Plasma sex steroids concentrations in *Xenopus laevis* exposed to atrazine in the field in South Africa.

Thanks To

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Why Atrazine?

Although banned in parts of Europe ATZ is still one of the most used broad-leave herbicides of the world (Hopenhayen-Rich et al., 2002)

ATZ is produced and used in high quantities (more than 30,000 tons are used annually in the U.S. alone; US Dept. Agr., 1994)

ATZ suggested to act as an endocrine disrupter (Colborne, 1993)

As a result of widespread use and persistence atrazine in surface waters and rainfall can be reach approximately 20 μg/L (Giddings et al., 2000)

Application of ATZ often correlates with the breeding season of aquatic vertebrates
Controversy about Atrazine (ATZ)

Effects of ATZ on *X. laevis* at concentrations as low as 0.1 ppb (Hayes et al., 2002)

Hypothesized mechanism of atrazine action via up-regulation of aromatase (Hayes et al., 2002)

Other studies could not confirm low level effects of atrazine (Carr et al., 2002; Coady et al., 2003)

Only evidence for aromatase based mechanism of action from human cell lines (Sanderson et al., 2001)
High degree of Uncertainty

Many of the results from different studies do not match.

No direct evidence for atrazine mediated induction of estrogen synthesis in laboratory and wild populations.

What is the environmental relevance of any observed effects ??
Hypothesis:

Atrazine exposure of *Xenopus leavis* in the wild affects androgen and estrogen plasma concentrations via induction of aromatase activity.
Hypothetical Mode of Atrazine Action: Induction of Aromatase Activity in Males
Study Design – Sampling

- 3 reference (R) sites
  20 males & 20 females

- 5 sites in a corn-growing region (E)
  10 males & 10 females per site

Season: Late Summer after ATZ application
Study Design - Biomarkers

- Plasma sex steroid hormones Testosterone (T) and 17b-Estradiol (E2) → *ELISA*
- **Gonadal aromatase activity → *3H-release assay***
- Pathological changes of the gonads → *histopathology*
- Pathological changes of the larynx → *histopathology*
Plasma Steroid Concentrations
(Median +/- Quartile)

Testosterone

Estradiol

*
E2/T Ratios & Aromatase Activity
(Median +/- Quartile)

male

female

E2/T Ratio

Aromatase

E2/T Ratios & Aromatase Activity
(Median +/- Quartile)
### Correlations between Biomarkers and Exposure to Atrazine and Metabolites

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Summary

• Females in agricultural areas had significantly reduced plasma hormone concentrations

• No clear effects on hormone levels in males

• No significant effects on estrogen/androgen ratios or on aromatase activity in either sex

• Significant negative relationships between Atrazine and most of its metabolites and plasma sex steroids in females
Conclusions

- There is evidence that the plasma concentrations of both T and E2 in females are affected in the agricultural region.
- Environmental effects of ATZ and its metabolites on the endocrine system of frogs cannot be excluded.
- No evidence for an aromatase-based mechanism of action.
- Need to clarify the remaining uncertainties with a focus on elucidating the modes of atrazine action and its role in the environment.
The Role of Sex Steroid Hormones

**FEMALES:**
- E2: control of exogenous yolk protein synthesis
- T: biochemical signaling for maturation and spawning
- progestins: maintenance of developing oocytes

**MALES:**
- Androgens (e.g. T, 11KT, DHT): sexual behavior and spermiation; development of secondary sex characteristics
Thank You

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