

# Absence of Effect of Intravaginal Miconazole, Clindamycin, Nonoxynol-9, and Tampons on the Pharmacokinetics of an Anastrozole/Levonorgestrel Intravaginal Ring

The Journal of Clinical Pharmacology  
2018, 58(1) 97–106  
© 2017, The Authors. *The Journal of Clinical Pharmacology* published by Wiley Periodicals, Inc. on behalf of American College of Clinical Pharmacology  
DOI: 10.1002/jcph.984

Rüdiger Nave, PhD<sup>1</sup>, Stefan Klein, PhD<sup>1</sup>, André Müller, MD<sup>2</sup>, Xinying Chang, MD<sup>1</sup>, and Joachim Höchel, PhD<sup>1</sup>

## Abstract

A study was performed to investigate the effect of an intravaginally administered antimycotic, an antibiotic, and a spermicide plus the co-usage of tampons on the pharmacokinetics (PK) of levonorgestrel (LNG) and anastrozole (ATZ) administered as an intravaginal ring (IVR) releasing 1050  $\mu\text{g}$  ATZ per day and 40  $\mu\text{g}$  LNG per day. In this parallel-group, randomized, open-label study, healthy premenopausal women received an IVR as the main treatment. Comedications were administered on 3 consecutive evenings during treatment with IVR on days 9–11 (group A, 400 mg miconazole; group B, 100 mg clindamycin; group C, 75 mg nonoxynol-9); tampon co-usage (group D) was performed on days 20–23. The primary PK parameter was the average plasma concentration ( $C_{\text{av,ss}}$ ) of ATZ and LNG at defined intervals, mainly prior to, during, and up to 7 days after the start of comedication. Fifty-two subjects were included, and at least 11 subjects per group completed the treatments. Overall, the medications and comedications were safe and well tolerated. Very similar ATZ and LNG plasma levels were observed across all groups. The calculated ratios of  $C_{\text{av,ss}}$  confirmed the absence of PK interactions because all relevant point estimates and 90% confidence intervals were within the range of 0.800–1.250, which is typically used in bioequivalence studies. These results demonstrate the absence of PK interactions between ATZ/LNG released from IVR and the tested antibiotic, antimycotic, spermicide, and tampons. Therefore, no restrictions for the use of the IVR are needed to continue the clinical program intended to treat endometriosis symptoms.

## Keywords

drug-drug interaction, women's health, gynecology, drug delivery, clinical pharmacology, pharmacokinetics and drug metabolism

Endometriosis is a chronic inflammatory disease affecting up to 10% of women of reproductive age.<sup>1,2</sup> Common symptoms, such as chronic pelvic pain, dysmenorrhea, and dyspareunia, can have a severe impact on patients' physical, social, and psychological functioning.<sup>3,4</sup> The disease is known to be estrogen dependent; therefore, a new treatment concept is aimed at the reduction of local estrogen production in endometriotic lesions using aromatase inhibitors such as anastrozole (ATZ).<sup>5</sup> Aromatase is a key enzyme involved in the biosynthesis of estrogens and has been shown to be locally overexpressed in endometriotic lesions.<sup>6–9</sup>

ATZ is licensed for the treatment of breast cancer in postmenopausal women<sup>10</sup> and has been shown to reduce endometriosis-associated pain in small-scale clinical trials<sup>11–13</sup> but is not approved for the treatment of endometriosis. Furthermore, ATZ may cause harm to the fetus, as an effect on fetal development was observed in preclinical studies. Therefore, effective contraception is required for women of childbearing potential when treated with ATZ.

An intravaginal ring (IVR) simultaneously releasing ATZ and levonorgestrel (LNG) for the treatment of

symptoms associated with endometriosis was developed. This is seen as an attractive approach to treat endometriosis-associated pelvic pain using an IVR that provides systemic delivery of ATZ and a low dose of LNG as a contraceptive. LNG is a well-characterized drug product approved for use as a contraceptive in various delivery systems: progestin-only pill (eg, Norgeston, corresponding to Microlut in other countries), subcutaneous implant (Jadelle), or intrauterine system (Mirena).

<sup>1</sup>Research and Development, Bayer AG, Berlin, Germany

<sup>2</sup>CRS, Berlin, Germany

This is an open access article under the terms of the Creative Commons Attribution-NonCommercial License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited and is not used for commercial purposes.

Submitted for publication 12 April 2017; accepted 21 June 2017.

## Corresponding Author:

Rüdiger Nave, PhD, Director Pharmacokinetic Expert, Bayer AG, Drug Discovery, Clinical Pharmacokinetics, Müllerstraße 178, 13353 Berlin, Germany

Email: ruediger.nave@bayer.com

One phase 1 study has been completed in Europe investigating the pharmacokinetics (PK), pharmacodynamics (PD), safety, and tolerability of IVRs delivering ATZ/LNG in healthy, ovulating women aged 18–35 years.<sup>14</sup> The results showed that doses of 40  $\mu\text{g}$  LNG/day combined with up to 1050  $\mu\text{g}$  ATZ/day reached the anticipated exposure levels for both drugs.<sup>15</sup>

Considering the route of administration, other intravaginally applied medications (eg, antimycotics and antibiotics for the treatment of vaginal infections) may be necessary concomitantly with the use of the IVR, which is intended for long-term treatment of endometriosis. Furthermore, spermicides are commonly used intravaginally as vaginal suppositories, coated condoms, or tampons. To allow such concomitant use, it needs to be excluded that such intravaginal treatments affect the release or absorption of ATZ or LNG from the IVR. In the present study, the effect of concomitant intravaginal use of the antimycotic miconazole (Gynodaktarin vaginal capsule),<sup>16</sup> the antibiotic clindamycin (Sobelin vaginal creme),<sup>17</sup> the spermicide nonoxynol-9 (Patentex oval),<sup>18</sup> and tampons<sup>19</sup> on the PK of ATZ and LNG released from IVR was investigated.

## Methods

The study (EudraCT number 2014-005167-32) was approved by the independent ethics committee institutional review board (Landesamt für Gesundheit und Soziales, Ethik-Kommission des Landes, Berlin, Germany), and all volunteers provided written informed consent.

### Dose Selection

For this study, a high dose was chosen. The nominal in vitro release rate of the IVR at the end of the wearing period of 4 weeks was 1050  $\mu\text{g}$  ATZ/day + 40  $\mu\text{g}$  LNG/day. The release rate of ATZ from the IVR achieves a maximum systemic exposure that is lower compared with the steady-state exposure after oral ATZ 1 mg/day (Arimidex). The release rate and dose of LNG resulted in an exposure similar to that of the approved low-dose progestin-only LNG pill (Norgeston/Microlut, 30  $\mu\text{g}$ /day) and the LNG implant (Norplant II/Jadelle) after a wearing period of 2 years.

The IVRs were made of silicone elastomer with an outer diameter of 54 mm and a cross-sectional diameter of 5 mm.

### Study Design

This investigation was a randomized, parallel-group, 4-arm, open-label study conducted at a single center in Germany. Following screening and assessment of the premenopausal women for eligibility to participate in the trial, the study participants were allocated equally to 1 of 4 treatment groups. Randomization was carried

out using a computer-generated randomization list provided by a group independent of the investigator. The screening procedures including physical examination, 12-lead electrocardiogram, vital signs, drug screening, safety laboratory parameters and gynecological examination should be performed within 6 weeks prior to administration of the first study drug.

All subjects received an IVR releasing 1050  $\mu\text{g}$  ATZ/day + 40  $\mu\text{g}$  LNG/day as a main treatment, which started between the second and fifth days of the subjects' menstrual bleeding. The IVRs were inserted by a gynecologist on day 1 in the morning. After the maximum exposure ( $C_{\text{max}}$ ) of ATZ and LNG was reached within the first week, 1 of 4 treatments (group A, miconazole; group B, clindamycin; group C, nonoxynol-9; or group D, tampons) was applied intravaginally in addition to the IVR for 3 consecutive days. For groups A, B, and C, comedications were administered in the evening (at bedtime) of days 9–11. For group D, tampon use started on day 20 in the evening, and the last tampon was removed on day 23 in the evening. In total, 4 tampons per day were planned, and all tampons were inserted and removed by the subjects themselves.

In addition, an extended wearing duration of 35 days without changing the ring and the elimination of ATZ and LNG after IVR removal were investigated in group D.

An overview of the study design per treatment group is given in Table 1.

### Healthy Volunteers

Healthy women aged 18–50 years with a body mass index between 18 and 30  $\text{kg}/\text{m}^2$  were eligible for inclusion in the study. Key exclusion criteria included conditions/diseases known to deteriorate with pregnancy or hormonal contraceptives; preexisting diseases that might affect the absorption, distribution, metabolism and elimination of the study drugs; known or suspected malignant tumors; known or suspected benign tumors of the liver, pituitary, or adrenal gland; known or suspected liver disorders; severe renal impairment; venous or arterial thromboembolic disease; preexisting ischemic heart diseases or increased susceptibility to these diseases; use of hormonal contraceptives; use of drugs inducing or inhibiting metabolizing liver enzymes; smoking more than 10 cigarettes a day; and regular alcohol consumption of more than 20 g/day.

### Treatments

For the IVR, the dose per day for ATZ and LNG reflected nominal doses based on an intended in vitro release rate of ATZ 1050  $\mu\text{g}/\text{day}$  and LNG 40  $\mu\text{g}/\text{day}$  at the end of the intended wearing period of 28 days. The same batch was used for all treatments in this

**Table I.** Study Flow Chart for All Groups (Top) and for Group D (Bottom)

Time: Study Day (Relative Time)	Pre-dose	4	8	9	10	11	12	13	14	15	16		
Ambulant visit	X	X	X	X				X	X	X	X		
Overnight phase <sup>b</sup>				X	→	→	X						
Insertion of intravaginal ring	X <sup>a</sup>												
Removal of intravaginal ring											X		
Treatment with miconazole (group A), clindamycin (group B) or nonoxynol-9 (group C) — evening				X	X	X							
Blood sample for pharmacokinetics (profile)			X	X	X	X	X						
Blood sample for pharmacokinetics — morning	X	X						X	X	X	X		
Blood sample for SHBG — morning	X	X		X	X	X	X		X		X		
C <sub>av</sub> (reference)			X→	X									
C <sub>av</sub> (DDI 72 hours) during treatment <sup>c</sup>				X	→	→	X						
C <sub>av</sub> (DDI 158 hours) during and after treatment <sup>c</sup>				X	→	→	→	→	→	→	X		
Time: Study Day (Relative Time)	Up to Day 16	20	21	22	23	24	25	26	27	28	30	32	35
Ambulant visit		X	X	X	X	X	X	X	X	X	X	X	X
Overnight phase		X	→	→	X								
Removal of intra-vaginal ring													X
Insertion of tampons – start in the evening	see	X	X	X									
Removal of last tampon					X								
Blood sample for pharmacokinetics (profile)	table	X	X	X	X								
Blood sample for pharmacokinetics — morning						X	X	X	X	X	X	X	X
Blood sample for SHBG — morning	above	X	X	X	X		X		X		X		X
C <sub>av</sub> (reference tampons)		X→X											
C <sub>av</sub> (DDI 72 hours) during tampon use		X	→	→	X								
C <sub>av</sub> (DDI 158 hours) during and after tampon use		X	→	→	→	→	→	→	X				
C <sub>av</sub> (extended wearing period)										X	→	→	X

X action to be done at the time indicated.

→ action to be done continuously, starting from the time indicated.

<sup>a</sup>Insertion of the intravaginal ring will be done on days 2–5 of menstrual bleeding.

<sup>b</sup>In addition for group A and B, overnight stays until day 15; no overnight stays for group D.

<sup>c</sup>No tampon use in group D.

study, and the actual in vitro release data are available for up to 35 days. The average in vitro release rates for ATZ and LNG over the wearing period of 16 days were 1249 and 43.5  $\mu\text{g}/\text{day}$  respectively, and the corresponding rates over 35 days were 1102 and 42.0  $\mu\text{g}/\text{day}$ . A study duration of 16 days is justified. Stable plasma concentrations for both drugs can be reached in approximately 1 week, thus allowing for 3 days for comedication as well as an additional observation period of 3.5 days.

On days 9–11 of IVR use, the following treatments were administered in the evening at bedtime by a physician.

- Group A: 400 mg miconazole (Gyno-Daktarin)<sup>16</sup>;
- Group B: 100 mg clindamycin (Sobelin vaginal creme)<sup>17</sup>;
- Group C: 75 mg nonoxynol-9 (Patentex oval).<sup>18</sup>

If bleeding (classified as more than “spotting”) occurred during day 9 of IVR use, the above-mentioned

treatments were not allowed to be postponed, and consequently the subject was excluded from the study.

The use of tampons in group D was independent of bleeding status.

#### Pharmacokinetics

Blood samples for the pharmacokinetic analysis were taken: before IVR insertion and 96, 202, 216, 226, 232, 238, 250, 256, 262, 274, 280, 286, 298, 312, 336, 360, and 384 hours (16 days) after IVR insertion for groups A, B, and C. The samples taken on days 8 and 9 (202, 216, and 226 hours) served as the reference period. For group D, samples were taken at almost the same times up to 384 hours after IVR insertion (exceptions: 240 hours instead of 232 and 238 hours, 264 hours instead of 256 and 262 hours, 288 hours instead of 280 and 286 hours). In addition, there were time 466, 480, 490, 496, 502, 514, 520, 526, 538, 544, 550, 562, 576, 600, 624, 648, 672, 720, 768, and 840 hours (35 days) after IVR insertion. Furthermore, samples were taken 3, 24, 48, 72, 120, and 168 hours after IVR removal.

Plasma concentrations of ATZ and LNG were determined using validated liquid chromatography–tandem mass spectrometry methods. Quality control and calibration samples were analyzed concurrently. For ATZ, the lower limit of quantification (LLOQ) was 0.10  $\mu\text{g/L}$ . Quality control (QC) samples for ATZ above the LLOQ were determined with an accuracy of 98%–102% and a precision of 2.43%–12.79%. The LLOQ for LNG was 0.01  $\mu\text{g/L}$ ; at the LLOQ, the accuracy was 101% and the precision 6.5%. QC samples for LNG above the LLOQ were determined with an accuracy of 94.3%–97.2% and a precision of 2.04%–4.24%.

The primary pharmacokinetic variable was the average concentration ( $C_{\text{av,ss}}$ ) of ATZ and LNG in plasma at defined intervals after insertion of the IVR as indicated in Table 1. For all treatment groups, the  $C_{\text{av}}$  (reference),  $C_{\text{av}}$  (drug–drug interaction [DDI] 72 hours), and  $C_{\text{av}}$  (DDI 158 hours) were estimated as a reference (interval, 202–226 hours), during treatment (interval, 226–298 hours), or during and after treatment (interval, 226–384 hours), respectively. The DDI intervals were chosen to investigate acute effects on the absorption during comedication as well as a potential prolonged effect including 3.5 days thereafter. In addition for group D the corresponding  $C_{\text{av}}$  prior (reference T; interval, 466–490 hours), during (DDI 72 hours T; interval, 490–562 hours), or during and after (DDI 158 hours T; interval, 490–648 hours) tampon usage were estimated as well as the concentrations on day 28 and day 35.

Terminal half-life after removal of the IVR in group D was estimated for ATZ and LNG.

Sex hormone-binding globulin (SHBG) data are of interest because of the interaction of LNG with SHBG, that is, the binding of LNG to SHBG and the inhibitive effect of LNG on SHBG synthesis.<sup>20,21</sup>

Blood samples were collected for the determination of SHBG concentrations in serum using an immunoassay during treatment (7 or 15 time points over 16 or 35 days of the IVR-wearing period, respectively) as well as prior to the start of and after treatment. The calibration range of the procedure was from 10 (LLOQ) to 512 nmol/L. QC samples in the concentration range from 20 to 384 nmol/L were determined with an accuracy of 105% to 109% and a precision of 6.3% to 9.5%. No PK evaluation was done for SHBG concentrations; however, the serum concentration was summarized by descriptive analysis.

#### Safety and Tolerability

Adverse events (AEs) were reported by the study volunteers spontaneously or on questioning and were recorded regularly until the follow-up visit. Safety laboratory parameters as well as vital signs were assessed at regular intervals during the study.

The gynecological examinations included a cytological cervical smear and a transvaginal ultrasound examination (TVU) as part of the screening examination to detect abnormalities that represent exclusion criteria or AEs. They were performed by a gynecologist at the study site. Furthermore, visual assessment of the vagina was performed once daily during the application time of the comedication/tampon. TVU was repeated in the follow-up period 14 days after removal of the IVR. Any findings during the study were documented as AEs. The findings in the TVU were based on clinical expertise and ovarian cysts with an average diameter of  $\geq 30$  mm were defined as an AE.

#### Statistical Analysis

This was an exploratory study; hence, no confirmatory statistical analysis was performed. Exploratory statistical analysis was done per treatment. The study was planned with 13 participants per treatment arm. Subjects who dropped out of the study or had major protocol deviations were not replaced, as long as the overall group size did not drop below 11 participants.

Using 11 subjects per treatment group, the 90% CIs for the intraindividual ratios were planned to fall within the interval ( $0.8\bar{x}$ ;  $1.25\bar{x}$ ) with a probability of 79%, assuming a coefficient of variation  $\leq 0.35$ . Under a coefficient of variation of 0.4, the 90% CIs for the ratios described above were planned to fall within the interval ( $0.75\bar{x}$ ;  $1.33\bar{x}$ ) with a probability of 92%.

Here,  $\bar{x}$  denotes the geometric mean. PK parameters were assumed to be lognormally distributed. Assumptions on the coefficient of variation were derived from a reanalysis of differences between logarithmized trough levels for ATZ and LNG in a previous study.<sup>19</sup> Power and sample size calculations were performed using SAS version 9.2, PROC POWER. PK data are presented as geometric means with percent coefficient of variation (% CV), unless otherwise specified.

## Results

#### Healthy Volunteers

In total, 87 women were screened, and 52 subjects were randomized to 1 of the 4 treatment groups. All randomized subjects were included in the safety analysis set. They were white with a mean age at screening of 37.1 years and had a mean body mass index of 23.4  $\text{kg/m}^2$ . Of the 52 treated subjects, 47 subjects (90.4%) completed the study course as planned, and 49 subjects (94.2%) were included in the PK evaluation. Three subjects were excluded from the analysis of PK data because of early treatment termination prior to the administration of any comedication. In 2 cases in group A, miconazole was not administered because of an AE (bacterial vaginosis or bleeding on day 9), whereas in

**Table 2.** Demographics for Treatment Groups (Mean  $\pm$  SD, PK Analysis Set, n = 49)

	Group A (Miconazole), n = 11	Group B (Clindamycin), n = 12	Group C (Nonoxynol-9), n = 13	Group D (Tampon), n = 13	Total, n = 49
Age (years)	36.9 $\pm$ 8.5	34.8 $\pm$ 8.0	37.4 $\pm$ 7.8	37.8 $\pm$ 8.5	36.8 $\pm$ 8.0
Weight (kg)	63.60 $\pm$ 10.71	64.38 $\pm$ 6.84	68.38 $\pm$ 8.77	65.08 $\pm$ 6.34	65.45 $\pm$ 8.21
BMI (kg/m <sup>2</sup> )	22.68 $\pm$ 3.19	22.95 $\pm$ 2.18	24.19 $\pm$ 3.19	23.60 $\pm$ 1.46	23.39 $\pm$ 2.57

1 case in group B, clindamycin was not administered because of an AE (bacterial vaginosis on day 9).

The descriptive statistics for the demographic data of the 49 subjects included in the PK set are provided in Table 2.

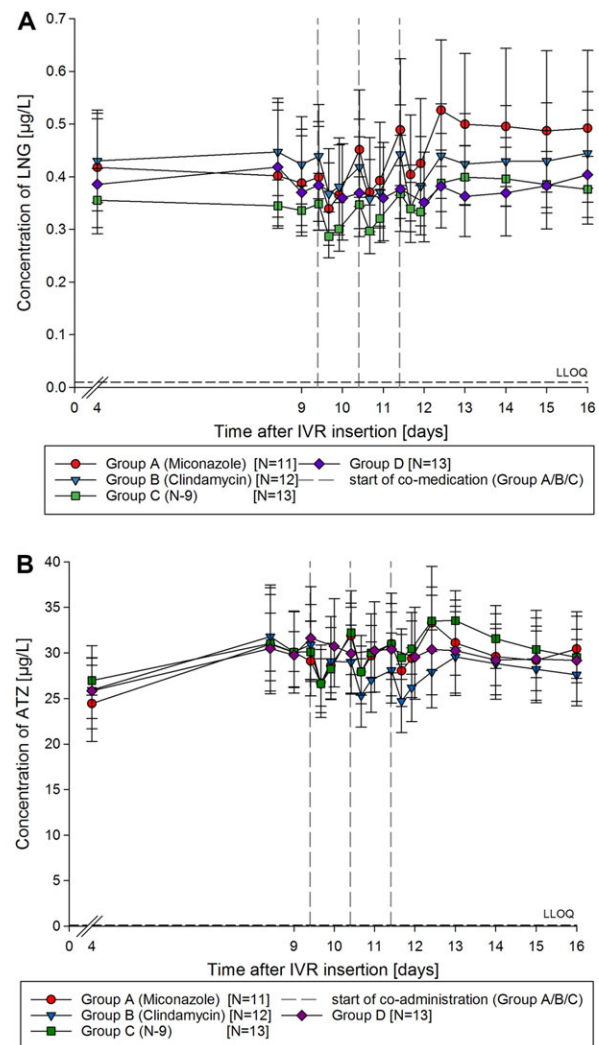
### Pharmacokinetics

At the first sample point (ie, 96 hours after insertion of the IVR), LNG concentrations seemed to be already at a plateau, whereas ATZ concentration increased further until day 10. Plasma concentrations of ATZ and LNG were measured up to 16 days (384 hours) after the start of the insertion of the ring. Overall, the concentrations of LNG and ATZ in the groups were comparable during the entire wearing period of the IVR (Figure 1). In particular, the concentrations on day 9 indicate similar exposure in the groups before the start of comedication. Furthermore, there seems to be no strong effect on PK during or after the 3 days of comedication. The concentration–time profiles for group D represent reference profiles, because no interaction was tested during the first 16 days.

During the use of tampons, the plasma concentration–time profiles were slightly more variable; however, no huge impact was seen for either drug, in particular, considering the variability as indicated by the error bars in the figures (Figure 2). After removal of the last tampon, the plasma concentrations of ATZ and LNG were relatively stable during the following week. After removal of the IVR, the concentrations declined, and plasma concentrations could be measured up to 6 days thereafter.

The  $C_{av}$  was calculated by using the corresponding partial AUCs (Table 3). The  $C_{av}$  (reference) reflects the concentration prior to the start of the DDI evaluation. The corresponding geometric mean values for ATZ of all 4 groups were very similar (30.2–30.8  $\mu\text{g/L}$ ), whereas for LNG the range was slightly wider (0.341–0.433  $\mu\text{g/L}$ ) and overall more variable.

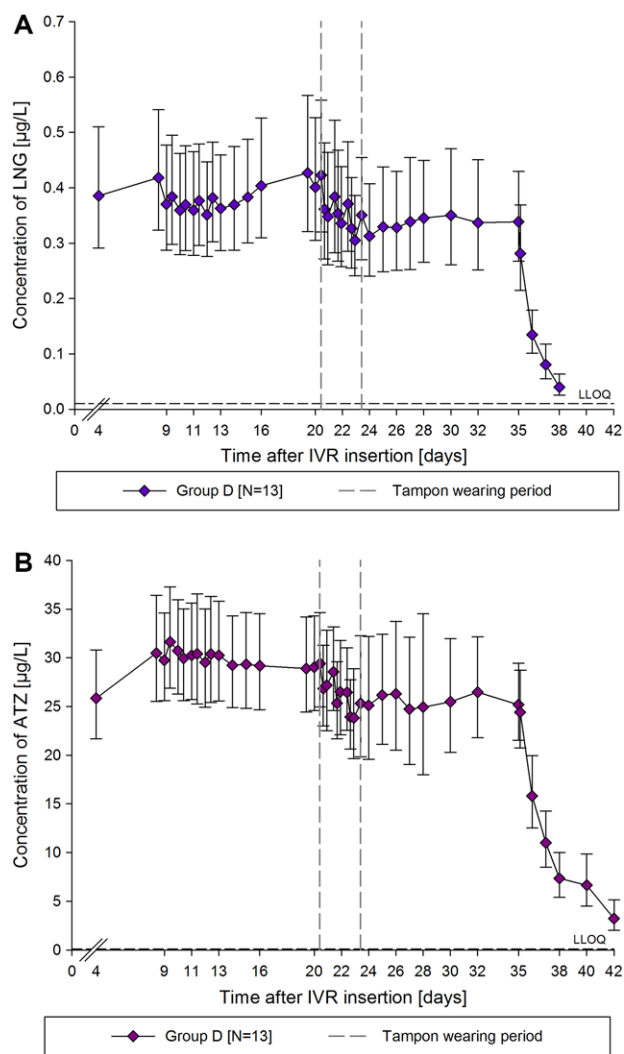
Group D served as a reference because no tampons were inserted during the “treatment period” of the other groups, and consequently the exposure was unchanged. The  $C_{av}$  (reference T) reflects the concentration prior to insertion of the first tampon. The corresponding geometric means for ATZ and LNG



**Figure 1.** Geometric mean (with geometric SEM) for plasma concentration of ATZ (top) and LNG (bottom) before, during, and after coadministration of miconazole, clindamycin, or nonoxynol-9 or the concomitant use of tampons. Note: concentration–time data from group D within the wearing period of 16 days are included in Figure 1, whereas the complete concentration–time profiles for group D are provided in Figure 2.

were in the same range as the  $C_{av}$  (reference). During the use of tampons, the  $C_{av}$  decreased slightly for both drugs.

The intended wearing period of the IVR is 28 days; however, it may happen that the IVR is worn longer, for



**Figure 2.** Geometric mean (with geometric SEM) plasma concentration for ATZ (top) and LNG (bottom) before, during, and after the concomitant use of tampons.

example, 1 week longer. Therefore, exposure on day 35 compared with day 28 is of interest. The  $C_{28d}$  and  $C_{35d}$  in group D were similar, with geometric mean concentrations of 24.9 and 25.3  $\mu\text{g/L}$  for ATZ, respectively. The corresponding LNG plasma concentrations on day 28 and day 35 were 0.338 and 0.334  $\mu\text{g/L}$ , respectively (Figure 2).

The geometric mean  $t_{1/2}$  after IVR removal was 40.4 and 25.6 hours for ATZ and LNG, respectively.

A similar pattern across all treatments was observed in the mean concentration of SHBG-versus-time curves. Baseline values of SHBG in groups A, B, C, and D were 69, 76, 55, and 65 nmol/L, respectively. Geometric mean values of SHBG on day 9 in groups A, B, C, and D were 53, 61, 45, and 52 nmol/L, respectively. Furthermore, the geometric mean of SHBG on day 20 in group D prior to the start of tampon use was 52 nmol/L. The follow-up values (about 14 days after

IVR removal) of SHBG in groups A, B, C, and D were 70, 71, 59, and 55 nmol/L, respectively.

In summary, mean serum concentration of SHBG decreased after insertion of the IVR, and after removal of the IVR, the SHBG concentration returned to the baseline level. Overall, at the start of the DDI investigation, SHBG levels were comparable between the groups.

#### Statistical Evaluation

Point estimates of ratios  $C_{av}$  (DDI 72 hours)/ $C_{av}$  (reference) and  $C_{av}$  (DDI 158 hours)/ $C_{av}$  (reference) for ATZ and LNG as well as their 90% CIs were calculated for all groups (Table 4). All point estimates and 90% CIs of the ratios were within the bioequivalence range of 0.800 to 1.250. It should be noted that group D served as a reference, because no interaction was investigated during this period.

For the investigation of the tampon's interaction using point estimates of ratios  $C_{av}$  (DDI 72 hours)/ $C_{av}$  (reference T) and  $C_{av}$  (DDI 158 hours)/ $C_{av}$  (reference T) for ATZ and LNG as well as their 90% CIs are relevant (Table 4). For ATZ and for LNG (with 1 exception for LNG for the ratio  $C_{av}$  [DDI 158 hours]/ $C_{av}$  [reference T]), all point estimates and 90% CIs were within the bioequivalence range of 0.800 to 1.250.

#### Safety

The safety analysis set consisted of 52 subjects. Of these, 43 (82.7%) had at least 1 treatment-emergent adverse event (TEAE). Most TEAEs were of mild intensity. Thirteen subjects (25.0%) had at least 1 TEAE with moderate intensity. No severe TEAEs were reported. The most frequently occurring TEAEs ( $\geq 10\%$  of the total population) were headache, in 10 subjects (19.2%); ovarian cyst (defined as follicle-like structure with an average diameter of  $\geq 30$  mm), in 10 subjects (19.2%); and nasopharyngitis, in 6 subjects (11.5%).

Study drug-related TEAEs related to IVRs containing ATZ and LNG occurred in 31 subjects (57.7%). The most frequently ( $>10\%$ ) occurring drug-related TEAEs were headache, in 9 subjects (17.3%), and ovarian cyst, in 10 subjects (11.5%). AEs considered related to comedication (or co-usage of tampons) were reported in 9 of 52 subjects (17.3%). None of these TEAEs were reported as being related to the same comedication in more than 1 subject. There were no deaths or serious AEs in this study. Two subjects discontinued the study prematurely because of AEs (both because of bacterial vaginosis).

There were no notable changes from baseline in vital signs or laboratory parameters during treatment.

#### Discussion

The primary objective of this study was to investigate the effect of vaginally administered comedications or

**Table 3.** Summary Statistics of  $C_{av}$  of ATZ and LNG in All Treatment Groups (Geometric Mean [ $\mu\text{g/L}$ ] and Coefficient of Variation [%])

Interval	$C_{av}$					
	Reference 202–226 Hours	DDI 72 Hours 226–298 Hours	DDI 158 Hours 226–384 Hours	Reference T 466–490 Hours	DDI 72 Hours T 490–562 Hours	DDI 158 Hours T 490–648 Hours
ATZ						
Group A (miconazole), n = 11	30.2 (27.7)	29.7 (25.7)	30.1 (23.5)			
Group B (clindamycin), n = 12	30.8 (27.7)	27.5 (27.5)	28.2 (26.5)			
Group C (nonoxynol-9), n = 13	30.4 (17.1)	30.0 (18.3)	30.9 (19.2)			
Group D (tampon), n = 13 <sup>a</sup>	30.8 (29.8)	30.5 (31.1)	30.0 (30.9)	29.4 (30.6)	26.5 (29.1)	26.8 (31.8)
LNG						
Group A (miconazole), n = 11	0.395 (46.7)	0.418 (45.5)	0.461 (47.6)			
Group B (clindamycin), n = 12	0.433 (39.7)	0.395 (39.8)	0.415 (41.8)			
Group C (nonoxynol-9), n = 13	0.341 (28.0)	0.335 (27.9)	0.366 (28.3)			
Group D (tampon), n = 13 <sup>a</sup>	0.380 (49.9)	0.368 (48.1)	0.373 (47.5)	0.415 (55.1)	0.349 (49.7)	0.335 (49.1)

T, tampon.

<sup>a</sup>n = 12 for 490–562 hours and n = 11 for 490–648 hours.

tampons on the PK of ATZ and LNG administered as an IVR releasing ATZ and LNG. As the vaginal route of administration avoids hepatic first-pass metabolism and potential food effects, such interactions are considered unlikely; however, local interactions affecting the absorption of drugs in the vagina may occur. The latter also needs to be considered in particular for tampon use. Before this study, no PK data were available on these topics for the ATZ/LNG IVR.

In the present study, the PK results for ATZ and LNG without comedication or tampon use are in line with the expectations based on the previous study investigating similar IVRs with 3 different ATZ/LNG ratios.<sup>19</sup> In the previous study, the geometric mean  $C_{av}$  plasma concentrations of ATZ and LNG were 27.4 and 0.36  $\mu\text{g/L}$  for IVRs delivering 1000  $\mu\text{g}$  ATZ/day or 40  $\mu\text{g}$  LNG/day, respectively. In the present study, the geometric mean plasma concentrations of the ATZ (1050  $\mu\text{g/day}$ ) and LNG (40  $\mu\text{g/day}$ ) were about 30 and 0.39  $\mu\text{g/L}$ , respectively (Table 3), and thereby well matched with the previous results.

This phase 1 study investigated the PK effect of a vaginally administered antimycotic (miconazole — group A), antibiotic (clindamycin — group B), and spermicide (nonoxynol-9 — group C) and the concomitant use of tampons (group D) on ATZ and LNG administered as an IVR releasing ATZ and LNG. A comparison of PK results across groups within this study could be done, because (1) the number of subjects per treatment group included in the analysis was similar

(n = 11–13); (2) the mean body weight across dose groups was very similar, ranging from 63.9 to 68.4 kg; and (3) the concentrations of ATZ and LNG prior to the start of the DDI investigation were comparable across all groups.

The investigation of potential DDIs with intravaginally administered drugs started 9 days after the IVR insertion, at which time stable conditions had been reached for ATZ, LNG, and SHBG. Following intravaginal administration via a continuously releasing IVR, it was expected that the drug concentrations would not show the fluctuations typically seen after daily oral administration. Therefore, only 3 blood samples were taken as a reference within 24 hours, confirming that the ATZ and LNG concentrations were stable over the day. The concentration–time data from these samples have been used to calculate the average concentration of the reference ( $C_{av}$  [reference]). Within all 4 treatment groups, similar exposures were observed for ATZ, with geometric mean values for  $C_{av}$  (reference) in the range of 30.2–30.8  $\mu\text{g/L}$ , and for LNG, with geometric mean values of 0.341–0.433  $\mu\text{g/L}$ .

In this context, SHBG concentrations should be considered because of the interaction of LNG with SHBG and because SHBG is a major binding protein for LNG.<sup>20,21</sup> The geometric mean SHBG concentrations for this period were comparable in all treatment groups as well as on day 20 (group D). Therefore, an overall similar unbound LNG concentration in all groups prior to the start of the comedication or usage

**Table 4.** Point Estimates and 90% Confidence Intervals for Comparison of  $C_{av}$  of ATZ and LNG at Different Intervals

Group	Ratio of Parameters	n	ATZ		LNG	
			Point Estimate (LS Mean)	90%CI	Point Estimate (LS Mean)	90%CI
A	$C_{av}$ (DDI 72 hours)/ $C_{av}$ (reference)	11	0.986	0.957–1.015	1.060	1.012–1.109
B		12	0.895	0.871–0.921	0.911	0.872–0.952
C		13	0.988	0.961–1.015	0.981	0.940–1.023
D <sup>a</sup>		13	0.990	0.963–1.017	0.971	0.930–1.013
A	$C_{av}$ (DDI 158 hours)/ $C_{av}$ (reference)	11	0.996	0.967–1.025	1.168	1.116–1.223
B		12	0.915	0.890–0.941	0.958	0.917–1.001
C		13	1.015	0.988–1.043	1.072	1.027–1.118
D <sup>a</sup>		13	0.973	0.947–1.000	0.984	0.943–1.026
D	$C_{av}$ (DDI 72 hours T)/ $C_{av}$ (reference T)	11 <sup>b</sup>	0.899	0.860–0.940	0.882	0.806–0.966
D	$C_{av}$ (DDI 158 hours T) / $C_{av}$ (reference T)	11 <sup>b</sup>	0.887	0.849–0.927	0.873	0.797–0.955

<sup>a</sup>For this evaluation, D is a reference for the other groups, as no tampons are used.

<sup>b</sup>All participants who provided PK data on both periods were included in the statistical analyses.

of tampons was expected, and the DDI investigation using the measured total concentration of LNG was justified.

The length of 3 days for antimycotic as well as antibiotic treatment was chosen because it reflects real-life clinical practice. Furthermore, from a PK point of view, repeated dosing with the “to-be-tested” interacting drug is favored compared with single dosing. Consequently, a similar dosing regimen was also implemented for the spermicide and tampon investigations. It should be noted that different dosage forms for the comedication were used. Miconazole nitrate was administered as a vaginal capsule. Clindamycin was administered as vaginal cream, whereas nonoxynol-9 was administered as a vaginal suppository. None of these different dosage forms showed an interaction with the IVR.

Based on the statistical evaluation, no DDI was observed for miconazole, clindamycin, or nonoxynol-9. All 90% CIs of the analysis of variance (ANOVA) performed for ATZ and LNG were within the range of 0.800 to 1.2500.

A further primary objective of the study was to investigate the PK effect of concomitant use of tampons during the use of an IVR releasing ATZ and LNG. For this purpose, the tampons were used in group D on days 20 to 23. No parallel reference group was available for this period; however, the continuous, stable concentration–time profiles of ATZ and LNG for this period could be seen in a previous study.<sup>14</sup> Interestingly, the concentrations of ATZ and LNG appeared to be slightly decreased during the use of tampons. Overall this happened in a similar manner for both drugs, indicating that the use of tampons might affect absorption of ATZ and LNG. Considering the entire tampon-wearing period, the plasma concentrations of ATZ and LNG decreased by about 10.1% and 11.7%, respectively (see Table 4). As for the ANOVA above, the  $C_{av}$  (here  $C_{av}$  [reference T]) prior to the start of tampon

use served as a reference, resulting in point estimates for the ratios within the bioequivalence range for ATZ and LNG (see Table 4). Furthermore, the entire 90%CI for ATZ was in the bioequivalence range, whereas the lower 90%CI limit for LNG was slightly below 0.800 (0.797). The slight decrease in LNG concentrations is not considered meaningful and might be allocated to a chance finding because of the overall variability of LNG in plasma. In summary, there was no relevant PK effect on ATZ and LNG delivered from IVRs because of concomitant use of tampons.

Similarly, no effect of tampon co-usage was reported on systemic exposure of etonogestrel (ENG) and ethinylestradiol (EE) delivered from the combined contraceptive intravaginal ring (NuvaRing).<sup>22</sup> In a crossover design, 14 healthy women were randomized to use both NuvaRing and tampons or NuvaRing alone for 1 cycle of ring use (3 weeks followed by a 1-week ring-free period, in total 84 days without follow-up visit). The first tampon was inserted on the morning of day 8 of the interaction cycle, and 4 tampons a day were to be used for 3 consecutive days. Blood samples were collected on cycle days 9, 10, 11, 13, 15, 18, and 21, and the systemic exposure of ENG and EE was considered a short-term (24-hour) PK interaction and for interaction during cotreatment (72 hours). As there were no statistically significant effects of tampon co-usage on the systemic exposure of ENG and EE, it was concluded that the concomitant use of tampons with NuvaRing does not interfere with either the release of hormones from the ring or vaginal absorption of the delivered drugs.

In summary, the results of the NuvaRing and our study show that interactions between tampon co-usage and delivery of these drugs from intravaginal rings are unlikely. Nevertheless, this statement should not be generalized for other drugs, considering the huge differences in the physicochemical properties of the investigated drugs.



As there is a relatively high incidence of bacterial vaginosis in the general female population,<sup>23</sup> an investigation of potential DDIs of intravaginally administered antimicrobics on the exposure of drugs delivered by an IVR is of interest. In our study, 3 × 400 mg of miconazole nitrate as a vaginal capsule was administered at bedtime according to the instructions for use. No significant DDI was observed in any of the investigated periods.

Although the investigated IVR should be a contraceptive, the effects of a spermicide on ATZ and LNG release and absorption were studied using a vaginal gel spermicide containing 4% nonoxynol-9. Administration was on 3 consecutive evenings. Despite the repeated dosing of the spermicide, no relevant DDI for ATZ and LNG was observed.

Similar DDI studies were performed for NuvaRing.<sup>22,24,25</sup> However, there are major differences with our study. The menstrual cycles of the participants were synchronized, and in the following 2 cycles NuvaRing was administered alone or together with the to-be-investigated agent (spermicide/antimicrobial/tampon). Therefore, the total duration of each study without follow-up visit was at least 84 days. The number of subjects (12 or 14) per group is comparable to our study, whereas the study duration was much shorter in our study, which was also favorable for the study participants. Furthermore, we followed the recommendation for many vaginally administered drugs, which suggests using medication in the evening prior to bedtime. Consequently, the design of the study differed from most other phase 1 studies, because the subjects stayed overnight during the DDI investigations, whereas their normal daily life was almost unaffected.

In addition, an extended wearing duration of 35 days without ring change was investigated, indicating overall extension of the expected exposure of ATZ and LNG (Figure 2). Therefore, at least a 5- instead of a 4-week wearing period of the IVR seems to be feasible. Following removal of the IVR, blood samples were taken to investigate the elimination of the drugs. The geometric mean  $t_{1/2}$  was 40.4 and 25.6 hours for ATZ and LNG, respectively, and overall was in line with the product information data.<sup>10,26</sup>

Overall, ATZ and LNG delivered by IVR were well tolerated, as expected. ATZ and LNG are marketed drugs with well-established pharmacological and safety profiles.

## Conclusion

The results of the present study demonstrate the absence of pharmacokinetic interactions of the tested antibiotic, antimicrobial, and spermicide as well as tampons

on ATZ and LNG released from IVR. Therefore, no restrictions on the use of this IVR delivering ATZ and LNG are needed.

## Acknowledgments

The authors thank Katrin Kettelhake and Antonia Kohnke for contributing to the analyses described in this article and thank April Fulcher for her support improving the language in the article. All listed authors meet the criteria for authorship and were involved in the analysis and interpretation of the data.

## Declaration of Conflicting Interests

All authors are employees of Bayer AG. André Müller is a contracted consultant (gynecologist to CRS Berlin, Germany), who received funding for this study from Bayer AG.

## Funding

This study was funded by Bayer AG.

## References

1. Crosignani P, Olive D, Bergqvist A, et al. Advances in the management of endometriosis: an update for clinicians. *Hum Reprod Update* 2006;12:179–189.
2. Dunselman GA, Vermeulen N, Becker C, et al. ESHRE guideline: management of women with endometriosis. *Hum Reprod* 2014;29(3):400–412.
3. Sinaï N, Plumb K, Cotton L, et al. Differences in characteristics among 1,000 women with endometriosis based on extent of disease. *Fertil Steril*. 2008;89(3):538–545.
4. Culley L, Law C, Hudson N, et al. The social and psychological impact of endometriosis on women's lives: a critical narrative review. *Hum Reprod Update* 2013;19(6):625–639.
5. Bulun SE, Zeitoun KM, Takayama K, Sasano H. Estrogen biosynthesis in endometriosis: molecular basis and clinical relevance. *J Mol Endocrinol*. 2000;25:35–42.
6. Simpson ER, Clyne, C, Rubin G, et al. Aromatase -a brief overview. *Annu Rev Physiol*. 2002;64:93–127.
7. Noble LS, Simpson ER, Johns A, et al. Aromatase expression in endometriosis. *J Clin Endocrinol Metab*. 1996;81(1):174–179.
8. Kitawaki J, Kusuki I, Koshiha H, et al. Detection of aromatase cytochrome P-450 in endometrial biopsy specimens as a diagnostic test for endometriosis. *Fertil Steril*. 1999;72(6):1100–1106.
9. Velasco I, Rueda J, Acíen P. Aromatase expression in endometriotic tissues and cell cultures of patients with endometriosis. *Mol Hum Reprod*. 2006;12(6):377–381.
10. Arimidex® Summary of product characteristics. <http://www.azpicentral.com/arimidex/arimidex.pdf>. Accessed February 23, 2017.
11. Ailawadi R, Jobanputra S, Kataria M, et al. Treatment of endometriosis and chronic pelvic pain with letrozole and norethindrone acetate: a pilot study. *Fertil Steril*. 2004;81(2):290–296.
12. Amsterdam LL, Gentry W, Jobanputra S, et al. Anastrozole and oral contraceptives: a novel treatment for endometriosis. *Fertil Steril*. 2005;84:300–304.
13. Abushahin F, Goldman KN, Barbieri E, et al. Aromatase inhibition for refractory endometriosis-related chronic pelvic pain. *Fertil Steril*. 2011;96:939–942.

14. Schultze-Mosgau MH, Waellnitz K, Nave R, et al. Pharmacokinetics, pharmacodynamics, safety and tolerability of an intravaginal ring releasing anastrozole and levonorgestrel in healthy premenopausal women: a Phase I randomized controlled trial. *Hum Reprod.* 2016;31(8):1713–1722.
15. Reinecke I, Schultze-Mosgau MH, Nave R, et al. Model-based dose selection for intravaginal ring formulations releasing anastrozole and levonorgestrel intended for the treatment of endometriosis symptoms. *J Clin Pharmacol.* 2017;57(5):640–651.
16. Janssen-Cilag Gyno-Daktarin<sup>®</sup>, Summary of product characteristics. <https://www.medicines.org.uk/emc/medicine/894>. Accessed March 7, 2017.
17. Pharmacia Pfizer Pharma Sobelin<sup>®</sup> Vaginal Cream, Summary of product characteristics. <https://www.medicines.org.uk/emc/medicine/7594>. Accessed March 7, 2017.
18. Package insert of Patentex oval<sup>®</sup>. Merz Consumer Care GmbH. [http://www.patentex.com/de/download\\_area/downloads/PTX\\_oval\\_GA\\_Internet.pdf](http://www.patentex.com/de/download_area/downloads/PTX_oval_GA_Internet.pdf). Accessed March 7, 2017.
19. Package insert of o.b. [http://www.ob.de/sites/ob\\_de/files/ob\\_verpackungsbeilage\\_de.pdf](http://www.ob.de/sites/ob_de/files/ob_verpackungsbeilage_de.pdf). Accessed March 7, 2017.
20. Kuhnz W, Schütt B, Woloszczak R, et al. Influence of changes in the concentration of sex hormone-binding globulin in human serum on the protein binding of the contraceptive steroids levonorgestrel, 3-keto-desogestrel and gestodene. *J Steroid Biochem Mol Biol.* 1994;48(5–6):573–580.
21. Dowsett M, Attree S, Virdee S, Jeffcoate S. Oestrogen-related changes in sex hormone binding globulin levels during normal and gonadotrophin-stimulated menstrual cycles. *Clin Endocrinol.* 1985;23:303–312.
22. Verhoeven CHJ, Dieben TOM. The combined contraceptive vaginal ring, NuvaRing, and tampon co-usage. *Contraception.* 2004;69:197–199.
23. Sobel JD. Vaginitis. *N Engl J Med.* 1997;337:1896–1903.
24. Verhoeven CHJ, van den Heuvel MW, Mulders TMT, et al. The contraceptive vaginal ring, NuvaRing, and antimycotic co-medication. *Contraception.* 2004;69:129–132.
25. Haring T, Mulders TMT. The combined contraceptive ring NuvaRing and spermicide co-medication. *Contraception.* 2003;67:271–272.
26. Bayer Norgeston<sup>®</sup> Summary of product characteristics. <http://www.medicines.org.uk/emc/medicine/1834>, Accessed February 27, 2017.