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Effects of the endocrine active compound triphenyltin on development of male and female rats after pre- and postnatal exposure

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1. Zusammenfassung

Organozinnverbindungen finden in zahlreichen Bereichen, wie z.B. industrielle Katalysatoren und Stabilisatoren oder industrielle und landwirtschaftliche Biozide Anwendung. Sie können verschiedene Auswirkungen auf die männlichen und weiblichen Reproduktionsorgane bei Nagetieren induzieren und erzeugen außerdem in solchen Organen Tumore (Hoden, Hypophyse), für deren Ursache endokrine Störungen vermutet werden können.

Untersucht wurden die Auswirkungen der Behandlung mit der Organozinnverbindung Triphenylzinn während der prä-, und postnatalen Entwicklungsphase von Ratten. Für die prä,- peri,- und postnatale Entwicklung der weiblichen wie männlichen Reproduktionsorgane spielt das Enzym Aromatase, welches die Umwandlung von Testosteron zu Östradiol katalysiert eine zentrale Rolle.

Die Untersuchungen umfassten bei männlichen Nachkommen die Körpergewichtsentwicklung, den Zeitpunkt der Präputialseparation, das Gewicht von Hoden, Nebenhoden, Samenblase und ventraler Prostata, das Gewicht von Leber und Thymus, die Testosteronkonzentration im Blut und die Messung der Aromataseaktivität im Gehirn und im Gonadengewebe.

Die Untersuchungen der weiblichen Nachkommen umfassten die Körpergewichtsentwicklung, den Zeitpunkt der Vaginalöffnung, das Gewicht von Uterus und Ovarien, das Gewicht von Leber und Thymus, die Estradiol- und Progesteronkonzentration im Blut, sowie die Messung der Aromataseaktivität im Gehirn und im Gonadengewebe.

Muttertiere (trächtige Wistar Ratten) wurden ab dem Tag 6 der Trächtigkeit bis zum Ende der Laktationsperiode per Sonde mit Dosierungen von 2 mg TPT/kg Körpergewicht und 6 mg TPT/kg Körpergewicht behandelt. Dies bedeutet, dass die Nachkommen sowohl transplazentar, als auch via Muttermilch exponiert wurden. Nach Absetzen der Nachkommen wurde eine Gruppe weiter behandelt und die andere nicht mehr exponiert.

Ausgewählte Tiere wurden an den postnatalen Tagen (PND) 1, 21 und im Erwachsenenalter (PND 58) getötet.

Die Organe der männlichen Nachkommen wurden an PND 1, PND 21, sowie PND 58 entnommen und für die weiteren Untersuchungen entsprechend aufgearbeitet. Bei den weiblichen Nachkommen wurden ab PND 58 Vaginalabstriche zur Zyklusbestimmung angefertigt und die Organe dann in der ersten Östrusphase nach PND 58 entnommen und ebenfalls für die weiteren Untersuchungen aufgearbeitet.

In der 6 mg TPTCl Dosisgruppe wurde eine verzögerte Geburt beobachtet. Darüber hinaus führte die Behandlung zu einer deutlichen Steigerung der perinatalen Mortalität, zu einer Abnahme der Gewichtszunahme in der Stillzeit sowie zu einer verzögerten körperlichen Entwicklung der Nachkommen.

Die Exposition mit 2 mg TPTCl / kg Körpergewicht führte zu einer signifikanten Erhöhung der perinatalen Mortalität und einem verzögerten Öffnen der Augen.

Es wurde ein deutlicher Geschlechtsunterschied bei der Reaktion auf die Behandlung beobachtet.

Während der postnatalen Entwicklung der männlichen Nachkommen wurde eine Abnahme des Körpergewichts, des Gewichts der Fortpflanzungsorgane und der Testosteronkonzentration beobachtet, sowie eine erhebliche Verzögerung der Präputialseparation.
Im Gegensatz dazu war bei den weiblichen Nachkommen war der Zeitpunkt der der Vaginalöffnung verfrüht, während alle anderen Endpunkte unverändert waren.
Während im Gehirn der weiblichen Nachkommen die Aktivität der Aromatase an PND 21 und bei den adulten Tieren deutlich erhöht war, zeigte sich bei den männlichen Nachkommen eine signifikante Abnahme der Aromataseaktivität im Gehirn nur bei den Adulten.
Die Aromataseaktivität in den Eierstöcken wurde zu beiden Zeitpunkten untersucht und war unverändert. Im Gegensatz dazu war die Aromataseaktivität im Hoden bei den männlichen Nachkommen an PND 21 deutlich erhöht.
Die Schlussfolgerung ist, dass TPT besonders während der vulnerablen Phase der Entwicklung die postnatalen und sexuelle Entwicklung, sowie die Aktivität der Aromatase in Rattennachkommen geschlechtsabhängig beeinflusst, wobei männliche Ratten anfälliger für Störungen durch diesen endokrinen Wirkstoff sind als die Weibchen.

1.1. Abstract

Triphenyltin (TPT) is an organotin compound (OTC) previously widely used as an antifouling agent in paints applied in the marine environment, a fungicide, and as an agricultural pesticide. Certain OTCs are immunotoxic and may also have endocrine disrupting properties resulting in adverse effects on the reproductive tract in mollusks and mammals. In recent years, many physical and behavioral disorders in humans have been attributed to the exposure to ubiquitous environmental chemicals such as OTCs. Since effects of *in utero* exposure to endocrine disrupting chemicals on the reproductive system are dependent on the critical window of exposure during its development, we conducted a comprehensive study with the aim to identify the most sensitive window of exposure to TPT and to investigate the effects of *in utero* and lactational exposure to TPT on pregnancy outcome, postnatal and sexual development as well as aromatase activity in rats. Gravid Wistar rats were treated per gavage from gestational day 6 until the end of lactation at dose levels of 2 or 6 mg TPT/kg bw. In the 6 mg TPTCl dose group, gestational mortality in dams and an increased incidence of anticipated and delayed parturition were observed. Furthermore, treatment resulted in a significant increase in perinatal mortality, a decrease in lactational body weight gain as well as in delayed physical maturation of offspring. Similarly, exposure to 2 mg TPTCl/kg b.w. resulted in a significant increase in perinatal mortality and in delayed eye opening. At weaning, the remaining offspring in the 2 mg/kg bw dose group were divided into two groups, one of which was treated until sacrifice in adulthood while the other received no further treatment. Selected animals were sacrificed on postnatal days (PND) 1, 21 and in adulthood from PND 58. A clear sex difference in response to treatment was observed. Male postnatal development was severely affected with decreases in body weight gain, reproductive organ weights and testosterone concentration as well as a significant delay in the age at preputial separation. In contrast, females exhibited a precocious completion of vaginal opening while all other

endpoints were unaffected. While brain aromatase activity was significantly increased on PND 21 and at adulthood in female offspring, male offspring exhibited a significant decrease in brain aromatase activity only at adulthood. Ovarian aromatase activity was unaffected at both time points investigated. In contrast, testicular aromatase activity was significantly increased in males on PND 21 and significantly decreased at adulthood irrespective of the duration of treatment. We conclude that TPT administered during the particularly vulnerable period of development affects postnatal and sexual development as well as aromatase activity in rat offspring in a sex-dependent manner, with the male rat being more susceptible to disturbances through this endocrine active compound than the female.

1.2. Introduction and objectives

The organotin compound (OTC) triphenyltin (TPT) was previously used extensively as a herbicide, pesticide and fungicide in agriculture as well as, together with tributyltin (TBT), in marine antifouling paints. The main source of organotin intake for humans is contaminated fish and seafood. This is a result of OTCs leaching from ship paint into the aquatic environment (Kannan and Falandysz, 1997, Takahashi *et al*., 1999, Kannan *et al*., 1999). Certain OTCs are known hormone disrupters in molluscs (Oehlmann *et al*., 1996) and have shown to inhibit a variety of enzymes responsible for the production of sex steroid hormones in higher male and female organisms (Doering et al., 2002, Lo et al., 2003). OTCs easily cross the placental as well as the blood-brain barrier in higher species (Hasan *et al*., 1984, Adeeko *et al*., 2003, Cooke *et al*., 2004) and have shown various effects on the immune (Snoeij *et al.*, 1985,1986; Vos *et al*.,1990, Tryphonas *et al*., 2004) and the reproductive system (Ema *et al*., 1999, Ogata *et al*., 2001, Omura *et al.*, 2001, Grote *et al*., 2004, 2006) in experimental studies. The toxicological mechanisms of OTCs have not yet been fully elucidated. An increase in androgen levels through inhibition of aromatase activity has been made responsible for the induction of imposex in mollusks and has also been observed *in vitro* (Heidrich *et al*., 2001; Cooke, 2002, Lo *et al*., 2003).
Due to these properties OTCs may have the potential to affect hormone-regulated developmental processes during pre- and postnatal development resulting in effects that may not be manifested until the offspring reaches maturity. The developing organism is particularly susceptible to the effects of endocrine disrupters (Gray *et al*., 1999) and the developmental vulnerability begins at conception and extends through gestation, parturition and the postnatal period up to adolescence. During development the organism undergoes many complex integrated processes and interference with those processes through e.g. altered cell division, hormone activity or enzyme function can result in significant alterations in physiological developmental processes. In an earlier study we used a modification of the Rodent 20-day pubertal female assay and the Rodent 20-day pubertal male assay to investigate the effects of TPTCl on pubertal rats (Grote *et al*., 2004, 2006). The data reported in the present publication are part of a comprehensive study, in which we aimed to further evaluate TPT and its effects on pregnancy outcome, postnatal development and aromatase activity in rats and to investigate possible differences of chemical susceptibility/sensitivity of fetuses and neonates in comparison to pubertal rats which we studied earlier at the same dose levels.

1.3. Materials and methods

1.3.1. Animals, dose selection and treatment

Wistar rat dams were administered TPT or peanut oil by daily gavage from day 6 of gestation (mating=day 0) to day 21 of lactation at dose levels of 2 or 6 mg/kg bw/day.
One group of animals received only peanut oil and served as vehicle control. A total number of 14-17 rat dams (litters) per group were used. Dose selection was based on results from our previous studies in male and female pubertal rats (Grote et al., 2004, 2006). Dams in the 6 mg TPTCl/kg b.w. dose group exhibited a high rate of mortality. Due to the high number of either stillborn or dead offspring until PND 4 and the resulting insufficient number of offspring no further evaluation was carried out after weaning at this dose level. After weaning on postnatal day (PND) 21 the remaining offspring in the 2 mg TPT/kg bw treatment group were divided into two groups. In one group treatment was continued until termination, the other group received no further treatment. The same procedure was applied to offspring of the vehicle control, resulting in a separate control group for each 2 mg TPTCl treatment group.

1.3.2. Evaluation of dams

In dams the following parameters of pregnancy outcome and reproductive success were evaluated:

- Gestational and lactational body weight gain
- Day of parturition
- Litter size
- Litter weight
- Number of live and stillborn pups at birth

1.3.3. Evaluation of offspring

Offspring were investigated at different stages of life to detect possible effects on development and induction of effects on reproductive and endocrine functions. A list of the endpoints evaluated is shown in table 1.

Table 1. Endpoints investigated in offspring exposed *in utero* and during lactation to TPT

Age	Number of animals	Endpoints
PND 1	11-13 offspring/sex/dose group	Body weights Brain hypothalamic pre-optic area (HPOA) aromatase activity.
From PND 6	All available offspring	Evaluation of developmental landmarks: fur development, tooth eruption, eye opening, testes descent
PND 21	11-13 offspring/sex/dose group	Lactational body weight gain Brain hypothalamic pre-optic area (HPOA) aromatase activity. Gonadal aromatase activity
From PND 30	13-27 females/dose group	Completion of vaginal opening
From PND 35	13-28 males/dose group	Completion of preputial separation
PND 58	11-15 males/dose group	Body weights and body weight gain Organ weights Testosterone concentrations Brain hypothalamic pre-optic area (HPOA) aromatase activity. Gonadal aromatase activity
From PND 58 (at estrus determined by vaginal smear)		Body weights and body weight gain Organ weights Estradiol and progesterone concentrations Ovarian follicle count Brain hypothalamic pre-optic area (HPOA) aromatase activity. Gonadal aromatase activity

1.4. Results and Discussion

1.4.1. Effects on pregnancy outcome and postnatal development

(Grote K., **Hobler C**, Andrade A.J.M., Wichert Grande S., Gericke C., Talsness C.E., Appel K.E., Chahoud I., 2007. Toxicology 238, 177 185)

Endocrine disrupters (Eds) are capable of activating or blocking hormone receptors, in particular estrogen or androgen receptors, and may interfere with synthesis and metabolism of steroid hormones. The developing organism is particularly susceptible to the effects of endocrine disrupters (Gray *et al*., 1999) and the developmental vulnerability begins at conception and extends through gestation, parturition and the postnatal period up to adolescence.

In the present study, mortality of gravid dams was observed after exposure to 6 mg TPTCl/kg b.w., while no mortality or other signs of general toxicity were observed in the 2 mg TPTCl dose group. Dams in the 6 mg TPTCl dose group exhibited a significant increase in anticipated and delayed parturitions compared to the control animals. This effect could have been caused by an indirect impact of TPTCl disrupting the homeostasis needed for the maintenance of late pregnancy and for the hormonal preparation of parturition in the dams. In particular the increased number of dams that delivered their pups on GD 24 indicates a possible dystocia as in our historical control data this finding was observed in only 3 out of 292 dams. It has been shown that treatment of pseudopregnant rats with 6.3 mg TPTCl/ kg b.w. and 16.5 or 24.8 mg diphenyltin/kg b.w. resulted in a significant decrease in serum levels of the pregnancy maintaining hormone progesterone (Ema *et al*., 1999; Ema and Miyawaki, 2002). Baroncelli *et al*. (1995) also reported a significant shift in the time of parturition after exposure of pregnant mice to the organotin compound bis(tri-n-butyltin)oxide (TBTO) at dose levels from 5 to 30 mg/kg b.w.

Mean litter size as well as distribution of litter size between the classified groups were not significantly affected by TPT treatment.

Exposure to both doses of TPT significantly increased the number of stillborn pups compared to the vehicle control. On PND 2 a significantly increased number of pups died in both dose groups compared to control, while on PND 3 and 4 only the 6 mg TPTCl dose group exhibited a significant increase in offspring mortality. No postnatal mortality was observed after PND 4. This observation is in agreement with reports from the study of Ema *et al*. (1999) in which treatment with TPTCl at dose levels between 6.3 and 12.5 mg/kg bw led to a significant increase in the number of dead fetuses when uteri were examined on GD 20. The authors suggest that the embryolethal effects observed in their study may be caused by direct fetotoxic effects of the substance or hormonal disruption in the dams.

Offspring mean body weights were significantly decreased during the first two days of lactation in the 6 mg TPTCl dose group when compared to the control animals. After PND 2 no significant difference in lactational body weight gain was observed in this dose group. This is in agreement with data from the

studies with butyltins of Cooke *et al*. (2004) and Adeeko *et al*. (2003) reporting decreased offspring body weights on PND 1 and on the day of caesarean section, respectively.

The indicative developmental landmarks fur development, tooth eruption, eye opening and testes descent in male offspring were significantly delayed in the 6 mg TPTCl dose group. In the 2 mg TPTCl dose group offspring showed a statistically significant delay only in the day of eye opening, which on PND 16 occurred in approx. 70% of the animals compared to 90% in the control group. The delayed physical maturation in the present study observed in the 6 mg TPTCl dose group might be primarily due to the suppressed body weights on PND 1 and 2. With regard to the androgen dependent parameter testes descent, however, a possible antiandrogenic impact by TPTCl should also be considered. Omura *et al*. (2001) also observed a delay in eye opening after *in utero* exposure to approx. 2 and 10 mg TBTCl in male rat offspring, while no effect on testes descent was detected.

1.4.2. Effects on offspring sexual development

(Grote K., **Hobler C**., Andrade A.J.M., Wichert Grande S., Gericke C., Talsness,C.E., Appel K.E., Chahoud, I., 2009. Toxicology 260, 53-59)

The aim of the present study was to investigate whether rats are more susceptible to exposure of TPTCl at the same dose levels during the sensitive period of pre- and postnatal development and whether a possible impact caused perinatally would be reversible.

The surviving offspring in the 2 mg/kg bw/d dose group exhibited a clear sex-dependent difference regarding susceptibility to effects induced by developmental exposure to TPTCl in most endpoints investigated. While body weight development in female offspring was unaffected, body weights of surviving male offspring were similar to the control until the end of lactation but exhibited a decrease in body weight gain during the postweaning period that gained statistical significance around the onset of puberty even in animals that received no further treatment. At termination male body weights were significantly decreased in both treatment groups given 2 mg TPTCl/kg bw during different durations of treatment. Given that fact, exposure to TPTCl may have caused an impact either in *utero* or during lactation that resulted in an irreversible effect on bodyweight development only detectable around the beginning of puberty when sexual hormones are beginning to play an important role in male physical development.

While liver and thymus weights in female offspring were unaffected, male offspring exhibited a significant decrease in absolute organ weights.

We observed a clear sex-dependent difference in the onset of puberty that did not correlate with the duration of treatment. Male offspring exhibited a significant delay in the completion of PS with a significant decrease in testosterone concentration only being present in the animals exposed until termination. In female offspring, however, perinatal exposure to TPTCl resulted in a significantly earlier completion of vaginal opening in both treatment groups but did not alter progesterone or estradiol levels. The effects on male sexual development are in contrast to our previously reported results obtained from the male pubertal assay (Grote *et al*., 2004) in which no changes in the completion of

PS after treatment with 2 TPTCl/kg bw were observed. The accelerating effects on sexual development observed in female offspring are in agreement with results from our previous study, in which peripubertal treatment with 2 mg TPTCl/kg also caused precocious completion of vaginal opening while sexual hormone levels remained unaffected (Grote *et al*., 2006). In males, absolute testes weights were significantly decreased in both treatment groups while absolute epididymal weights were decreased only in the animals that received treatment until weaning. Furthermore, a significant decrease in absolute and relative prostate weight occurred in the group undergoing longer treatment only. Absolute weights of seminal vesicle were decreased irrespective of the duration of treatment. In female offspring, neither reproductive organ weights nor ovarian follicle counts were affected by treatment. In conclusion, both postnatal development of the male reproductive tract and sexual maturation in male and female offspring were affected. Most of these effects were more pronounced in male than in female offspring and were also present in animals that were only exposed until weaning, indicating that these effects may be irreversible. Continued treatment until termination seemed to have contributed less than expected to the severity of the observed effects. The results suggest that in this study the sensitive window for these endpoints seems to be the period of prenatal development and that male offspring rats were more susceptible to treatment.

1.4.3. Effects on brain and gonadal aromatase activity

(**Hobler C**, Andrade AJ, Grande SW, Gericke C, Talsness CE, Appel KE, Chahoud I, Grote K. Toxicology 2010, 276: 198-205)

OTC-induced environmental endocrine disruption occurs in fish and mammals and a number of *in vivo* and *in vitro* studies have argued that OTCs may act through inhibition of the aromatase enzyme. *In vivo* studies supporting the aromatase inhibition hypothesis in mammals are lacking. In the present study, we investigated the effects of perinatal treatment with 2 mg TPTCl/kg bw on gonadal and brain aromatase activity in order to evaluate whether altered aromatase activity serves as a potential mediator of the previously observed effects of TPT on postnatal development.

There was an obvious, sex-dependent qualitative difference in the response to treatment. Male rats exhibited a decrease in brain aromatase activity on PND 21 which gained statistical significance at adulthood in the group treated until termination on PND 58. Evaluation of testicular aromatase activity revealed a significant increase on PND 21 followed by a significant decrease at adulthood in animals treated until weaning, as well as in animals treated until termination. In contrast, exposure of female rats to 2 mg TPTCl/kg bw resulted in a significant increase in brain aromatase activity on PND 21 and at adulthood in those animals treated until termination. Ovarian aromatase activity remained unaffected at all time points investigated irrespective of the duration of treatment. These results suggest that the duration of treatment played a decisive role in the characteristic of the effects in the brain. The changes in brain aromatase activity on PND 21 in both sexes could not be observed in adult animals that did not receive further treatment suggesting a possible reversibility of these effects.

In contrast, the decrease in testicular aromatase observed in young adult males that were only exposed until PND 21 indicates that TPT has prolonged effects on testicular aromatase.

In vivo and *in vitro* effects of OTCs on aromatase activity have been frequently reported in different species including humans. In female mollusks, exposure to OTCs caused the development of male sex characteristics, a phenomenon known as imposex (Horiguchi et al., 1995; Oehlmann et al.,1996; Matthiessen and Gibbs, 1998). Similarly, masculinization of OTC, exposed female fish has been observed (McAllister and Kime, 2003; Shimasaki et al., 2003; Santos et al., 2005). These alterations occurred at low exposure levels and were attributed to an increase in androgen concentrations through the inhibition of aromatase. The molecular target of OTCs in mammals has not yet been identified but there is evidence that OTCs bind as agonists to several nuclear receptors and thus modulate homeostasis of sex steroids through transcriptional up- or down-regulation of aromatase gene expression (Saitoh et al., 2001; Kanayama et al., 2005; Nakanishi et al., 2005). Furthermore, it has been reported that OTCs enhance histone acetyltransferase activity in rat liver nuclear extracts (Osada et al., 2005) and that they suppress testosterone synthesis via decreased expression of steroidogenic enzymes (Doering et al., 2002; Lo et al., 2003; Nakajima et al., 2003; Ohno et al., 2005; Reddy et al., 2006; Kim et al., 2008). It remains unclear whether the effects on aromatase activity reported in the present investigation are primarily mediated by direct enzyme inhibition or rather by modulation of the aromatase gene expression. The interpretation of the observed effects on aromatase is complicated by interdependent hormonal regulation mechanisms, gender-specificity for OTC-toxicity, critical window of exposure as well as the existence of other molecular targets for OTC-toxicity. Nevertheless, based on the results of this study, we can confirm that TPT administered during the particularly vulnerable period of development affects aromatase activity in rodents.

1.5. Conclusion

The present study provides comprehensive data regarding the effects of TPT on pregnancy outcome, postnatal development and aromatase activity after *in utero* exposure of rats. Treatment severely affected pregnancy outcome and perinatal survival of offspring. Furthermore, postnatal development of the male reproductive tract as well as sexual maturation in male and female offspring were affected. *In utero* exposure to TPT also resulted in a significant reduction in male brain and testicular aromatase activity at adulthood, while adult female rats exhibited an increase in brain aromatase activity only. Most effects observed were dependent on age, duration of treatment, sex-dependent susceptibility and organ specificity. This study provides evidence that age-related effects on susceptibility to a given compound should be taken into consideration and adequate attention should be given to the greater sensitivity of the developing organism to chemical toxicity.

In reproductive toxicology effects are exerted through complex mechanisms that are influenced by factors, such as dose, age and gender. These factors should be taken into account when assessing the risk of hormonally active environmental chemicals and appropriate precaution measures should be taken to protect children as the subpopulation most susceptible to endocrine disruption.

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2. Anteilserklärung

Carolin Hobler hatte folgenden Anteil an den vorgelegten Publikationen:

1. Publikation:

Grote, K., **Hobler, C**, Andrade, A.J.M., Wichert Grande, S., Gericke, C., Talsness, C.E., Appel, K.E., Chahoud, I., 2007. Effects of *in utero* and lactational exposure to triphenyltin chloride on pregnancy outcome and postnatal development in rat offspring. Toxicology 238, 177 185

40 Prozent

Beitrag im Einzelnen:

Durchführung der Experimente, Auswertung

2. Publikation:

Grote, K., **Hobler, C**., Andrade, A.J.M., Wichert Grande, S., Gericke, C., Talsness, C.E., Appel, K.E., Ibrahim Chahoud, I., 2009. Sex differences in effects on sexual development in rat offspring after pre- and postnatal exposure to triphenyltin chloride. Toxicology 260, 53-59

40 Prozent

Beitrag im Einzelnen:

Durchführung der Experimente, Auswertung

3. Publikation:

Hobler, C, Andrade, AJ, Grande, SW, Gericke, C, Talsness, CE, Appel, KE, Chahoud, I, Grote, K., (2010) Sex-dependent aromatase activity in rat offspring after pre- and postnatal exposure to triphenyltin chloride. Toxicology , 276: 198-205

60 Prozent

Beitrag im Einzelnen:

Durchführung der Experimente, Aromatase-Messungen, Literaturrecherche, Abfassen der Publikation.

3. Publikationen

1.)

Hobler, C, Andrade, AJ, Grande, SW, Gericke, C, Talsness, CE, Appel, KE, Chahoud, I, Grote, K., 201**0.** Sex-dependent aromatase activity in rat offspring after pre- and postnatal exposure to triphenyltin chloride. Toxicology **,** 276: 198-205

2.)

Grote, K, **Hobler, C**, Andrade, AJM, Wichert Grande, S, Gericke, C, Talsness, CE, Appel, KE, Chahoud, I, 2007. Effects of *in utero* and lactational exposure to triphenyltin chloride on pregnancy outcome and postnatal development in rat offspring. Toxicology 238, 177 185

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Grote, K, **Hobler, C**, Andrade, AJM, Wichert Grande, S, Gericke, C, Talsness, CE, Appel, KE, Ibrahim Chahoud, I, 2009. Sex differences in effects on sexual development in rat offspring after pre- and postnatal exposure to triphenyltin chloride. Toxicology 260, 53-59

4. Lebenslauf

Mein Lebenslauf wird aus datenschutzrechtlichen Gründen in der elektronischen Version meiner Arbeit nicht veröffentlicht.

5. Liste eigener Publikationen

1.)

Hobler, C, Andrade, AJ, Grande, SW, Gericke, C, Talsness, CE, Appel, KE, Chahoud, I, Grote, K., 201**0.** Sex-dependent aromatase activity in rat offspring after pre- and postnatal exposure to triphenyltin chloride. Toxicology **,** 276: 198-205

2.)

Grote, K, **Hobler, C**, Andrade, AJM, Wichert Grande, S, Gericke, C, Talsness, CE, Appel, KE, Chahoud, I, 2007. Effects of *in utero* and lactational exposure to triphenyltin chloride on pregnancy outcome and postnatal development in rat offspring. Toxicology 238, 177 185

3.)

Grote, K, **Hobler, C**, Andrade, AJM, Wichert Grande, S, Gericke, C, Talsness, CE, Appel, KE, Ibrahim Chahoud, I, 2009. Sex differences in effects on sexual development in rat offspring after pre- and postnatal exposure to triphenyltin chloride. Toxicology 260, 53-59

6. Selbständigkeitserklärung

„Ich, Carolin Hobler, erkläre, dass ich die vorgelegte Dissertation mit dem Thema: Effects of the endocrine active compound triphenyltin on development of male and female rats after pre- and postnatal exposure selbst verfasst und keine anderen als die angegebenen Quellen und Hilfsmittel benutzt, ohne die (unzulässige) Hilfe Dritter verfasst und auch in Teilen keine Kopien anderer Arbeiten dargestellt habe.“

7. Danksagung

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