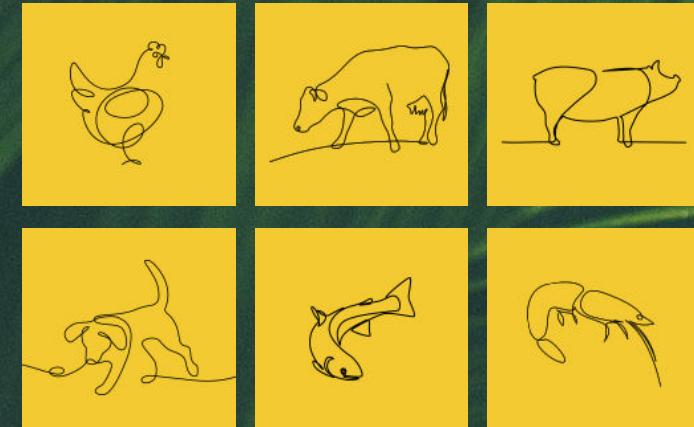


Natu-B4

Product presentation



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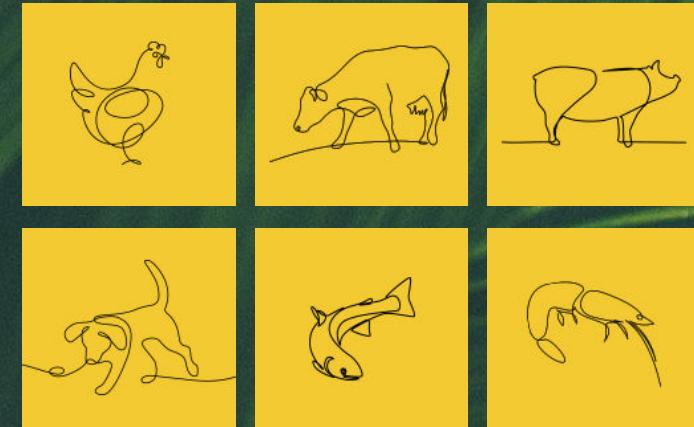


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Natu-B4

Concept and mode of action



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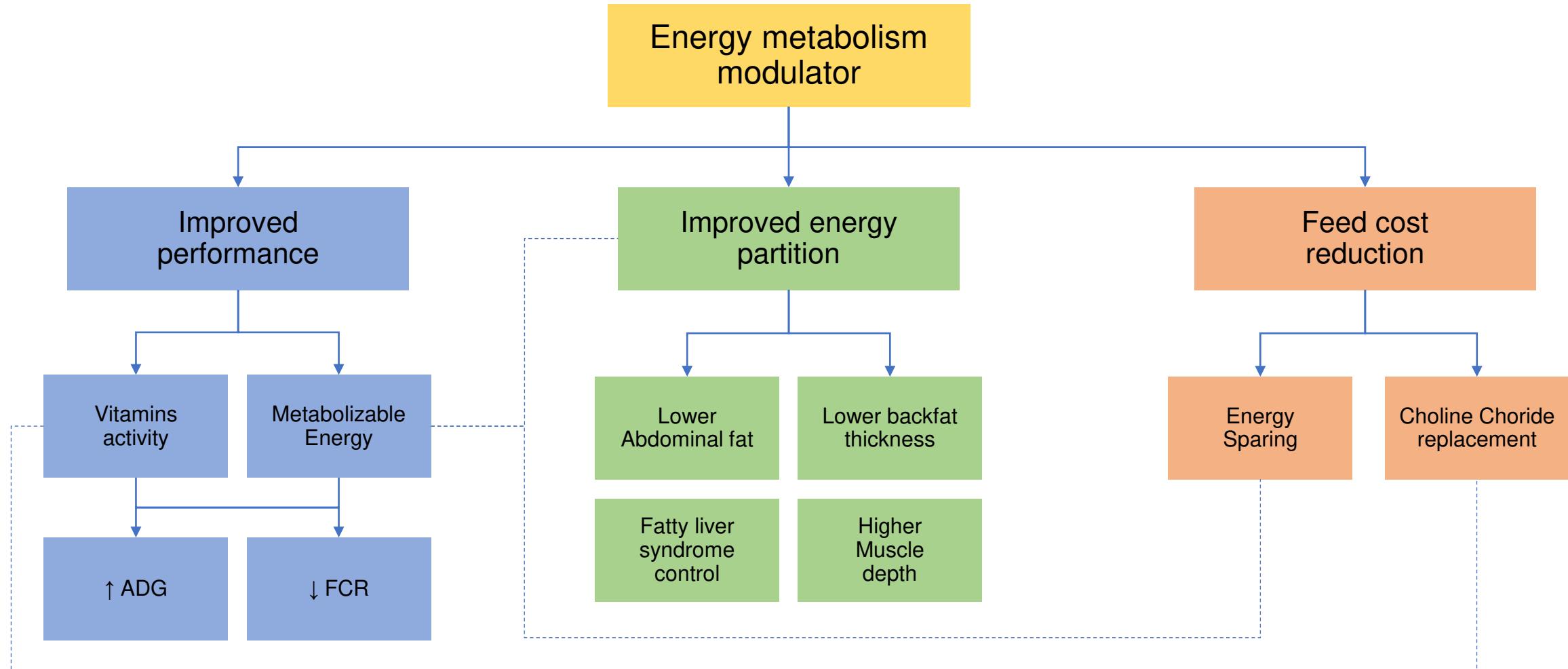


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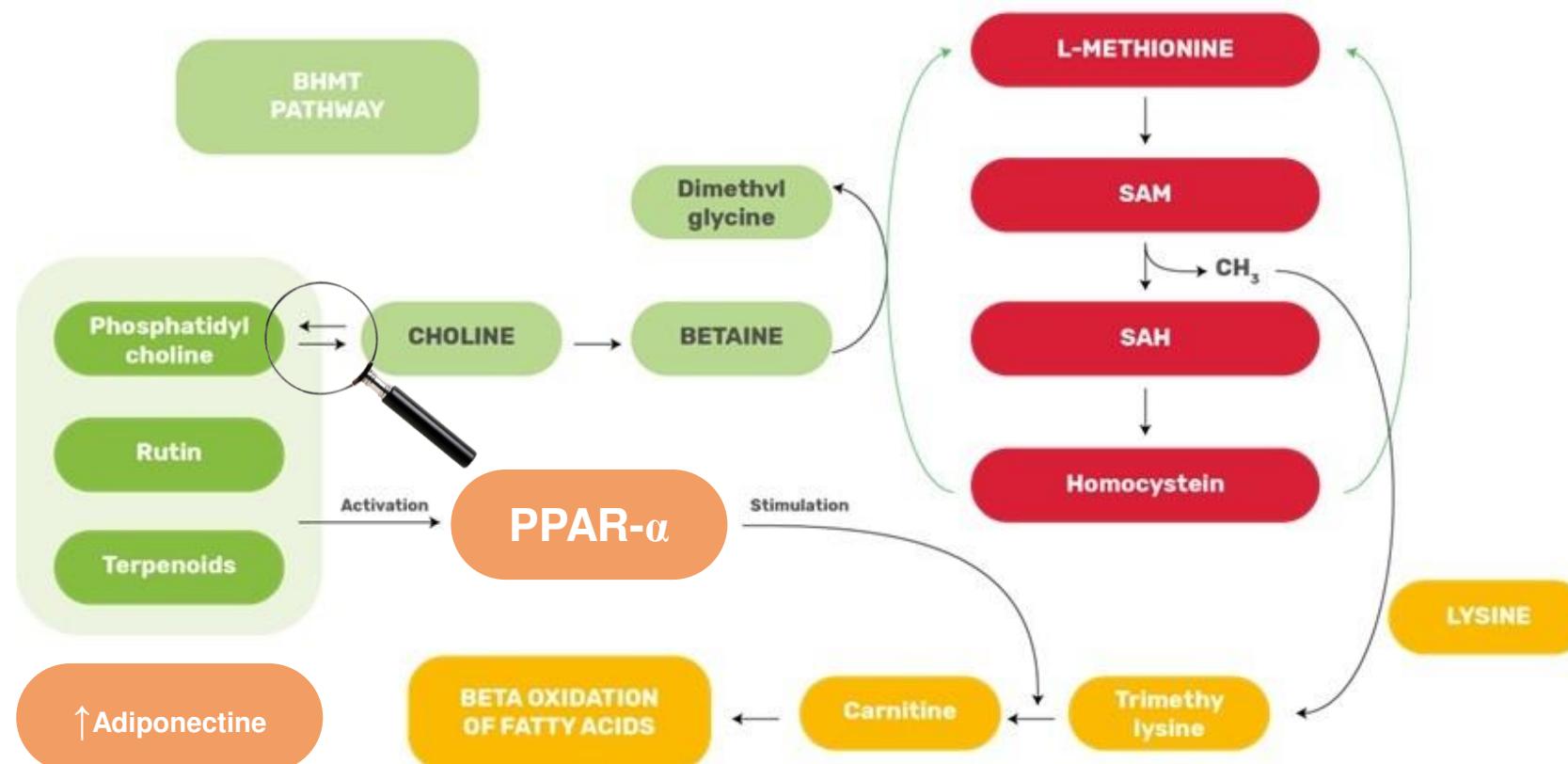
Product Family summary

Natu-B4®



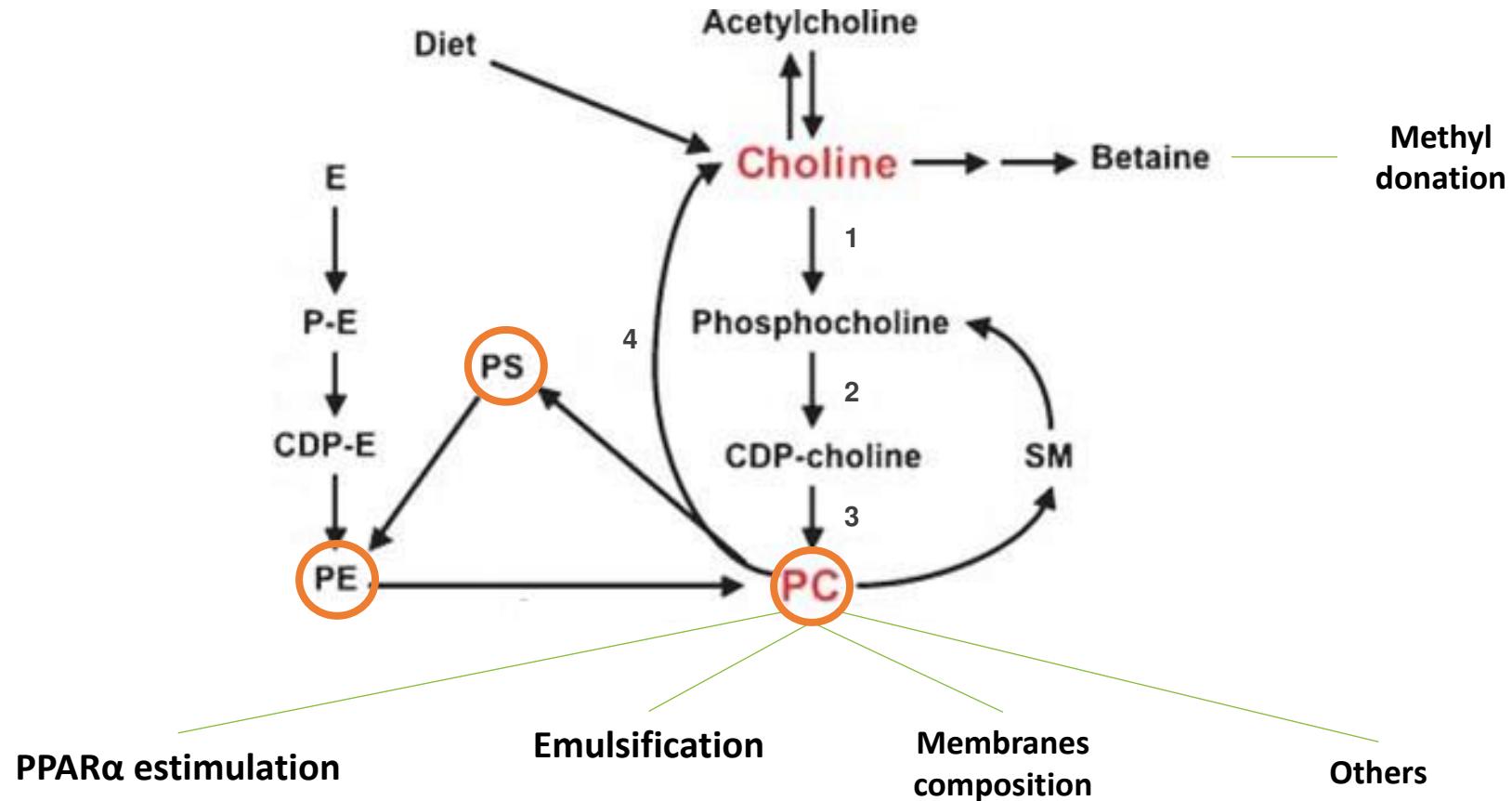
Mode of action

Natu-B4®



Mode of action detail

Natu-B4®



- Phosphatidylcholine is 3 metabolic steps (1, 2 and 3) ahead Choline for the most interesting functionalities;
- Phosphatidylcholine is just 1 step (4) behind Choline in the pathway to methyl donation;
- The Other Phosphatidyl conjugates as Phosphatidylserine (PS) and Phosphatidylethanolamine (PE) are also closer to Phosphatidylcholine than to Choline, being converted into PC with just 1-2 metabolic steps;

Mode of action validation

Role of Natu-B4™ on gene expression

- 240 Cobb 500 broiler chicks
- T1 – Control diet + CC60: Starter 900g/ton, Grower and Finisher 700g/ton
- 2 treatments x 6 replicates x 20 birds per pen
- T2 – Control diet + Natu-B4: Starter 315g/ton , Grower and Finisher 245g/ton (35% of the choline chloride dose)

- Acetyl COA Carboxilase (ACC) **+12,18%**
- Fatty Acid Synthase (FAS) **+11,41%**

Enzymes involved in the fatty acids synthesis

Fatty acids transport

- Fatty Acid Binding Protein (FABP) **+18,39%**

- **Peroxisome proliferator Activated receptor (PPAR)** **+39,03%**
- **Adiponectin** **+14,61%**
- **Pyruvate Kinase** **+159,51%**
- **Perlipin (PLIN)** **+24,97%**

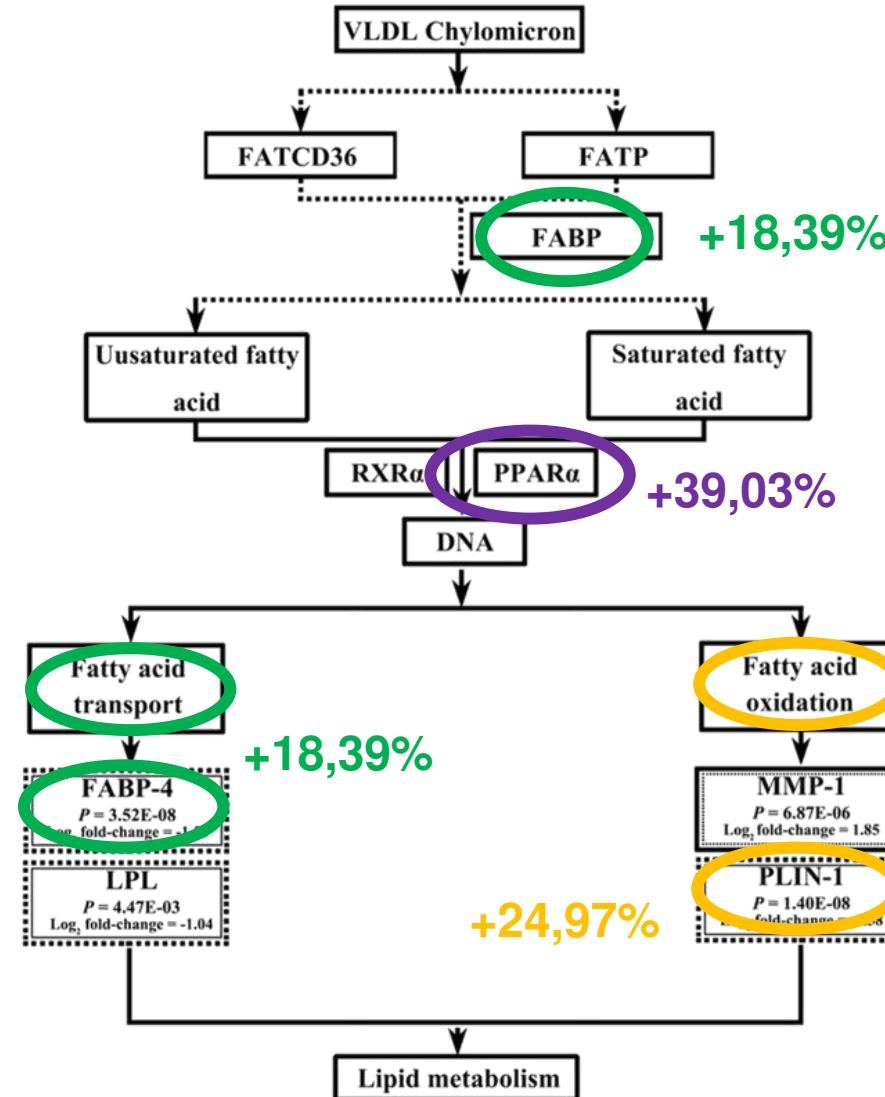
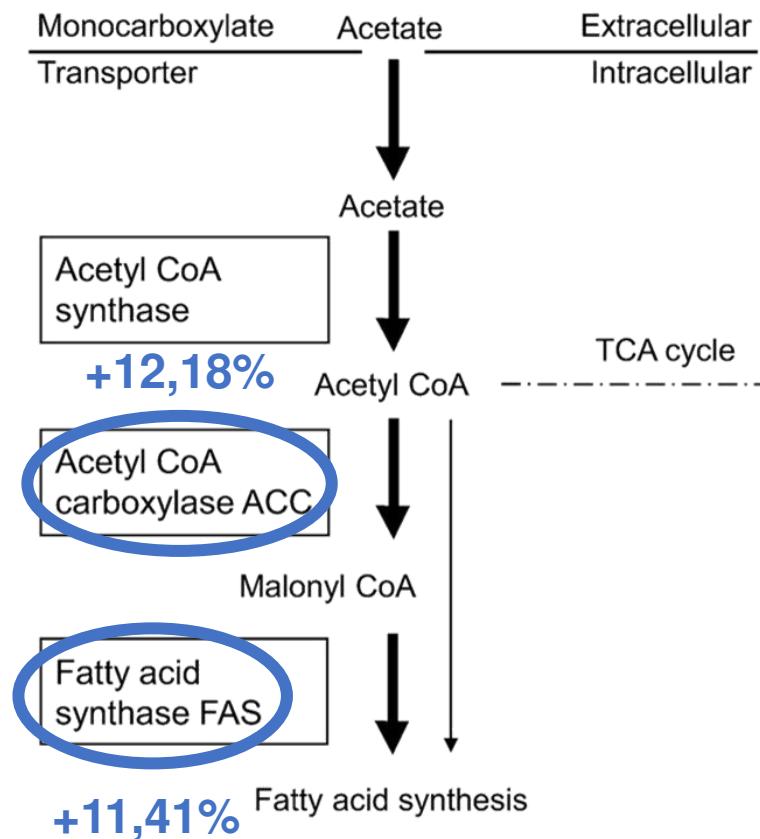
Glucose levels regulation and fatty acids degradation

Use of fatty acids and glycogenesis

- Cholesterol Acyltransferase (ACAT) **+16,15%**
- Phosphoenolpyruvate carboxykinase (PEPCK) **+47,69%**

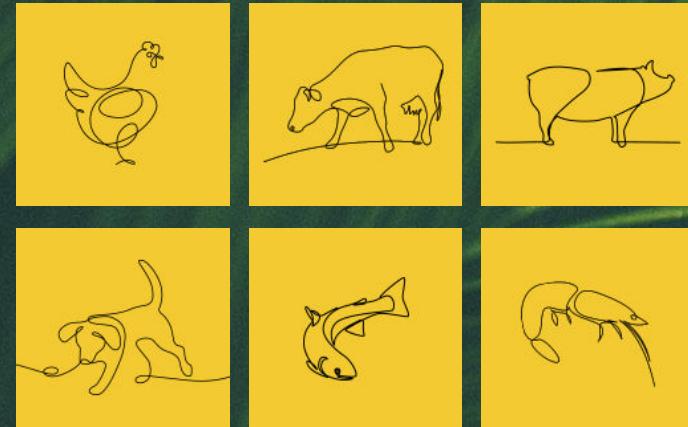
Kim et al. (2019)

Mode of action validation



Kim et al. (2019)

IMPROVING PERFORMANCE



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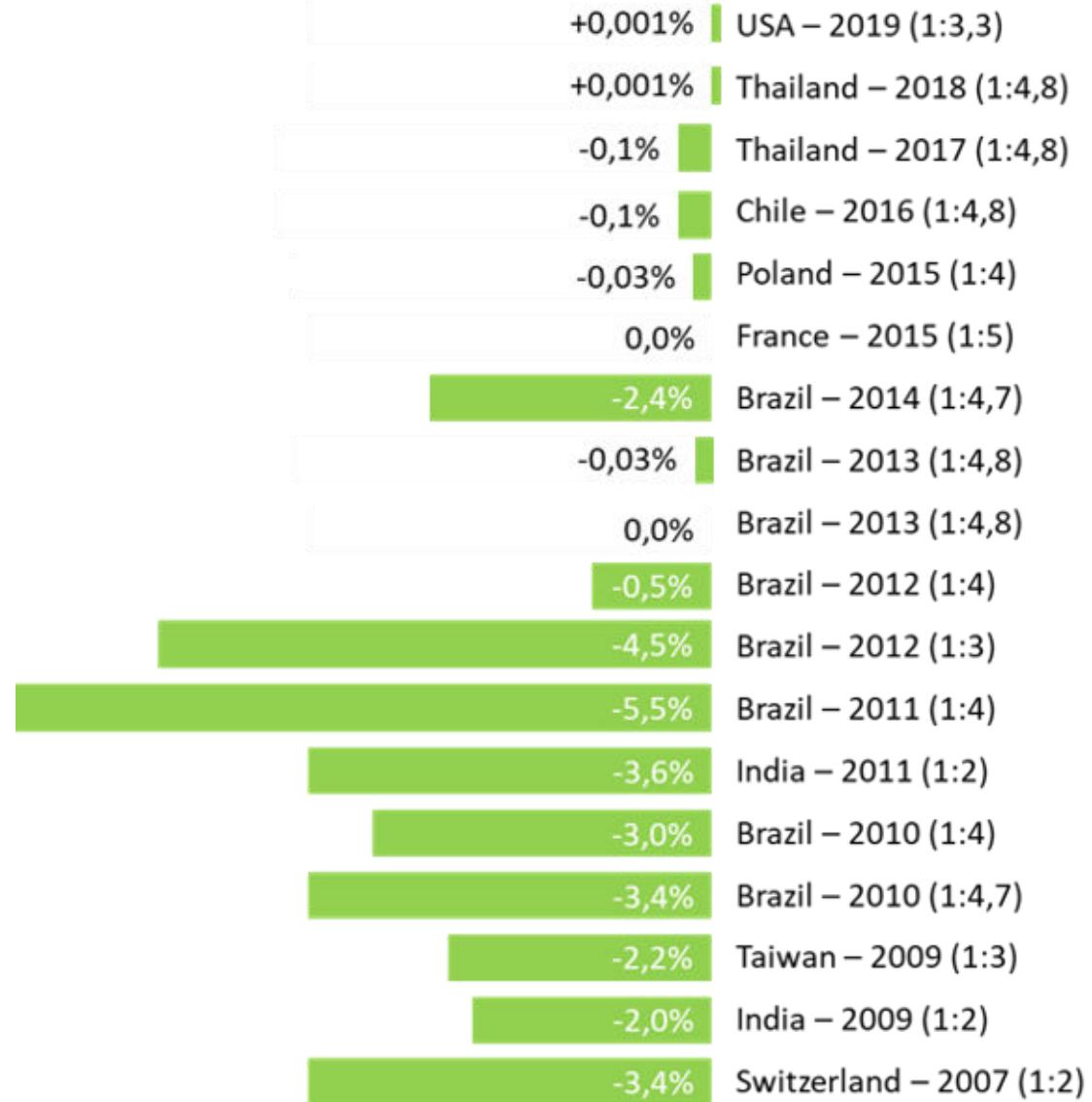


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SUMMARY OF TESTS IN BROILERS

Feed conversion (FCR)

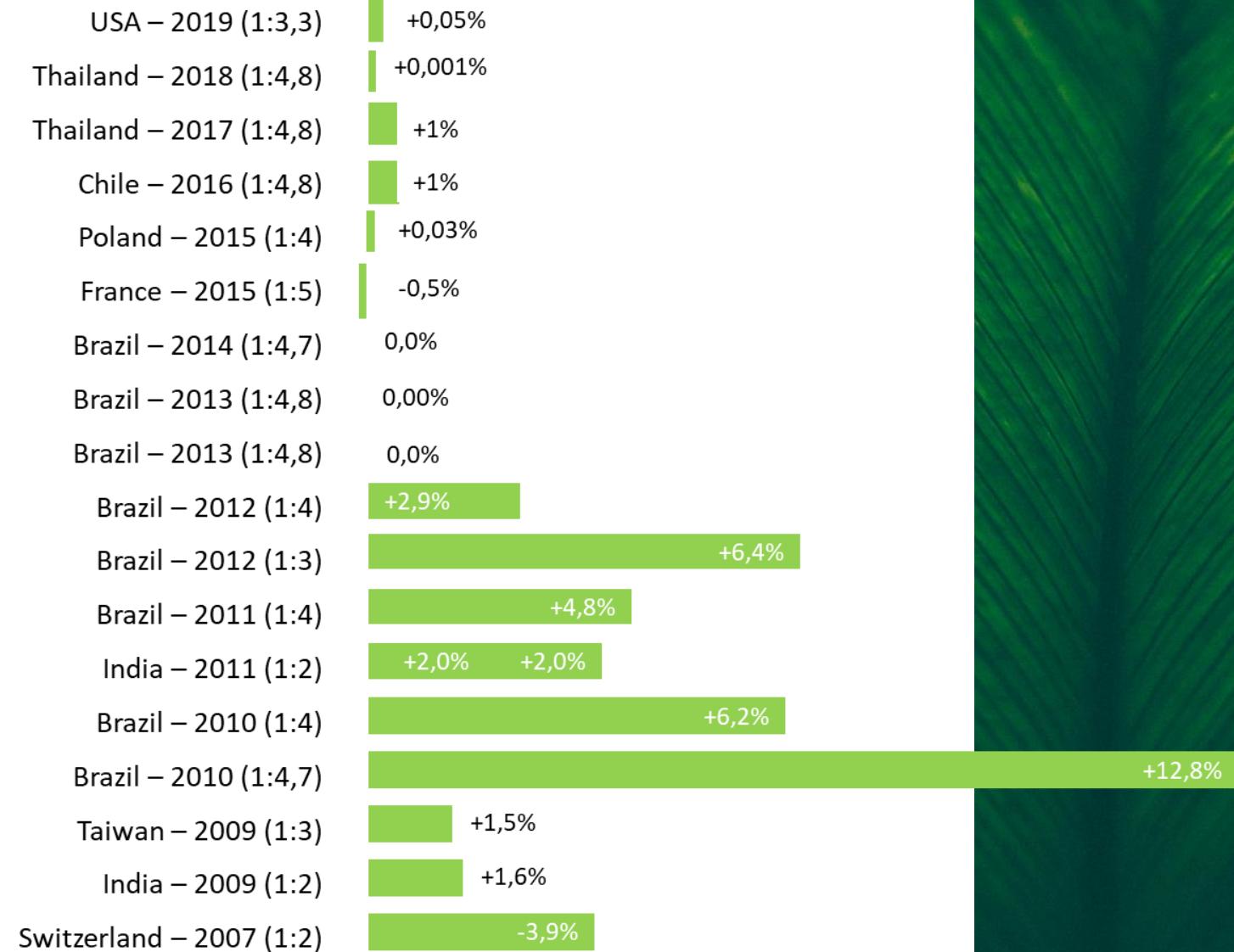
- Comparison between NatuB4 and Choline Chloride
- Different substitution ratios
- Significant change since 2013: bioequivalence



SUMMARY OF TESTS IN BROILERS

Weight Gain (WG)

- Comparison between NatuB4 and Choline Chloride
- Different substitution ratios
- Significant change since 2013: bioequivalence



Performance trial in broilers

Natu-B4®

PROTOCOL

216 COBB birds (4 replicates x 18 birds x 3 treatments)

Duration: 42 days

3 treatments:

- Negative control (no choline)
- Choline Chloride 60 at 1 kg/T
- NatuB4 at 0.5 kg/T

Dose Relationship CC:NatuB4

1 : 2

RESULTS

	Negative control	Choline chloride	NatuB4
Final BW (g)	1926 b	1927 b	1964 b
FCR	1,93 a	1,90 ab	1,84 b
Abdominal Fat content	2,51% a	2,20% ab	2,03% b
Liver fat content (mg/g)	66,2 a	58,9 b	55,2 b
Fatty liver incidence	2,8%	2,8%	0%

Performance trial in broilers

Natu-B4®

Protocol

- 2 treatments
- 10 Replicates
- 41 male birds per replicate
- 1-35 days of age

Doses per phase in each Treatment	D 1 – 12	D13 – 21	D 22 a 28	D 29 – 35
Choline Chloride 70 (g/ton)	857	643	643	429
NatuB4 (g/ton)	200	150	150	100

ADG	D0-12	D0-24	D0-28	D0-35
Choline Chloride	31,9	54,8	58,7	66,5
NatuB4	32,3	54,7	58,3	65,9

FCR	D0-12	D0-24	D0-28	D0-35
Choline Chloride	1,03	1,34	1,43	1,54
NatuB4	1,02	1,33	1,42	1,54

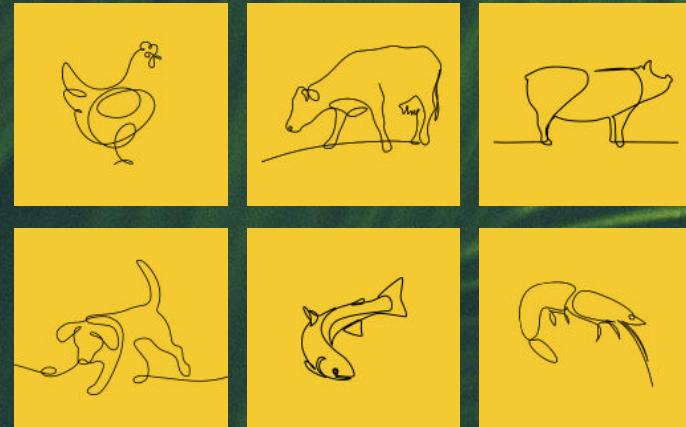
Blood analysis	Glucose (g/dL)	Colesterol (g/dL)	Triglycerides (mg/L)	Albumin (g/L)	AST (IU/L)	Alkaline phosphatase
Choline Chloride	2,23	1,33	0,47	12,7	449	10493
NatuB4	2,23	1,31	0,45	12,4	441	10442
Reference	2-5,0	1-2,5	0,37-1,15	-	<800	-

Liver weight at D35	ABW	Liver Weight	Relative liver weight
Choline Chloride	2361	55,4 b	2,35 % b
NatuB4	2353	51,3 a	2,18 % a

Average weight D35 = 2,355g

Le Roux
(2015)

PERFORMANCE trials in layers



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Performance trial in layers 2

Choline chloride replacement

Natu-B4®

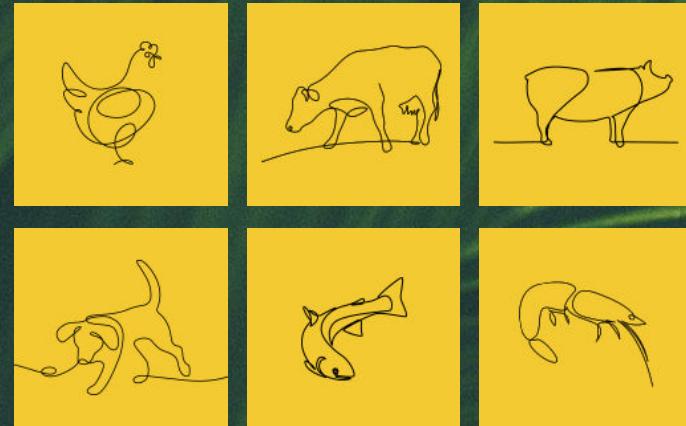
	Choline chloride	Natu-B4
Production phase	800 g/T	208 g/T

11 replicates x 20 layers x 2 treatments
 n = 440 gallinas
 63 to 70 semanas de edad

	Production (%)	Egg weight (g)	BW (g)	FI (g)	FCR (g)
Choline Chloride	91,6	69,6	1826	116,2	1,82
Natu-B4	91,6	69,3	1803	113,8	1,79

Le Roux (2017)

CHOLINE CHLORIDE REPLACEMENT



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Choline Chloride points of attention

Natu-B4®

1/3 of the choline is absorbed

- 2/3 are transformed into trimethylamine (TMA) during digestion by intestinal flora.
- Toxic, when absorbed and can negatively affect the hepatic metabolism
- Can be associated at the fishy odor of eggs

Low concentration

- 50, 60% powder and 75% liquid
- Chloride ~ 25%
- High inclusion levels

Risk of adulteration

- QC – indirect analysis
- Calculated [Choline]
- Common salt = 60% Chloride

Adds chloride to the diet

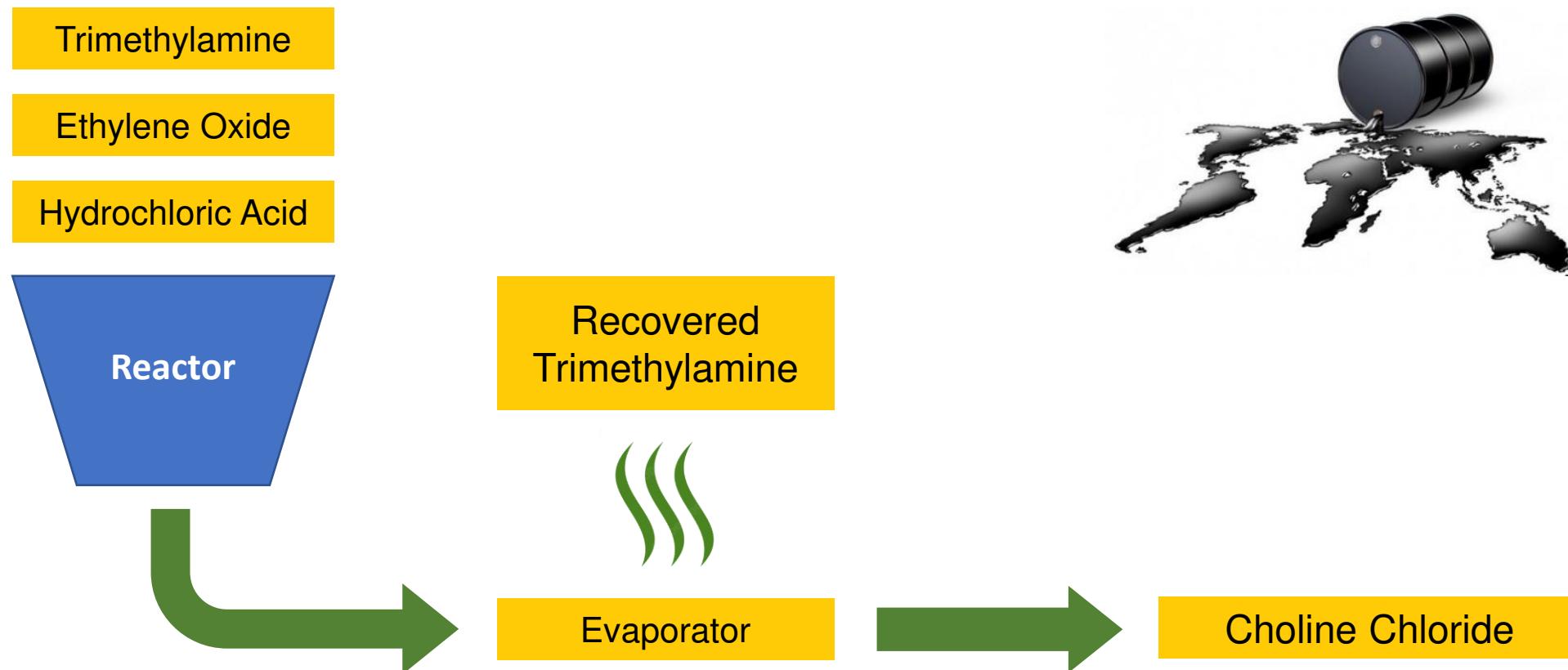
- Electrolytic balance

Associated with metabolic diseases such as: Metabolic acidosis, Ascitic syndrome, Tibial dyschondroplasia

Hygroscopic and corrosive

- Unstable and hard to handle in the factory
- Increases moisture and causes destruction of vitamins and pigments in premixes and feeds

SYNTHESIS OF CHOLINE CHLORIDE



ELECTROLYTIC BALANCE Cost reduction

Natu-B4®

- Excess Cl- (above 0.40%) ↓ growth and ↑ incidence of tibial dyschondroplasia (Luo et al., 1992);
- Recommended Cl level in male chickens: 0.200 to 0.165% (Rostagno, 2011);
- Sources of Cl-

L-Lysine-HCl

NaCl

Choline Chloride

Balancing the Na Level: Important Benefit

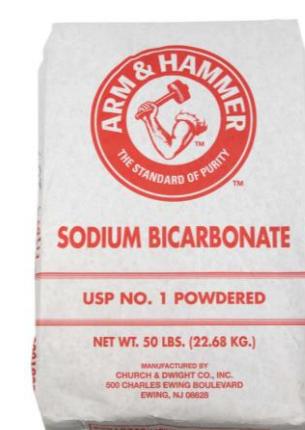
1T Natu-B4



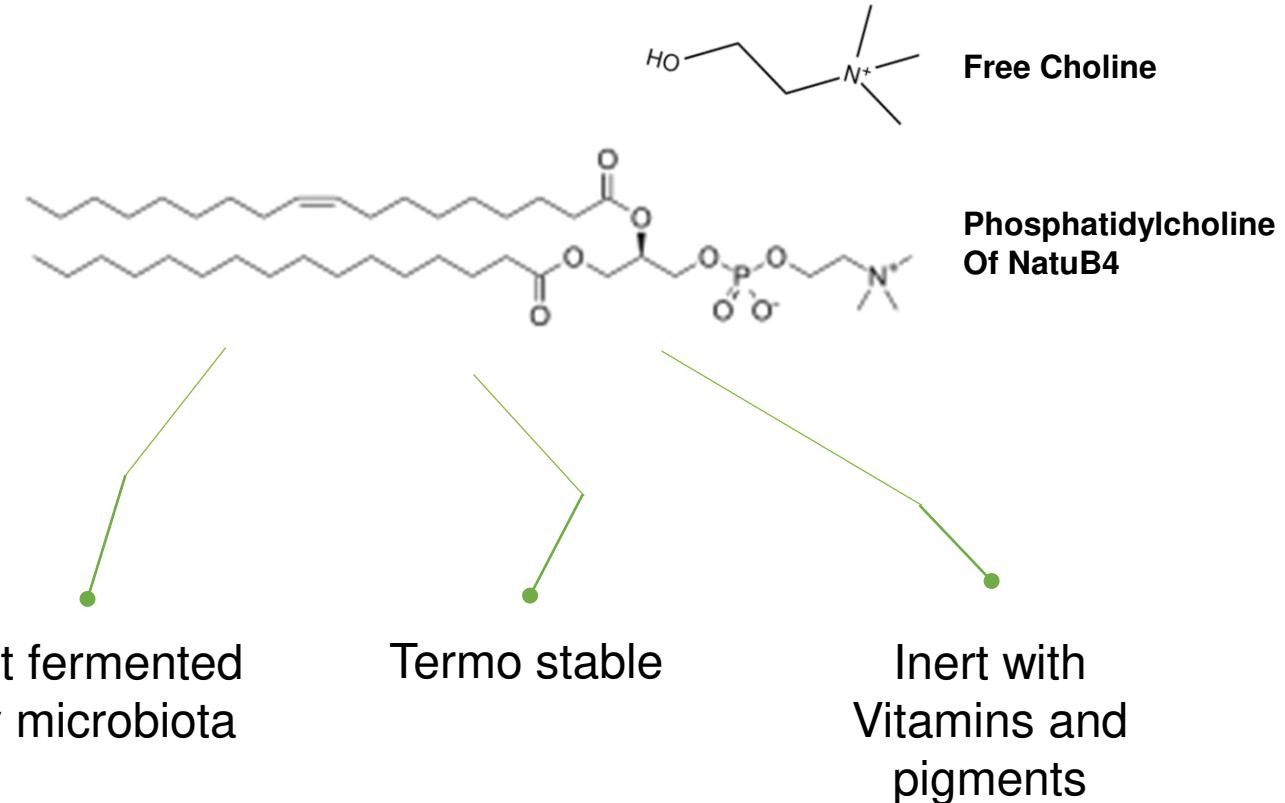
+ 1.3T NaCl



-1.7T NaHCO₃



Natu-B4® contains phosphatidylcholine and other molecules that are important for the metabolism



Stability of vitamins in a premix with Choline Chloride

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Natu-B4®

Vitamins	% Activity Retention		Average loss per month (%)
	1 month	6 months	
Vitamin A	85	58	8
Vitamin D ₃	91	65	6
Vitamin E	95	82	3
Vitamin K	64	0	38
Tiamine	70	27	17
Riboflavine	95	56	9
Pyridoxine	92	56	9
Pantotonic acid	95	58	8
Niacin	95	58	8
Biotin	93	57	9
Vitamin B ₁₂	98	89	2
Folic acid	85	43	12

Whitehead
(2002)

Stability of vitamins in a premix with Choline Chloride or Natu-B4

Natu-B4®

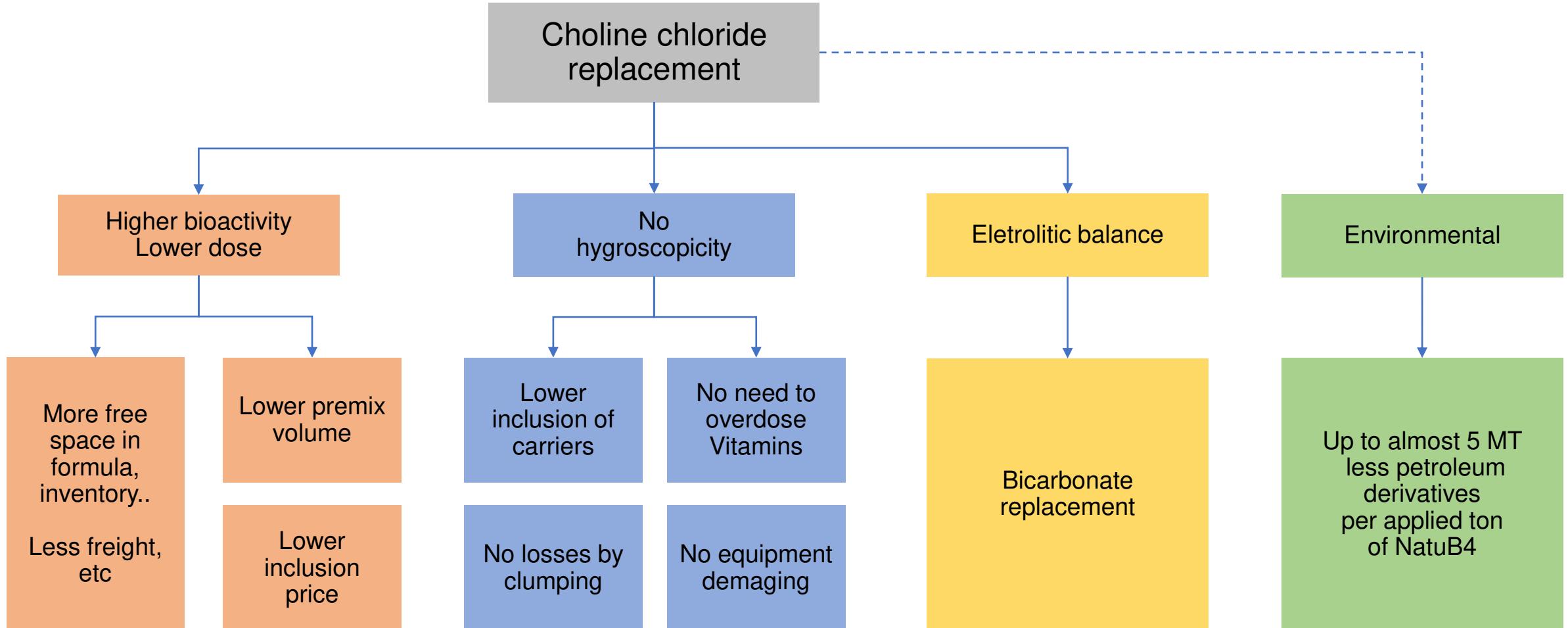
Vitamins	Control			Choline chloride 60%			NatuB4™		
	24°C	40°C	45°C	24°C	40°C	45°C	24°C	40°C	45°C
Vitamin B1	16,8%	30,4%	33,0%	70,0%	78,9%	79,0%	18,3%	40,4%	34,6%
Vitamin E	18,7%	30,6%	40,8%	44,4%	55,6%	60,0%	25,0%	30,0%	33,3%
Vitamin B6	17,9%	35,7%	38,2%	28,8%	62,5%	63,8%	22,3%	30,1%	32,0%
Vitamin B9	10,3%	29,4%	38,7%	25,0%	60,0%	63,8%	18,3%	32,7%	34,6%
Vitamin B12	6,3%	7,8%	15,1%	36,1%	57,0%	83,7%	12,3%	15,1%	19,8%
Vitamin B2	3,0%	15,9%	18,4%	34,1%	31,8%	43,2%	8,0%	11,0%	11,0%
Vitamin C	14,3%	14,3%	24,8%	11,1%	43,2%	47,7%	11,1%	14,0%	11,1%

(2010)

6 Months Trial. With temperatures of 24 °C, 40°C and 45°C, vitamin loss has a considerable reduction with NatuB4™

Financial and environmental benefits

Natu-B4®





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