Czech University of Life Sciences Prague

Selected Non-Protein Nitrogen Compounds in Insects for Food and Feed Purposes

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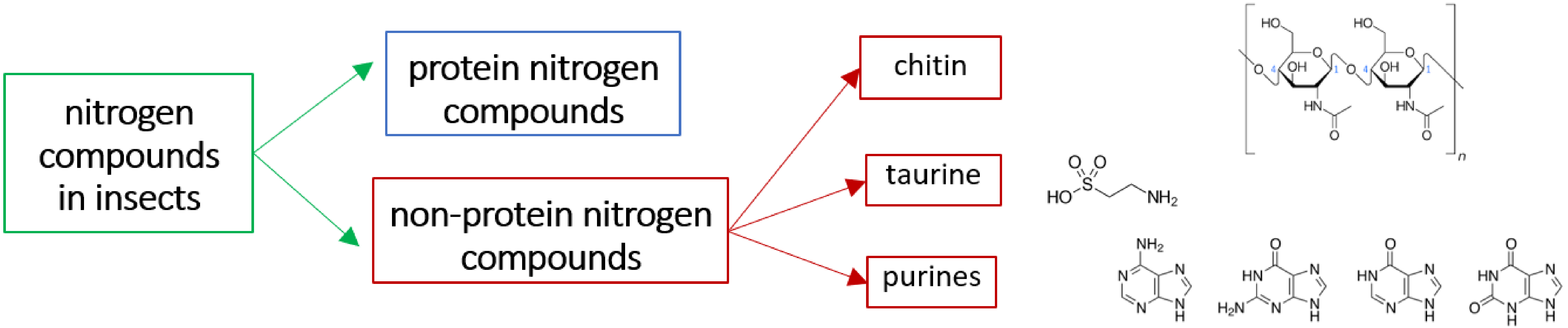
Food and Natural Resources

OVERWIEW

- Nitrogen compounds in food
- Nitrogen-to-protein conversion factor
- Non-protein nitrogen compounds
  - Purines
  - Taurine
  - Chitin



Kjeldahl method

Nitrogen in compounds is converted to NH_3, followed by acido-basic reactions and titration

Nitrogen-to-protein conversion factors

generally used: 6.25 (16% N)

eggs, meat: 6.25

milk: 6.38

wheat: 5.83

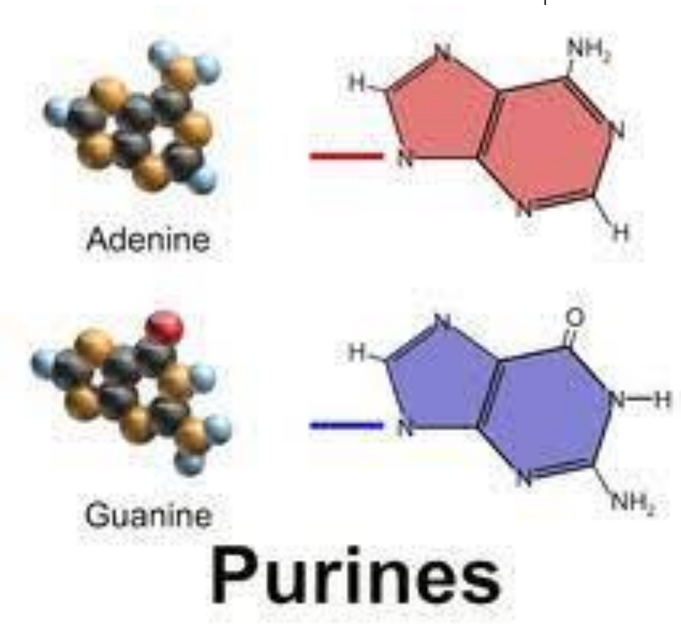
rice: 5.95

peanuts: 5.46

Non-protein nitrogen substances in insects: **11-26%** (Janssen et al. 2017)

for example: chitin, nucleic acids, phospholipids, and excretion products (e.g., ammonia) in the intestinal tract

PURINES



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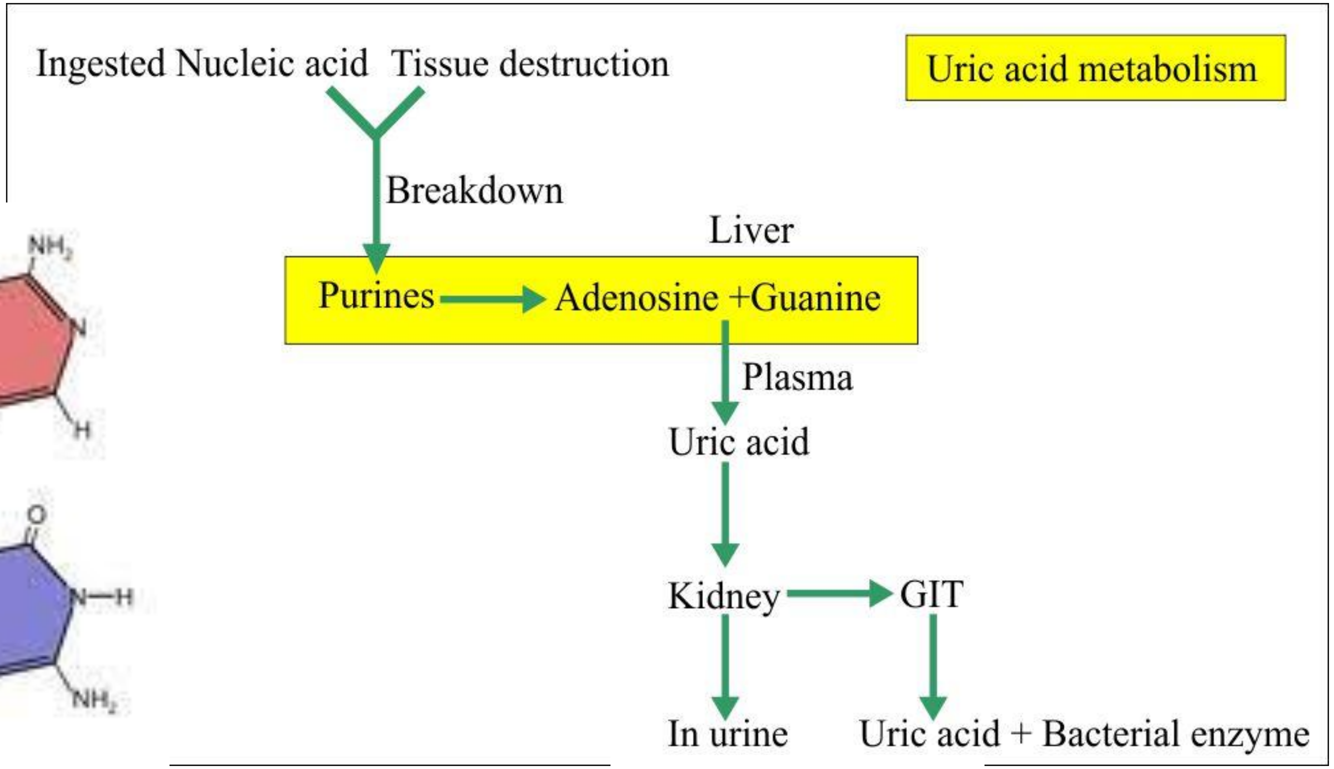
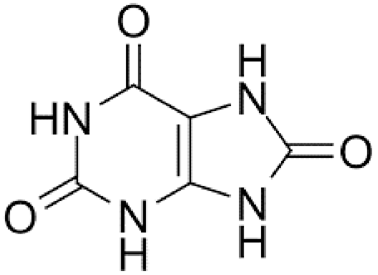


Uric acid



GOUT

- painful form of inflammatory arthritis
- extra **uric acid** in body ⇒ **sharp crystals** may form in the big toe or other joints (knee, ankle, foot, hand, wrist and elbow) ⇒ episodes of swelling and pain called gout attacks
- treatable with medications and changes in diet and lifestyle

Purines in food

- High levels (150-1000 mg/100 g): organ meats, seafood and yeast extracts
- Moderate levels (50-150 mg/100 g): beans, pulses and some vegetables
- Low levels (<50 mg/100 g): milk products, eggs, refined cereals and most fruits and vegetables

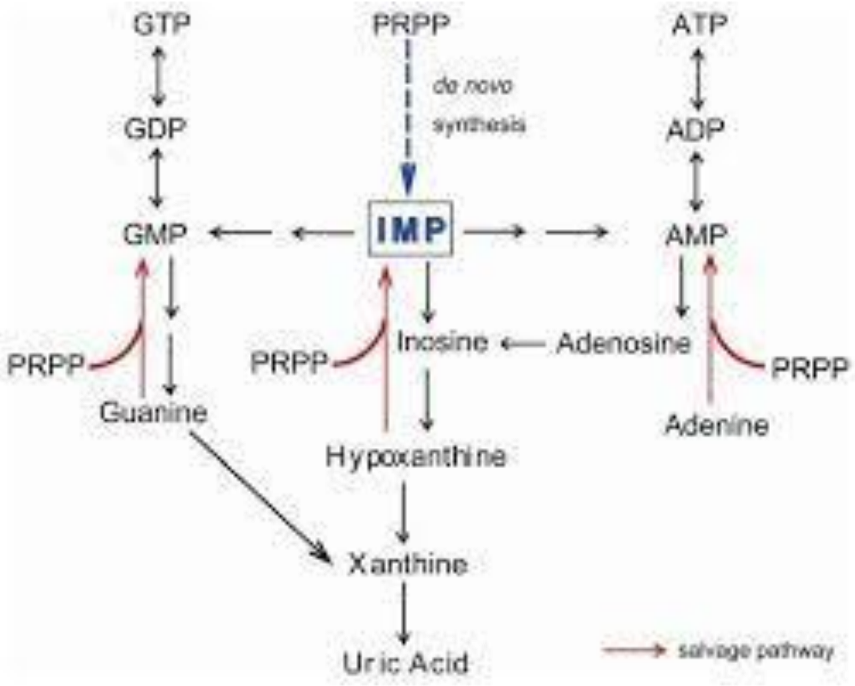
Lockyer & Stanner (2016)

Purines in insect???

AIM OF OUR RESEARCH IN PURINES

- to analyse the content of selected purine derivatives (adenine, guanine, hypoxanthine, and xanthine) and their metabolite (uric acid) in different insect species
- to compare the levels of purines with conventional meat sources
- to evaluate sex-dependent differences in purines content
- to see the effect of developmental stage

on purines and uric acid content



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MATERIAL

Insect samples:

Orthoptera


house cricket (*Acheta domesticus*) – nymph and adult

Jamaican field cricket (*Gryllus assimillis*) - nymph and adult


migratory locust (*Locusta migratoria*) – nymph and adult

desert locust (*Schistocerca gregaria*) - adult

Coleoptera

mealworm (*Tenebrio molitor*) - larvae and pupae

lesser mealworm (*Alphitobius diaperinus*) - larvae and pupae

Blattodea

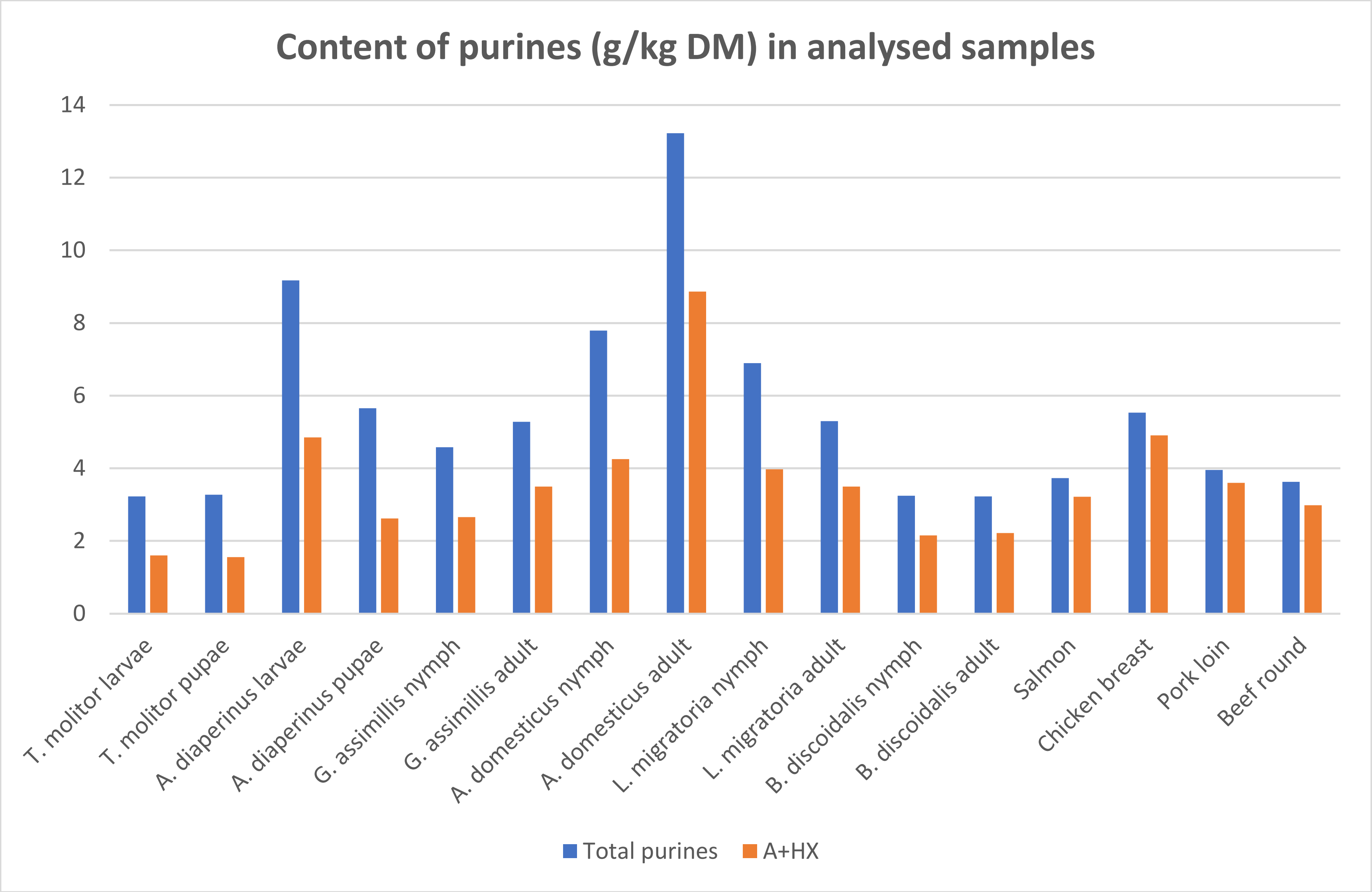
discoid cockroach (*Blaberus discoidalis*) – nymph and adult

MATERIAL AND METHODS

- **Conventional meat samples:**
  chicken, pork, beef, and salmon

- **Purine analysis:**
  RP-HPLC/DAD,
  mobile phase - 0.05 mol/L KH_2PO_4 in water (pH 3.6) as solvent A,
  and acetonitrile as solvent B,
  Luna C18 column (250 × 4.6 mm),
  detection - uric acid 285 nm,
  guanine and hypoxanthine 250 nm,
  xanthine and adenine, 260 nm

Insect vs. conventional meat sources



Total purines

insects:

3.23-13.22 g/kg DM

meat:

3.61-5.53 g/kg DM)

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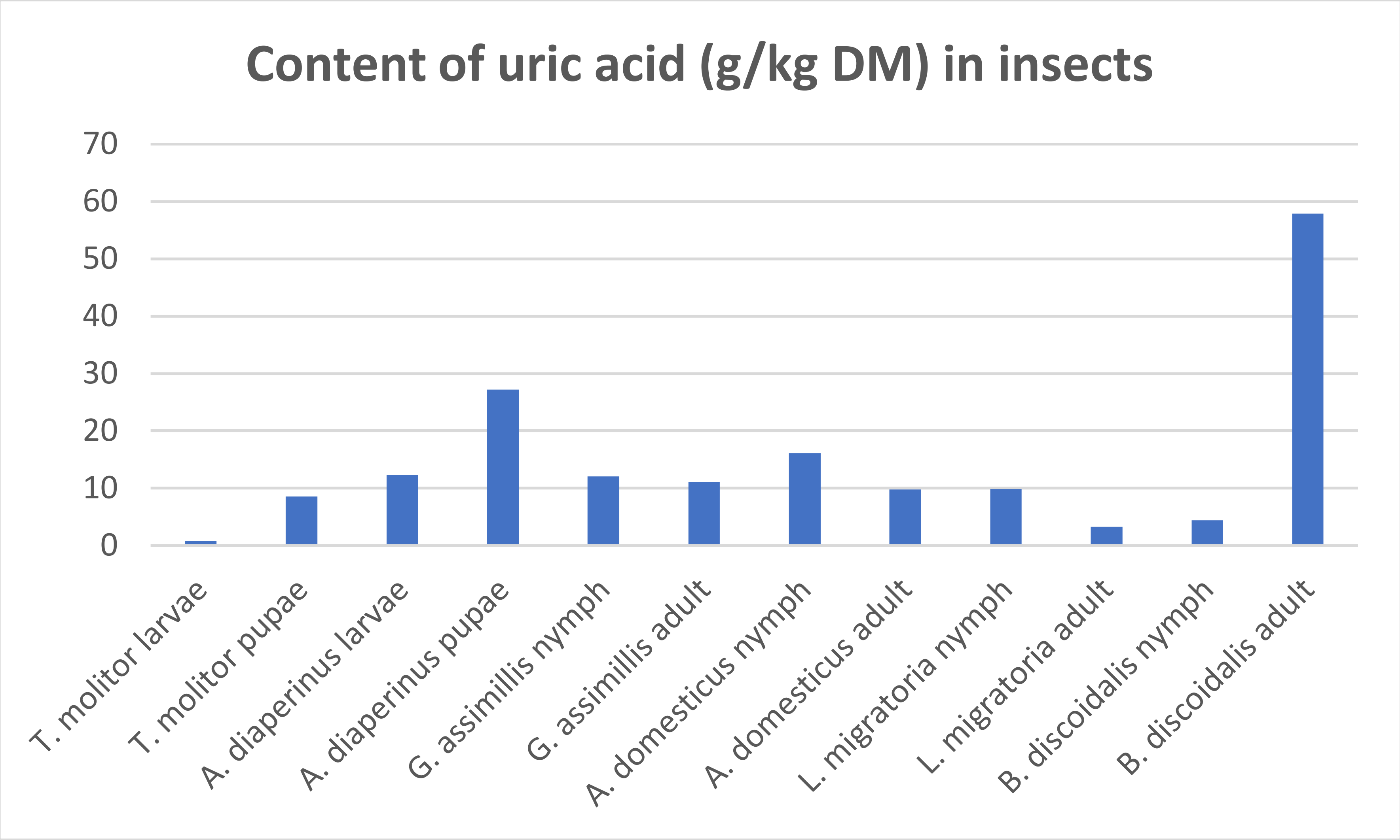
Uric acid in insects



Uric acid

insects:

0.81-57.86 g/kg DM

meat:

<LOD (0.1 g/kg DM)

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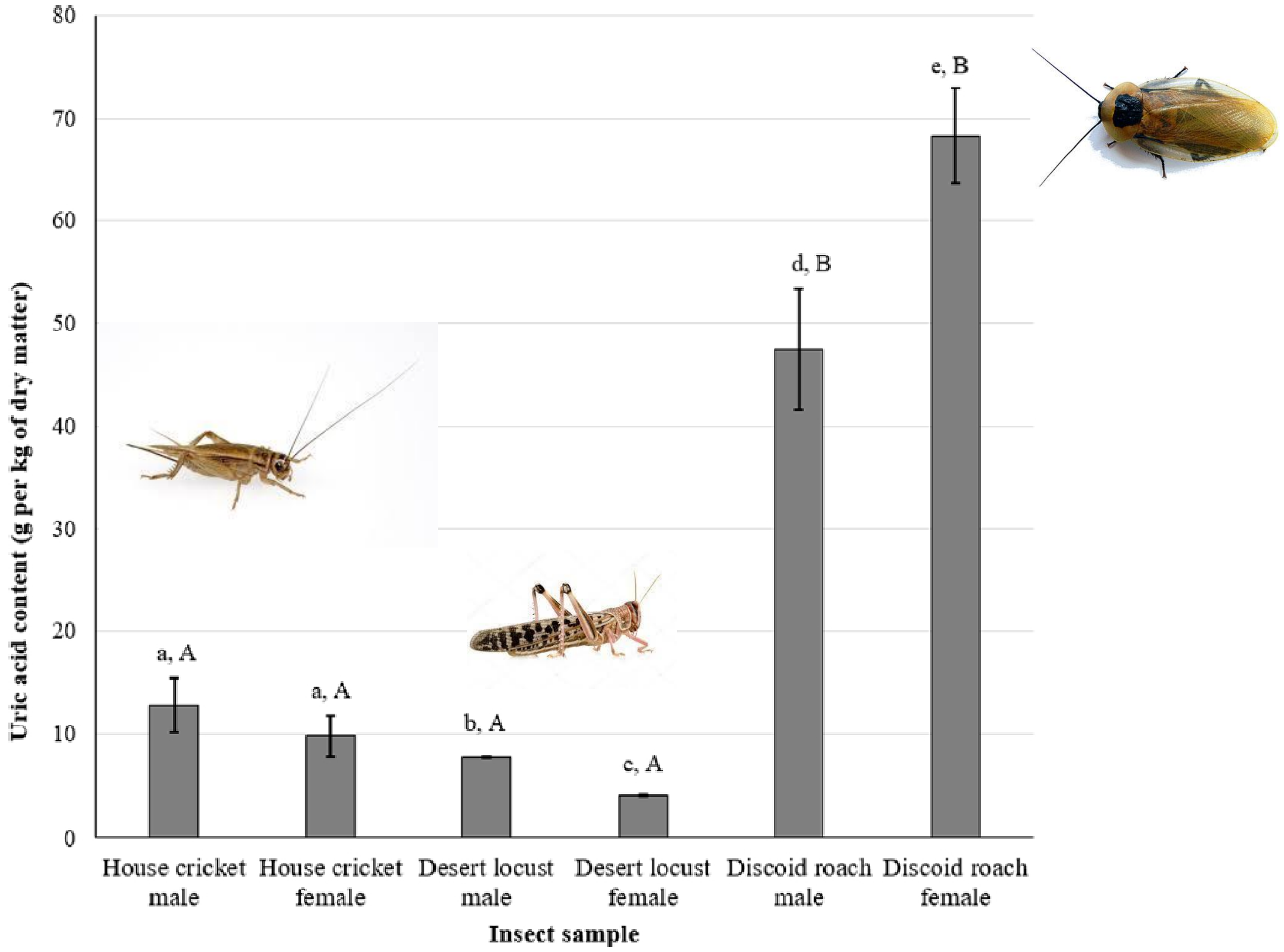
RESULTS – uric acid content – differences between sex and species



Insects vs. conventional meat sources

Total purine content in analysed insects was more less similar to the content of such compounds in commonly consumed meats.

The obtained results indicate that edible insects constitute a reasonable source of purines and uric acid, and patients suffering gout should be cautious about excessive and daily consumption of edible insects and the products made therefrom.

Contents lists available at ScienceDirect
Journal of Food Composition and Analysis
journal homepage: www.elsevier.com/locate/jfca
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Short communication

Sex-dependent differences in purine and uric acid contents of selected edible insects

Monika Sabolová [a,*], Martin Kulma [b], Lenka Kouřimská [a]

Faculty of Agrobiology,
Food and Natural Resources

RESEARCH ARTICLE

Investigating purine and uric acid contents of various development stages of artificially reared edible insects

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Journal of Insects as Food and Feed: 9 (1)- Pages: 77 - 85

https://doi.org/10.3920/JIFF2022.0011

Contents lists available at ScienceDirect
Food Chemistry
journal homepage: www.elsevier.com/locate/foodchem

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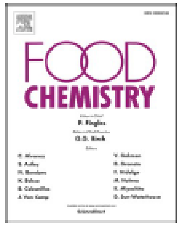


Check for updates

Changes in purine and uric acid content in edible insects during culinary processing

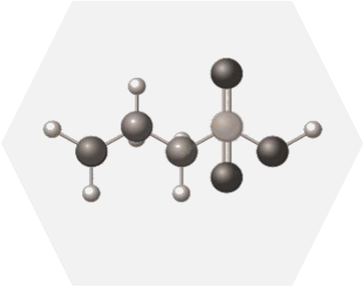
Monika Sabolová [a,*], Martin Kulma [b], Dora Petříčková [a], Kateřina Kletečková [a], Lenka Kouřimská [a]

Taurine

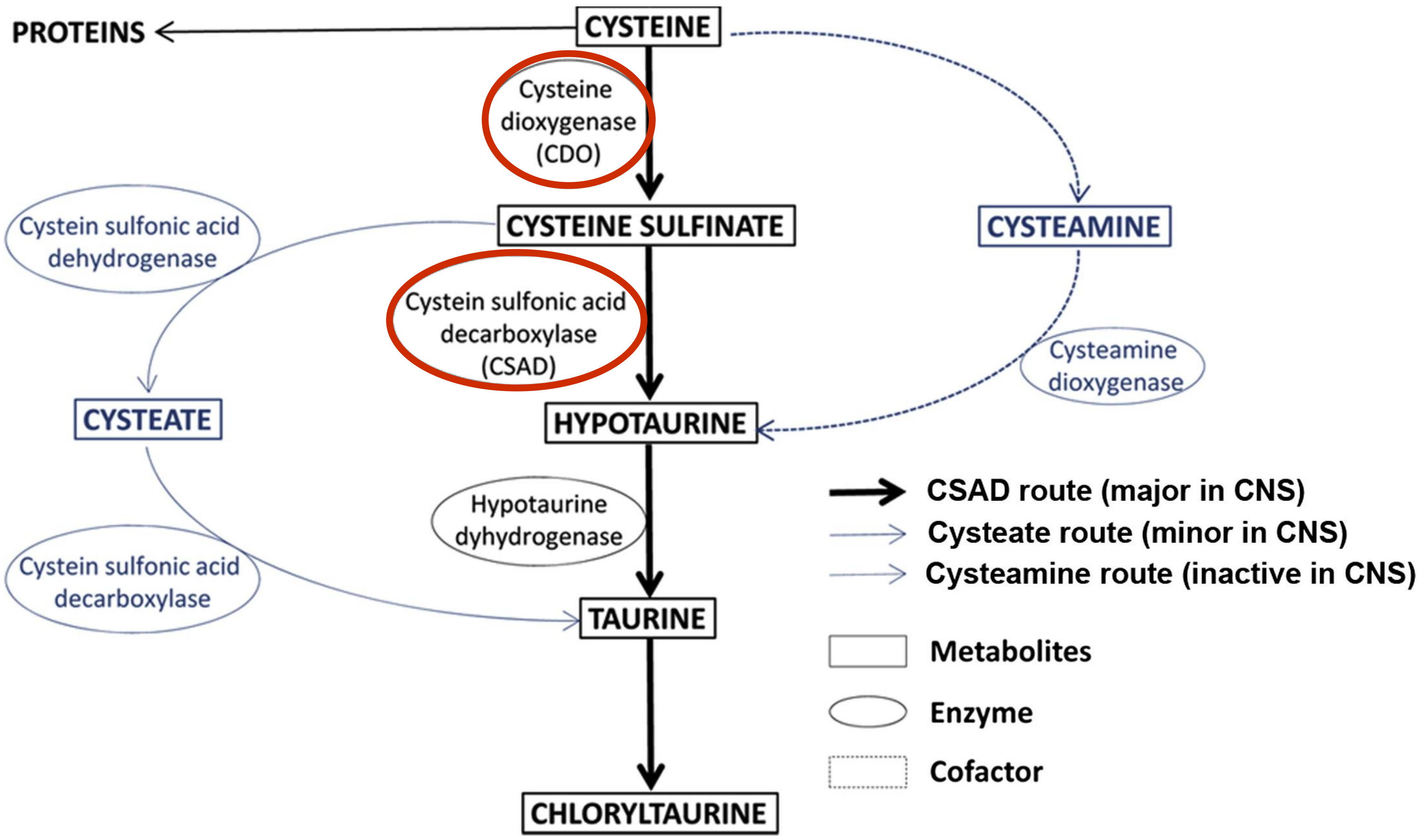
- 1827 - Friedrich Tiedemann and Leopold Gmelin – bile of *Bos taurus*
- **2-aminoethane sulfonic acid**

- plays a key role in the central nervous system, light-sensitive tissues in the retina, and in cardiac and vascular functions
- intracellular osmolyte
- neurotransmitter
- antioxidant

- **essential for some organisms**
    - low or no levels of enzymes cysteine dioxygenase and cysteine sulfonic acid decarboxylase



Metabolism



(Froger et al. 2014)

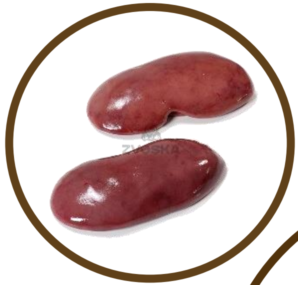
Taurine sources

Meat	Part	Taurine [mg/100 g]
Beef	spleen	96
	Lungs	87
Pork	kidney	69-96
	lungs	87
Chicken	thigh	169-379
	kidney	110
Fish		72-280

Product	Animal	Taurine [mg/100 g]
Milk	cow	2.5
	goat	8.5–11.4
Yoghurt	cow	0.8–3.3
	goat	5.3








Detection of taurine content in several commercially available insect species suitable for large scale production:

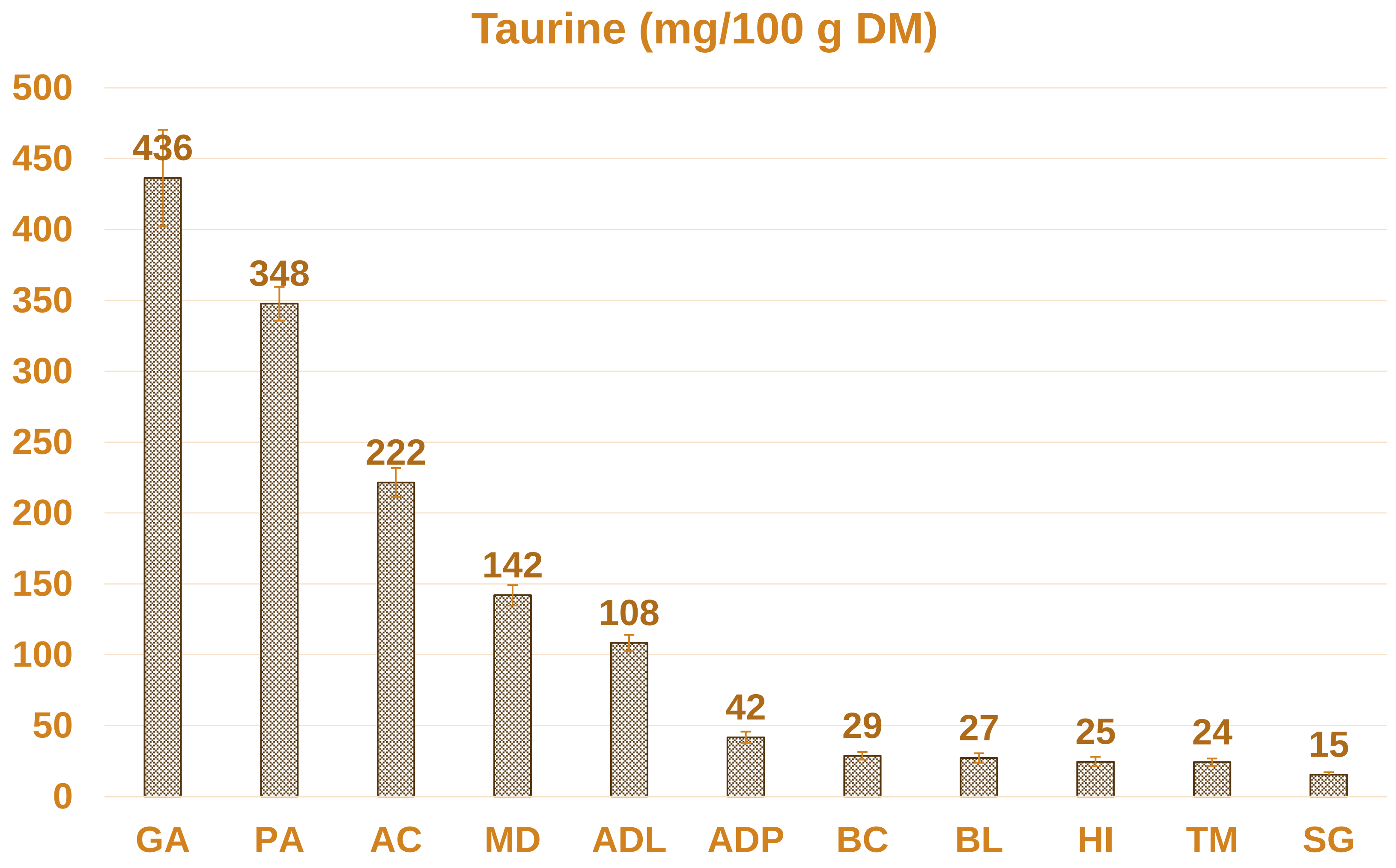
- lesser mealworm (*Alphitobius diaperinus*)
- house cricket (*Acheta domesticus*)
- death's head cockroach (*Blaberus craniifer*)
- Turkestan cockroach (*Blatta lateralis*)
- Jamaican field cricket (*Gryllus assimilis*)
- black soldier fly (*Hermetia illucens*)
- house fly (*Musca domestica*)
- American cockroach (*Periplaneta americana*)
- desert locust (*Schistocerca gregaria*)
- yellow mealworm (*Tenebrio molitor*)

Methods

- N-substances (crude protein) – Kjeltec
- Chitin – hydrolysis (6M sulphuric acid) – glucosamine – spectrophotometric method
- Taurine - conversion to isethionic acid - on-line coupled capillary isotachophoresis with capillary zone electrophoresis with conductivity detection

- *True protein content:*
- True protein = (total mineral nitrogen – chitin nitrogen – taurine nitrogen) × 6.25
  - chitin nitrogen = chitin/14.51 (conversion factor derived from the representation of nitrogen at 6.89 % in a molecule of chitin)
  - taurine nitrogen = taurine/8.54 (11.19% of N in a taurine molecule)

Tested insect	Stage	Order	Dry matter (g/100 g)	True protein (g/100 g FW)	Chitin (g/100 g FW)	Taurine (mg/100 g FW)	Taurine (mg/100 g protein)
G. assimilis	adult	Orthoptera	27.74 ± 0.91	20.83 ± 0.71	2.14 ± 0.11^{bc}	121.0 ± 10.2^{a}	588.6 ± 49.8^{a}
P. americana	adult	Blattodea	33.10 ± 1.11	21.95 ± 0.74	2.33 ± 0.08^{b}	115.1 ± 4.0^{a}	559.5 ± 19.2^{a}
A. domesticus	adult	Orthoptera	24.97 ± 0.26	20.57 ± 0.70	2.24 ± 0.06^{b}	55.3 ± 2.2^{b}	268.9 ± 10.8^{b}
M. domestica	larvae	Diptera	24.38 ± 0.82	10.75 ± 0.36	1.97 ± 0.06^{bc}	34.6 ± 1.8^{c}	168.3 ± 8.6^{c}
A. diaperinus	larvae	Coleoptera	31.21 ± 1.05	20.67 ± 0.70	1.93 ± 0.05^{bc}	33.8 ± 1.8^{c}	164.4 ± 8.6^{c}
A. diaperinus	pupae	Coleoptera	36.62 ± 1.23	30.07 ± 1.02	2.22 ± 0.11^{b}	15.3 ± 1.4^{d}	74.3 ± 7.0^{d}
H. illucens	prepupae	Diptera	43.25 ± 1.45	11.89 ± 0.40	2.25 ± 0.11^{b}	10.6 ± 1.4^{d}	51.7 ± 7.0^{d}
B. lateralis	adult	Blattodea	34.83 ± 1.17	22.78 ± 0.77	2.06 ± 0.07^{bc}	9.5 ± 1.2^{d}	46.0 ± 5.8^{d}
B. craniifer	adult	Blattodea	24.62 ± 1.36	12.47 ± 0.42	2.11 ± 0.03^{bc}	7.1 ± 0.7^{d}	34.3 ± 3.4^{d}
T. molitor	larvae	Coleoptera	26.96 ± 0.96	17.54 ± 0.60	1.64 ± 0.11^{c}	6.5 ± 0.7^{d}	31.7 ± 3.5^{d}
S. gregaria	adult	Orthoptera	34.67 ± 1.16	13.57 ± 0.46	3.03 ± 0.19^{a}	5.4 ± 0.6^{d}	26.0 ± 2.8^{d}



GA - *Gryllus assimilis*
PA - *Periplaneta Americana*
AC - *Acheta domesticus*
MD - *Musca domestica*
ADL – larvae of *Alphitobius diaperinus*
ADP – pupae of *Alphitobius diaperinus*
BC - *Blaberus craniifer*
BL - *Blatta lateralis*
HI - *Hermetia illucens*
TM - *Tenebrio molitor*
SG - *Schistocerca gregaria*

Results

4 significantly distinct groups according to taurine content:

a) *G. asimilis* and *P. americana* (121.0 and 115.1 mg/100 g FW)

b) *A. domesticus* (55.3 mg/100 g FW)

c) *M. domestica* and larvae of *A. diaperinus* (34.6 and 33.8 mg/100 g FW)

d) *A. diaperinus* pupae, *H. illucens, B. lateralis, B. craniifer, T. molitor,* and *S. gregaria* (15.3-5.4 mg/100 g)

Taurine and chitin in insects

- Insects have been proposed as one of the most promising alternatives to conventional feline as well as fish feeds ⇒ the knowledge of taurine content in insects is valuable.
- The most taurine-rich insect species are comparable with conventional feeding ingredients such as fishmeal, animal muscles, and visceral tissues.
- Chitin contend varied from 1.63 to 3.03 g/100 g FW.

RESEARCH ARTICLE

Taurine content of insects used as feed

L. Kouřimská, F. Kvasnička, M. Kurečka, A. Rajchl, P. Škvorová, M. Kulma
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Journal of Insects as Food and Feed: 0 (0)- Pages: 1 - 8

https://doi.org/10.3920/JIFF2022.0140

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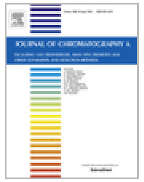
Journal of Chromatography A
Volume 1695, 26 April 2023, 463952



Electrophoretic determination of chitin in insects

František Kvasnička [a], Lenka Kouřimská [b], Roman Bleha [c], Petra Škvorová [b], Martin Kulma [d], Aleš Rajchl [a]

Conclusion

As non-protein nitrogen compounds (NPC) account for a relatively high percentage of the total nitrogen in insects, the amount of nitrogen present from NPC compounds should be considered when calculating the true protein content from the total amount of nitrogen substances.

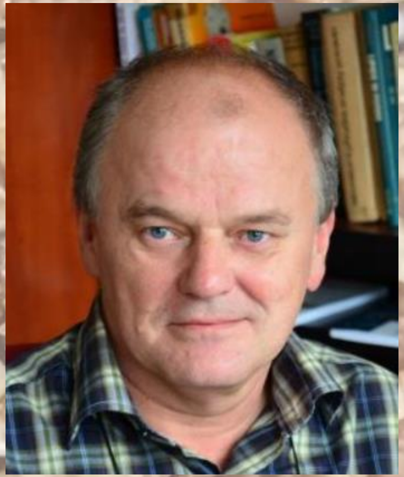
The nitrogen-to-protein conversion factor 6.25 overestimates the protein content, due to the presence of non-protein nitrogen in insects.

The conversion factor 4.76 was calculated for larvae from *Tenebrio molitor*, *Alphitobius diaperinus*, and *Hermetia illucens* (Janssen et al., 2017).

Boulos et al. (2020) calculated 5.41, 5.25, and 5.33 for mealworms, crickets, and locusts.

Thanks to my colleagues Monika, Martin, Petra, František et al.



Thank you for your attention

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This study was supported by the METROFOOD-CZ research infrastructure project (MEYS grant number: LM2018100), including access to its facilities, the Czech Science Foundation (GAČR), project number 21-47159L (INPROFF: Quality, Safety and Authenticity of Insect Protein-based Food and Feed Products), and the OP VVV, project number CZ.02.2.69/0.0/0.0/19_073/0016944, improving the quality of the internal grant scheme at the CZU.

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Research Field	Other
Researcher Profile	Established Researcher (R3)
Country	Czech Republic
Application Deadline	6 Jun 2023 - 06:06 (Europe/Prague)
Type of Contract	Permanent
Job Status	Full-time
Hours Per Week	40