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Effect of Cladribine Tablets on the Pharmacokinetics of a Combined Oral Contraceptive in Pre-Menopausal Women With Relapsing Multiple Sclerosis

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ABSTRACT

This study assessed the effect of cladribine tablets (CladT) on the pharmacokinetics (PK) of a combined oral contraceptive (COC) in pre-menopausal women with relapsing multiple sclerosis. It was a randomized, double-blind, two-period, two-sequence crossover study to assess steady-state plasma PK (area under the concentration–time curve and peak concentration) of COC (ethinylestradiol [EE] 30 μg and levonorgestrel [LNG] 150 μg) when co-administered with CladT or placebo. Participants received 2 weeks of active CladT treatment per course (Weeks 1 and 5 per year) to have a cumulative dose of 3.5 mg/kg over 2 years as per label. Of the 24 randomized participants, 23 completed the study. The results showed that the concentration–time profiles as well as PK parameters of EE and LNG in the plasma were similar when co-administered with CladT or placebo. Analysis of variance confirmed the bioequivalence of EE and LNG in COC when co-administered with either CladT or placebo. All participants were adequately exposed to cladribine. Repeat-dose administration of CladT had no apparent effect on serum luteinizing hormone, follicle-stimulating hormone, progesterone, or sex hormone-binding globulin concentrations during concomitant treatment with COC. Co-administration with COC did not change the known safety and tolerability profile of CladT and did not alter the PK of EE or LNG in a COC during the study. Therefore, the concomitant use of CladT is not expected to decrease the efficacy of COCs containing EE and LNG.

Trial Registration: EudraCT Number: 2018-001015-70.

1 | Introduction

Multiple sclerosis (MS) is the most common disabling neurological disease in young adults, affecting approximately 2.9 million people worldwide [1, 2]. It is a disease with approximately three times more prevalence in women than in men [2]. The typical age of MS onset is during the childbearing years [2]; however, it does not significantly impair fertility [3, 4]. Female patients using disease-modifying therapies (DMTs) to manage MS generally require effective contraception to avoid the risk of adverse foetal development effects [5, 6].

Summary

- What is the current knowledge on the topic?
  - Combined oral contraceptives (COCs) are metabolized by CYP3A4 as well as other drug-metabolizing enzymes. The concomitant use of enzyme inducers may reduce their efficacy. Previous in vitro studies have demonstrated that cladribine has no clinically meaningful inductive effect on CYP3A4 enzymes; however, the effect of cladribine tablets on the pharmacokinetics of COCs has not been evaluated.
- What question did this study address?
  - This study investigated the potential effects of cladribine tablets on the pharmacokinetics of a commonly prescribed COC containing ethinylestradiol and levonorgestrel in premenopausal women with relapsing multiple sclerosis.
- What does this study add to our knowledge?
  - Co-administration with cladribine tablets did not alter the pharmacokinetics of ethinylestradiol or levonorgestrel in a COC. This was confirmed by analysis of variance for bioequivalence between COC co-administered with cladribine tablets and COC co-administered with placebo. Therefore, the concomitant use of cladribine tablets is not expected to decrease the efficacy of COCs containing ethinylestradiol and levonorgestrel. Co-administration with COC containing ethinylestradiol and levonorgestrel did not change the known safety and tolerability profile of cladribine tablets during the study.
- How might this change clinical pharmacology or translational science?
  - Co-administration of CladT did not alter the PK of ethinylestradiol or levonorgestrel in a COC. Therefore, the concomitant use of cladribine tablets is not expected to decrease the efficacy of ethinylestradiol/levonorgestrel-based COC as a means of birth control in women with relapsing multiple sclerosis.

Cladribine tablets, a short-course oral therapy with established safety and efficacy profiles, are indicated for the treatment of adults with highly active relapsing MS (RMS) as defined by clinical or imaging features [7]. The recommended cumulative dose is 3.5 mg/kg body weight over 2 years, as one treatment course of 1.75 mg/kg/year. Each treatment course consists of two treatment weeks, one at the beginning of the first month and one at the beginning of the second month of the respective treatment year. Each treatment week consists of 4 or 5 days of treatment at 10 mg or 20 mg as a single daily dose, depending on body weight [7]. Since its first approval in 2017 in The European Union, an estimated 101,132 patients with MS have been treated with CladT, with a cumulative exposure of 251,900 years (until end-June 2024; the healthcare business of Merck KGaA, Darmstadt, Germany, data on file).

Cladribine is a prodrug that is preferentially phosphorylated in lymphocytes to its metabolite 2-chlorodeoxyadenosine triphosphate. Through its active metabolite, cladribine induces reversible, selective reduction in lymphocyte count, which contributes to the management of autoimmune processes involved in MS [8, 9]. The pharmacokinetics (PK) of cladribine have been extensively investigated in patients with MS previously [8]. The bioavailability of oral cladribine is approximately 40% when administered in a fasted state, and its absorption remains unaffected by food intake [7, 8]. The mean apparent volume of distribution of cladribine ranges from 480 to 490 L, and the estimated median apparent renal and non-renal clearance rates are 22.2 and 23.4 L/h, respectively [10]. It has a half-life of approximately 1 day with no drug accumulation upon once-a-day dosing [7, 8, 11]. Cladribine is not a substrate of cytochrome (CYP) P450 enzymes and has no clinically meaningful inductive effect on CYP1A2, CYP2B6, and CYP3A4 enzymes based on in vitro studies [7, 8]. Cladribine has been shown to exert teratogenic effects in preclinical studies, consistent with its pharmacologic mechanisms of action [7]. Therefore, women of childbearing age are recommended to use effective contraception during treatment with CladT and for at least 6 months after the last dose in each treatment course to avoid pregnancy [7].

Combined oral contraceptives (COCs) are widely used by women of reproductive age and are highly effective in preventing pregnancy when used appropriately. However, drug interactions with concomitant use of therapies can adversely affect the enzymes involved in the metabolism of progestins and estrogens, thereby affecting the efficacy and safety of COCs [12]. COCs contain a combination of two synthetic steroid hormones such as estrogen and progestin, which are metabolized by inducible drug-metabolizing enzymes, including CYP3A4. The effect of CladT on the PK of COCs had not been evaluated. Given its teratogenic potential, it is imperative to assess the impact of CladT treatment on the PK and PD of COC upon co-administration [7].

The primary objective of this study was to investigate the potential effect of repeat-dose CladT (10 mg/tablet) administration on the PK of a commonly used monophasic COC (microgynon tablets [Bayer AG]) containing ethinylestradiol (EE; 30 μg/tablet) and levonorgestrel (LNG; 150 μg/tablet), by assessing its components EE and LNG in pre-menopausal women with RMS. The secondary objectives were to examine cladribine exposure and to assess the safety and tolerability of the co-administration of EE/LNG COC and CladT. The PD effects of the co-administration of EE/LNG COC and CladT on serum hormone concentrations during ovulatory cycles were also assessed.

2 | Methods

This randomized, double-blind, two-period, two-sequence crossover study with a 1-month run-in period was conducted in pre-menopausal women with active RMS (EudraCT Number: 2018-001015-70). This study was conducted at six sites across Germany and Poland between January 2019 and March 2022.

The study design (Figure 1) comprised a ≤21-day screening period and an EE/LNG COC run-in period followed by two sequential treatment periods (CladT–placebo or placebo–CladT), each consisting of two 28-day study periods. The participants received EE/LNG COC for 21 days during the run-in period

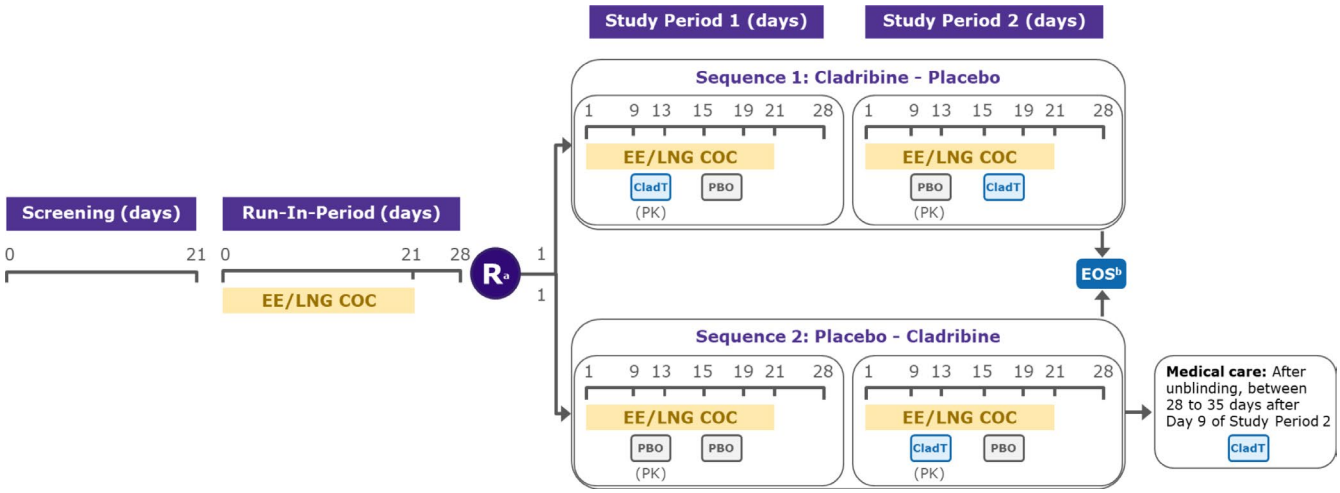

FIGURE 1 | Study design. [a]Randomisation within 10 days after Day 21; [b]End of study 4–7 days after Day 21 of study period 2. CladT, cladribine tablets; COC, combined oral contraceptive; EE, ethinylestradiol; LNG, levonorgestrel; PBO, placebo.

and both treatment periods and were off treatment for 7 days. In study periods 1 and 2, EE/LNG COC was co-administered with randomized treatment sequences of a 5-day once-daily CladT treatment (Days 9–13 of study period 1) followed by a 5-day once-daily matched placebo treatment (Days 9–13 of study period 2) or vice versa. Additionally, the participants received matched placebo treatment (Days 15–19 of study period 1) to ensure blinding of the CladT–placebo sequence among those who received their second weekly dose of CladT (Days 15–19 of study period 2). In turn, participants in the placebo–CladT sequence received the second weekly dose of CladT after the end-of-study visit as part of standard medical care. An end-of-study examination was performed within 4–7 days after the last visit during study period 2. Of note, only Days 9–13 CladT and matched placebo treatments cover the primary and secondary endpoints while Days 15–19 CladT and matched placebo treatments are factored in to allow for a timely initiation of the second 5-day CladT treatment for participants assigned to the CladT–placebo treatment sequence in a blinded fashion.

Due to the sustained lymphopenic effect and safety profile associated with CladT, no clinical pharmacology studies were conducted in healthy volunteers. Eligible participants were premenopausal women with RMS (as defined by either the 2001 McDonald or Poser criteria) [13, 14] aged 18–45 years, and having a body mass index (BMI) of 18–30 kg/m^2 at the screening visit. As the CladT dosing is based on body weight, participants with a very low body weight (i.e., < 50 kg) would receive only a 4-day treatment per treatment week instead of a 5-day treatment. Therefore, only participants with a body weight of ≥ 50 kg (up to 100 kg) were eligible for participation to allow for a standardized 5-day repeat-dose CladT treatment. All participants underwent a negative serum pregnancy test at the screening visit. Patients with moderate-to-severe renal (creatinine clearance < 60 mL/min) or hepatic (Child Pugh B and C) impairment were excluded from the study. Additionally, patients who used potent or moderate inducers/inhibitors of CYP3A4 or other CYP450 enzymes (e.g., CYP2C9), as well as conjugation enzymes (e.g., UGT1A1 and SULT1E1), or inducers of the drug transporters P-glycoprotein and breast cancer resistance protein within the past 4 weeks prior to Day 1 of the study were also excluded.

To assess the PK of EE and LNG, blood samples were obtained on Days 7, 14, and 21 of the run-in period, in the morning on Day 13 (pre-dose of EE/LNG COC administration), at pre-dose, 0.5, 1, 2, 4, 6, 8, and 12 h post-dose on Day 14, and at pre-dose on Day 15 (24 h post-dose on Day 14) of study periods 1 and 2. Of note, the serial PK assessment (0–24 h post-dose) of EE and LNG on Day 14 was designed to be on the fifth day of co-administration of CladT and the COC, thereby enabling PK evaluation at the point of maximal potential interaction between the co-administered treatments. Similarly, to assess the PK parameters of cladribine (C_{max} and t_{max}), blood samples were obtained in the morning on Day 9 (pre-dose of CladT/placebo administration) and at pre-dose and 0.25, 0.5, 1.0, 1.5, and 2.0 h post-dose on Days 10–13 of CladT/placebo administration. EE, LNG, and cladribine, along with their respective labeled internal standard, were isolated through liquid extraction, separated by reversed-phase chromatography, and detected via electrospray ionization in positive mode (Please refer to the Appendix S1 for further details regarding the bioanalytical methods). The PK analysis set included all participants who completed both study periods and had available data for primary endpoints.

The primary PK parameters of EE and LNG were area under the concentration–time curve from zero to tau at steady state ($AUC_{\tau,ss}$) and maximum plasma concentration at steady state ($C_{max,ss}$), both of which were derived from plasma concentration–time curves. The secondary PK parameters of EE and LNG were the average plasma concentration at steady state ($C_{av,ss}$), minimum observed concentration at steady state ($C_{min,ss}$), measured concentration at the end of a dosing interval (C_{trough}), peak-to-trough fluctuation over one complete dosing interval (PTF%), and time to reach the observed maximum concentration at steady state ($t_{max,ss}$).

Statistical analysis of PK parameters was performed using an analysis of variance (ANOVA) model that included treatment, period, sequence, and subject within sequence as fixed effects. The PK parameters were natural log-transformed before analysis, and the results were then transformed back to the original scale. If 90% confidence intervals (CIs) for the test/reference ratios of the primary PK endpoints of EE and LNG—$AUC_{\tau,ss}$

and $C_{max,ss}$—were entirely within the standard bioequivalence acceptance range (0.8–1.25), the absence of a drug–drug interaction effect (no effect) was concluded for each comparison (Test [CladT + EE/LNG COC] versus Reference [CladT + placebo]). Summary statistics were obtained for all secondary PK parameters of EE, LNG, and cladribine in the plasma by time point.

The intra-subject coefficient of variations (CVs) of EE and LNG were considered for the sample size calculation (EE: AUC_{ss} 13%, and $C_{max,ss}$ 14%; LNG: AUC_{ss} 16%, and $C_{max,ss}$ 17%) [15–17]. Assuming that AUC and C_{max} for the same analyte are highly correlated, it was planned to randomize at least 20–23 participants to obtain 16 evaluable participants who could provide at least 80% power to demonstrate the absence of an interaction.

To perform pharmacodynamic (PD) assessment of serum concentrations of progesterone, luteinising hormone (LH), follicle-stimulating hormone (FSH), and sex hormone-binding globulin (SHBG), blood samples were collected at the mid-cycle (Days 13, 14, and 15) of each CladT/placebo treatment period (i.e., study periods 1 and 2) before the morning dose. The PD analysis set included all participants who completed at least one treatment sequence and had measurements of the PD variable for at least one treatment. The statistical analysis of the PD parameters was performed using ANOVA. Adverse events (AEs) and lymphocyte counts were monitored throughout the study period. The safety analysis set included all participants who received at least one dose of the intervention.

TABLE 1 | Baseline demographics and disease characteristics (safety analysis set).

Characteristics	Patients $N=28$
Mean age (±SD), years	34 (±6.3)
Female, *n* (%)	28 (100)
Race, *n* (%)	
White	28 (100)
Mean weight (±SD), kg	61.4 (±10.38)
Mean BMI (±SD), kg/m^2	21.9 (±3.69)

Abbreviations: BMI, body mass index; SD, standard deviation.

3 | Results

Of the 28 enrolled participants, 24 (85.7%) were randomized and 23 (82.1%) completed the study. Five participants discontinued the study (withdrew consent, $n=1$; investigator decision, $n=1$; COVID-19-related study interruptions, $n=2$; and failed inclusion criteria for normal blood cell count on Day 21 of the run-in period, $n=1$). All enrolled patients received at least one dose of the study intervention and were included in the safety analysis set ($N=28$). In addition to those who discontinued, 2 participants were excluded from the PK and PD analysis sets due to the intake of prohibited concomitant medications, and 1 participant was excluded from the PD analysis set due to a deviation in the sampling during the study period (PK analysis set, $n=21$; PD analysis set, $n=20$). All participants were Caucasian with a mean (±SD) age of 34 (±6.3) years and a mean BMI of 21.9 (±3.69) kg/m^2 (Table 1). Of the 23 participants who completed the study, 22 received CladT and matched placebo as planned and were exposed to 1.75 and 0.875 mg/kg body weight CladT in sequences 1 ($n=12$ participants) and 2 ($n=10$ participants), respectively. Similarly, 22 participants received EE/LNG COC as planned (single daily dose of EE 30 µg and LNG 150 µg in sequences 1 and 2).

The mean plasma concentrations of EE and LNG over a 24-h period after oral co-administration of EE/LNG COC with CladT or with placebo are shown in Figure 2A,B. The concentration–time profiles of EE and LNG in the plasma were comparable when co-administered with CladT or placebo. The mean plasma concentrations of EE and LNG increased rapidly to reach the $C_{max,ss}$ between 1 and 2 h post-dose, irrespective of co-administration with either CladT or placebo (Figure 2A,B).

The primary and secondary PK parameters of EE and LNG in the plasma were similar when co-administered with CladT or placebo (Table 2). The measures of EE and LNG exposure, based on $AUC_{\tau,ss}$ and $C_{max,ss}$, when EE/LNG COC was coadministered with CladT or placebo are presented in Table 3. For EE, the geometric least-squares means for $AUC_{\tau,ss}$ and $C_{max,ss}$

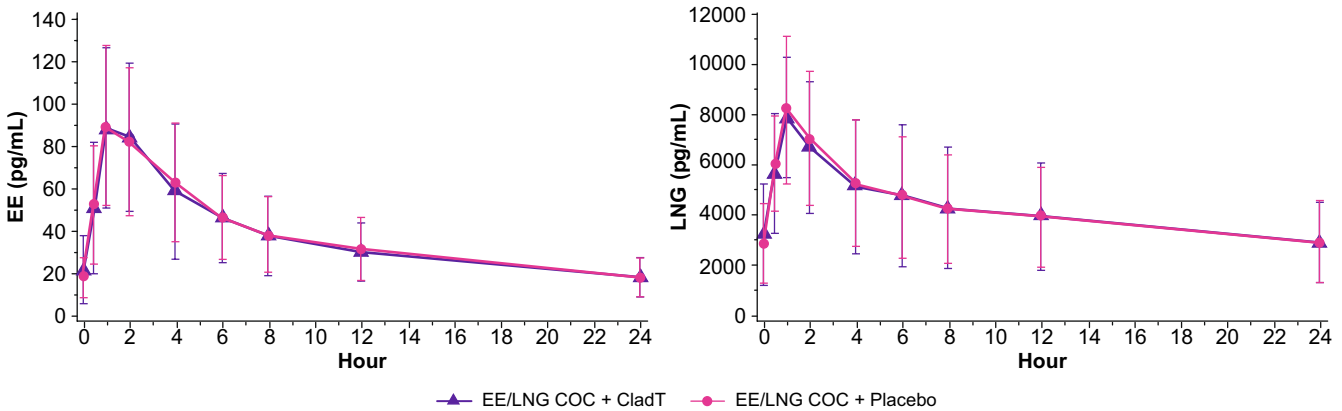

FIGURE 2 | Mean plasma concentrations of (A) EE and (B) LNG when co-administered with CladT or placebo (PK analysis set). Data points (error bars) represent the arithmetic mean (standard deviation); $n=21$.CladT, cladribine tablets; COC, combined oral contraceptive; EE, ethinylestradiol; LNG, levonorgestrel; PK, pharmacokinetics.

TABLE 2 | Summary of primary and secondary PK endpoints (PK analysis set).

	EE/LNG COC with CladT ($n = 21$)		EE/LNG COC with placebo ($n = 21$)	
Analyte/parameter	**GeoMean (95% CI)**	**GeoCV (%)**	**GeoMean (95% CI)**	**GeoCV (%)**
EE				
$AUC_{\tau,ss}$ (h × pg/mL)	789 (620–1000)	56.9	817 (653–1020)	52.6
$C_{max,ss}$ (pg/mL)	88.3 (72.2–108)	46.5	88.1 (72.3–107)	45.7
$C_{av,ss}$ (pg/mL)	32.9 (25.9–41.9)	56.9	34.1 (27.2–42.6)	52.6
$C_{min,ss}$ (pg/mL)	14.8 (11.3–19.5)	66.3	14.7 (11.2–19.3)	65.6
C_{trough} (pg/mL)	15.5 (11.7–20.4)	66.8	15.4 (11.7–20.4)	66.9
PTF% (%)	220 (194–250)	28.0	213 (192–237)	23.2
$t_{max,ss}$ (h), median (range)	1.0 (0–2.0)	—	1.0 (0.4–2.0)	—
LNG				
$AUC_{\tau,ss}$ (h × pg/mL)	91,400 (75,100–111,000)	45.2	92,000 (75,100–113,000)	46.8
$C_{max,ss}$ (pg/mL)	7950 (6840–9240)	33.9	8150 (6990–9500)	34.6
$C_{av,ss}$ (pg/mL)	3810 (3130–4630)	45.2	3830 (3130–4690)	46.8
$C_{min,ss}$ (pg/mL)	2520 (2040–3120)	49.5	2430 (1920–3090)	56.3
C_{trough} (pg/mL)	2580 (2090–3180)	48.5	2580 (2030–3270)	56.0
PTF% (%)	140 (122–159)	29.7	145 (128–166)	29.0
$t_{max,ss}$ (h), median (range)	1.0 (0.4–2.0)	—	1.0 (0.4–2.0)	—

Abbreviations: AUC, area under the plasma concentration–time curve; C_{av}, average plasma concentration; CI, confidence interval; C_{max}, maximum observed concentration; C_{min}, minimum observed concentration; COC, combined oral contraceptive; C_{trough}, measured concentration at the end of a dosing interval at steady state; EE; ethinylestradiol; GeoCV%, geometric coefficient of variation in percent; GeoMean, geometric mean; LNG, levonorgestrel; PK, pharmacokinetics; PTF%, peak-to-trough fluctuation over one complete dosing interval at steady state; t_{max}, time to reach the observed maximum concentration.

TABLE 3 | Effect of co-administration of CladT and EE/LNG COC on the PK of EE and LNG: primary PK endpoints (PK analysis set).

Analyte/parameter	EE/LNG COC with CladT	EE/LNG COC with placebo	GMR estimate (%)	90% CI	Intra CV (%)
EE					
$AUC_{\tau,ss}$ (h × pg/mL)	803 (760–847)	828 (784–874)	96.95	91.00–103.28	11.78
$C_{max,ss}$ (pg/mL)	89.9 (85.6–94.4)	89.1 (84.9–93.6)	100.93	95.34–106.84	10.59
LNG					
$AUC_{\tau,ss}$ (h × pg/mL)	93,000 (88100–98,200)	93,000 (88100–98,200)	99.98	93.85–106.52	11.79
$C_{max,ss}$ (pg/mL)	8120 (7730–8530)	8240 (7850–8650)	98.56	93.08–104.36	10.64

Note: Values are presented as the geometric least-squares mean (95% CI), unless otherwise stated; $n = 21$.
Abbreviations: $AUC_{\tau,ss}$, area under the plasma concentration–time curve at steady state during a complete dosing interval (τ); CI, confidence interval; $C_{max,ss}$ maximum observed concentration at a steady state; COC, combined oral contraceptive; EE, ethinylestradiol; GMR, geometric least-squares mean ratio; Intra CV, intra-subject coefficient of variation; LNG, levonorgestrel; PK, pharmacokinetics.

were 803 h × pg/mL and 89.9 pg/mL, respectively, when EE/LNG COC was co-administered with CladT, and 828 h × pg/mL and 89.1 pg/mL, respectively, when EE/LNG COC was co-administered with placebo. Similarly, for LNG, the geometric least-squares means for $AUC_{\tau,ss}$ and $C_{max,ss}$ were 93,000 h × pg/mL and 8120 pg/mL, respectively, when EE/LNG COC was co-administered with CladT, and 93,000 h × pg/mL and 8240 pg/mL, respectively, when EE/LNG COC was co-administered with placebo. The geometric least-squares mean ratios for EE were $AUC_{\tau,ss}$ 96.95% (90% CI: 91.00–103.28) and $C_{max,ss}$ 100.93% (90% CI: 95.34–106.84), whereas those for LNG were $AUC_{\tau,ss}$ 99.98% (90% CI: 93.85–106.52) and $C_{max,ss}$ 98.56% (90% CI: 93.08–104.36). The 90% CIs of the geometric least-squares mean ratios for primary endpoints were within the standard

TABLE 4 | Summary of PD endpoints (PD analysis set).

Parameter	EE/LNG COC with CladT ($n=20$)	EE/LNG COC with placebo ($n=20$)
Progesterone		
Day 13	0.28 ± 0.17	0.29 ± 0.15
Day 14	0.31 ± 0.17	0.32 ± 0.13
Day 15	0.31 ± 0.22	0.32 ± 0.15
LH		
Day 13	3.46 ± 3.25	3.88 ± 3.51
Day 14	2.83 ± 3.08	3.45 ± 4.09
Day 15	2.83 ± 2.80	3.14 ± 3.46
FSH		
Day 13	1.73 ± 1.40	1.92 ± 1.60
Day 14	1.60 ± 1.46	1.87 ± 1.69
Day 15	1.60 ± 1.36	1.81 ± 1.69
SHBG		
Day 13	139.03 ± 48.44	134.85 ± 50.52
Day 14	142.81 ± 46.18	140.51 ± 46.35
Day 15	146.44 ± 47.58	141.41 ± 47.42

Note: Values are presented as mean ± SD.
Abbreviations: CladT, cladribine tablets; COC, combined oral contraceptive; EE, ethinylestradiol; FSH, follicle-stimulating hormone; LH, luteinizing hormone; LNG, levonorgestrel; PD, pharmacodynamics; SHBG, sex hormone-binding globulin.

bioequivalence acceptance range of 80%–125%, confirming the lack of clinically relevant interactions between CladT and the COC components EE and LNG. The intra-subject coefficients of variation (CVs) for EE were estimated as $AUC_{\tau,ss}$ 11.78% and $C_{max,ss}$ 10.59%, whereas those for LNG were $AUC_{\tau,ss}$ 11.79% and $C_{max,ss}$ 10.64%. Furthermore, from Days 10 to 13, the geometric mean C_{max} for plasma cladribine confirmed that all participants were adequately exposed to cladribine (Table S1). The median t_{max} was 0.5 h across all days of assessment.

The serum concentrations of progesterone, LH, FSH, and SHBG, as exploratory PD biomarkers, were comparable when EE/LNG COC was co-administered with CladT or placebo from Days 13 to 15 (Table 4). Based on the results of the ANOVA test, it was concluded that no clinically meaningful difference was noted in the serum concentrations of progesterone, LH, FSH, and SHBG when EE/LNG COC was co-administered with CladT or placebo.

Overall, the co-administration of CladT and EE/LNG COC was generally well tolerated during the study. Across all sequences, periods, and treatments, 20 of 28 (71.4%) participants experienced 112 treatment-emergent AEs (TEAEs), of which 52 were considered related to the study interventions by the investigator (EE/LNG COC run-in period: 5; EE/LNG COC alone: 2; EE/LNG COC with CladT: 33; and EE/LNG COC with placebo: 12).

TABLE 5 | Most commonly reported TEAEs (in ≥ 5% participants) after co-administration of EE/LNG COC and CladT or placebo (safety analysis set).

	Overall ($N=28$) n (%)
Participants with TEAE(s)	20 (71.4)
Headache	15 (53.6)
Fatigue	10 (35.7)
Nasopharyngitis	3 (10.7)
Decreased appetite	3 (10.7)
Intermenstrual bleeding	3 (10.7)
Sinus bradycardia	2 (7.1)
Nausea	2 (7.1)
Pyrexia	2 (7.1)
Migraine	2 (7.1)
Sleep disorder	2 (7.1)

Abbreviations: CladT, cladribine tablets; COC, combined oral contraceptive; EE; ethinylestradiol; LNG, levonorgestrel; TEAE, treatment-emergent adverse event.

However, most AEs were of mild (86 [76.8%]) or moderate (25 [22.3%]) intensity, and only one (0.9%) AE was of severe intensity. One participant experienced Grade 3 lymphopenia 14 days post-administration of CladT, which was evaluated by the investigator as being related to MS treatment (Sequence 1, Period 2). The nature of the AEs experienced by participants receiving EE/LNG COC either with CladT or placebo was comparable. The most commonly reported AEs across treatments were headache and fatigue (Table 5). During the study period, there were no deaths, serious AEs, or discontinuations due to TEAEs, and no clinically relevant changes over time were observed in the standard clinical laboratory parameters, vital signs, ECG measurements, or physical examinations.

4 | Discussion

In this study, a repeat-dose administration of CladT did not have clinically relevant effects on the PK of a frequently prescribed EE/LNG COC, suggesting a lack of PK interaction between CladT and EE/LNG-based COC in pre-menopausal women with active RMS. ANOVA confirmed the bioequivalence of EE and LNG in COC when co-administered with either CladT or placebo. The primary and secondary PK parameters of EE and LNG in the plasma were similar when co-administered with CladT or placebo. The cladribine mean C_{max} observed in this study was generally consistent with previously reported estimates in patients with RMS at the approved dosage [7]. Furthermore, repeated administration of CladT had no apparent effect on serum LH, FSH, progesterone, or SHBG concentrations during concomitant treatment with COC containing EE and LNG.

No significant interactions have been reported with the concomitant use of oral contraceptives and MS DMTs [18]. Drug–drug interaction studies on the concomitant use of COCs and MS DMTs are scarce [19] or are limited to healthy women. COCs

are metabolized by CYP3A4 as well as other drug-metabolizing enzymes including conjugative pathways of sulfation and glucuronidation [20]. As such, the concomitant use of enzyme inducers may lead to their reduced efficacy [20]. Previous in vitro studies have shown that cladribine has no clinically meaningful inductive effect on CYP3A4 enzymes [8]. However, the effect of cladribine on the PK of oral contraceptive steroids has not been evaluated in humans. Due to the potential teratogenic effects of cladribine, the European Medicines Agency requested a drug–drug interaction study with an oral contraceptive as a post-approval commitment. Therefore, this study was designed to investigate the potential effects of CladT on the PK of COCs in pre-menopausal women with RMS.

CladT induces long-lasting decreases in lymphocyte counts as part of its principal mode of action, occasionally leading to severe lymphopenia (i.e., Grade 3). Therefore, it was not ethically feasible to conduct this study in a healthy population. The study design is consistent with drug–drug interaction studies examining the effects of investigational drugs on the PK of hormonal oral contraceptives [12], and the unique study design ensured that each participant received an effective treatment as per the approved label for CladT. The CladT dosing regimen is based on body weight, and all participants included in this study received a different total dose of cladribine. Participants with a very low body weight (i.e., <50 kg) would have received only a 4-day treatment per treatment week instead of a 5-day treatment; therefore, the study enrolled participants with a body weight of ≥ 50 kg to allow for a standardized 5-day repeat-dose CladT treatment for all study participants. This approach helped to standardize the CladT treatment duration, thereby reducing the complexity of the study design and ensuring the assessment of EE/LNG PK at the point of maximal potential interaction on Day 5 of co-administration with CladT.

In line with the previously reported non-clinically meaningful effect of cladribine on CYP3A4 [7, 8], the current study demonstrated a lack of PK interaction between CladT and the COC components EE and LNG. The metabolism of EE primarily occurs through hydroxylation, predominantly mediated by CYP3A, with a minor contribution from CYP2C9. Additionally, glucuronidation and sulfation are significant clearance pathways. Similarly, CYP3A4 is the primary P450 enzyme responsible for the reduction and hydroxylation of LNG, with CYP3A5 and CYP2C9 playing a lesser role, followed by conjugation [20]. It is acknowledged that the relative contributions of CYP3A4 and other inducible drug-metabolizing enzymes to the overall metabolic clearance of oral contraceptive steroids, and specifically CYP3A4-metabolized progestins, are variable [21, 22], and this may translate to potential differences in sensitivity to CYP3A4 inducers. Nevertheless, the absence of an interaction of CladT with EE/LNG in the present drug–drug interaction study, where exposures of EE and LNG were equivalent during co-administration with CladT in reference to during co-administration with placebo, provides important evidence for a lack of a clinically relevant interaction. Taken together with the lack of inductive effect of cladribine on CYP3A4 in vitro, it may be concluded that although this study specifically focused on the drug–drug interaction between CladT and COCs containing EE and LNG, the outcomes can be extrapolated to other hormonal contraceptives containing EE as an estrogen component and LNG or other progestin components metabolized by CYP3A4 (e.g., norethindrone and norelgestromin).

The co-administration of CladT and EE/LNG COC was found to be well tolerated overall during the study, with headache (53.6%) and fatigue (35.7%) the most commonly reported TEAEs. Additionally, there were no serious AEs or TEAEs leading to study discontinuation. However, one participant experienced Grade 3 lymphopenia, which was evaluated as being related to the study intervention by the investigator. Of note, lymphocyte count reductions are expected due to the mechanism of action of cladribine, and the reported frequency is consistent with previously published literature [23].

This study was affected by the COVID-19 pandemic in Poland and Germany between January 2019 and March 2022, which led to a temporary study discontinuation/interruption for safety reasons, as it was unknown whether patients with MS would experience a more severe course of COVID-19 infection. Two participants were affected by the temporary discontinuation for safety reasons and were withdrawn from the study during the run-in period. However, data collected during the COVID-19 pandemic showed that patients treated with CladT for MS are generally not at greater risk of serious disease and/or a severe outcome with COVID-19 compared with either the general population or the patients with MS [24]. Overall, the COVID-19 pandemic did not impact data reliability or the ability to monitor and manage participant safety during the conduct of the study, but had a substantial impact on the progress of the study on the time axis.

5 | Conclusions

Co-administration of CladT did not alter the PK of EE or LNG in a COC. Therefore, the concomitant use of CladT is not expected to decrease the efficacy of EE/LNG-based COC as a means of birth control in women with RMS.

Author Contributions

All authors wrote the manuscript. R.H., A.G., and A.K.-B. designed the research. K.H. performed the research. S.G., A.B., D.J., A.K.-B., C.V., A.N., K.V., and J.Q.D. analyzed the data. All authors reviewed the manuscript, approved the final version, and agree to be accountable for all aspects of the work.

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Ethics Statement

The protocol was approved by the independent ethics committee of Ruhr University Bochum, Germany (No. 18–6371 FF) and the Bioethical Committee of the Regional Medical Chamber in Warsaw, Poland (No. KB/1198/18). This study was conducted in compliance

with the principles of the Declaration of Helsinki and the International Council on Harmonization Good Clinical Practice Guidelines as well as applicable laws and regulations. Written informed consent was obtained from all participants prior to study commencement.

Consent

The authors have nothing to report.

Conflicts of Interest

R.H. is an independent Clinical Pharmacology consultant and served as an external Clinical Pharmacology expert advisor for various aspects in the clinical development of Cladribine. He received financial support for research, consulting, and training services from the healthcare business of Merck KGaA, Darmstadt, Germany. K.H. has received speaker honoraria and research support from Bayer, Biogen, the healthcare business of Merck KGaA, Darmstadt, Germany, Novartis, Sanofi Genzyme, Roche, and Teva; has received support for congress participation from Bayer, Biogen, the healthcare business of Merck KGaA, Darmstadt, Germany, Roche, Sanofi Genzyme, and Teva; and has served on scientific advisory boards for Bayer, Biogen, Sanofi, Teva, Roche, Novartis, and the healthcare business of Merck KGaA, Darmstadt, Germany. S.G., A.B., A.K.-B., C.V., and A.N. are employees of the healthcare business of Merck KGaA, Darmstadt, Germany. A.G. was an employee and is now a consultant to Ares Trading SA, Eysins, Switzerland, an affiliate of Merck KGaA, Darmstadt, Germany. D.J. is an employee of Merck Serono Ltd., Feltham, UK, an affiliate of Merck KGaA, Darmstadt, Germany. K.V. and J.Q.D. are employees of EMD Serono, Billerica, MA, USA.

Data Availability Statement

Any requests for data by qualified scientific and medical researchers for legitimate research purposes will be subject to the Data Sharing Policy of the healthcare business of Merck KGaA, Darmstadt, Germany. All requests should be submitted in writing to the data sharing portal for the healthcare business of Merck KGaA, Darmstadt, Germany https://www.emdgroup.com/en/research/our-approach-to-research-and-development/healthcare/clinical-trials/commitment-responsible-data-sharing.html. When the healthcare business of Merck KGaA has a co-research, co-development, co-marketing, or co-promotion agreement, or when the product has been out-licensed, the responsibility for disclosure might be dependent on the agreement between parties. Under these circumstances, the healthcare business of Merck KGaA will endeavor to gain agreement to share data in response to requests.

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Supporting Information

Additional supporting information can be found online in the Supporting Information section.