HORMONAL EFFECTS ON COGNITION IN ORAL CONTRACEPTIVE USERS AND

NATURALLY-CYCLING WOMEN

by

BRITTANY HAWKSHEAD

(Under the Direction of Lawrence H. Sweet)

ABSTRACT

The combined oral contraceptive pill (COCP) is the most common method of hormonal birth control for U.S. women, and there is a growing body of literature investigating hormone-related effects on cognition. Much of the research suggests that verbal memory and visuospatial functioning improve with COCPs in comparison to the natural menstrual cycle (MC). Hormonal variations during the natural MC have also been shown to impact cognition, with periods of high sex hormones (i.e., luteal phase) associated with improved verbal memory, and periods of low sex hormones (i.e., follicular phase) associated with improved visuospatial abilities. We analyzed group differences in cognitive performance using two separate designs: (a) discordant siblings and (b) demographics-matched groups. Overall, visuospatial functioning did not differ as a result of MC phase or COCP use. However, verbal memory scores were significantly higher for women during the follicular phase than during the luteal phase, contrary to expectations.

INDEX WORDS: sex hormones, cognition, oral contraceptives, menstrual cycle,

neuropsychology

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CHAPTER 1

INTRODUCTION

Hormonal Contraceptives

The most common method of birth control for U.S. women is the combined oral

contraceptive pill (COCP). A recent DHHS report revealed that over 17% of U.S. women

between the ages of 15 and 44 currently use COCPs as their primary contraceptive method, and

that 82% of women will use COCPs at some point in their lives (Mosher & Jones, 2010). COCPs

are composed of a mixture of synthetic forms of two naturally-occurring sex hormones –

estrogen and progesterone (via progestins) – in a combination designed to inhibit ovulation by

suppressing the release of gonadotrophin-releasing hormone, luteinizing hormone, and follicle-

stimulating hormone. A monthly COCP "pack" is intended to parallel the length of a typical

menstrual cycle (MC) and usually includes 28 pills – 21 of which contain the synthetic sex

hormones, and 7 which are placebo pills that allow for a monthly withdrawal bleeding intended

to mimic a typical MC (Hatcher, 2011). In comparison to a natural MC, COCPs have been found

to lower levels of endogenous estrogen and progestogen by 15-30% (Sahlberg, Landgren, &

Axelson, 1987). During use of COCPs, estrogen levels stay relatively low (i.e., 25-50 pg/ml) and

are without the typical peaks observed during the natural MC (Wuttke et al., 1975). Serum

progestogen levels are also reduced with COCP use and usually remain steady around 1 ng/ml,

without any significant fluctuations; this is in contrast to progestogen levels that vary throughout

a natural MC and whose peak can reach more than 9 ng/ml. In addition, COCPs have also been

shown to reduce endogenous *testosterone* levels (total and free) in comparison to levels seen at

any phase during a natural MC (Graham, Bancroft, Doll, Greco, & Tanner, 2007; Hietala, Sandberg, Borg, Olsson, & Jernström, 2007; Jung-Hoffman & Kuhl, 1987). A recent meta-analysis by Zimmerman, Eijkemans, Coelingh Bennink, Blankenstein, and Fauser (2013) found that free testosterone levels decrease by an average of 61% for women using COCPs in comparison to those who are naturally-cycling. Of note, this reduction in testosterone is believed to play a role in many of the non-contraceptive effects (e.g., cognition, emotion) linked to COCP use (Graham et al., 2007).

Reasons for COCPs use extend well beyond birth control. Several non-contraceptive benefits of COCPs include: improved control of menstrual-related symptoms (physical, psychological, and behavioral); acne and hirsutism; improved bone health; prevention of ovarian, endometrial, and colorectal cancers; and overall reduction in mortality (Dragoman, 2014). In addition, they are often prescribed as a method of menstrual regulation for women with amenorrhea or oligomenorrhea, and they are used to aid in the treatment of other disorders such as endometriosis, polycystic ovary syndrome, menorrhagia, and dysmenorrhea. Broadly, there is growing evidence that the use of COCPs has considerable influence outside of the reproductive system and into far-reaching systems including cognition (Gogos, Yeewen, Williams, & Byrne, 2014; Warren, Gurvich, Worsley, & Kulkarni, 2014).

Hormone Effects on Cognition with COCP Use

It has long been known that hormones affect cognition. For instance, the cognitive effects of hormone replacement therapy (HRT) in post-menopausal women have been well documented (LeBlanc, Janowsky, Chan, & Nelson, 2001). Current research suggests that HRT improves scores on tasks of verbal and spatial episodic memory, abstract reasoning, information

processing, and working memory (Doty et al., 2015; Hogervorst, 2000). However, it is important to note that in post-menopausal women, HRT is used to *increase* hormone levels during a time when women's natural levels are reduced; this contrasts with COCPs, which *decrease* levels of estrogen and progesterone in pre-menopausal women. Nevertheless, it is clear that administration of hormones may play a key role in a variety of cognitive processes for women across the lifespan.

Verbal Functions

Although the current literature on COCPs and cognitive functioning is mixed, verbal and visuospatial domains appear to be particularly sensitive to COCP use (Gogos et al., 2014; Warren et al., 2014). While some studies have failed to find significant COCP-related effects (Gordon & Lee, 1993; Islam, Sparkes, Roodenrys, & Astheimer, 2008), many other researchers have found that verbal memory improves (e.g., better scores on word memory tests) during active COCP use (Gogos, 2013; Mordecai, Rubin, & Maki, 2008). Specifically, Gogos (2013) demonstrated that scores significantly differed between the four groups tested (i.e., women using COCPs, "Low estrogen and progesterone" [i.e., follicular] women, "High estrogen and progesterone" [i.e., luteal] women, and men), with those on COCPs performing the best, followed by luteal women, follicular women, and then men; these patterns held true for tasks assessing immediate and delayed verbal memory, as well as for information presented in word list and narrative format. Some investigators have theorized that despite COCPs reducing overall levels of estrogen and progesterone (typically associated with worse verbal abilities), the accompanying reduction in testosterone levels due to COCP use may be implicated in this improved verbal memory performance (Mordecai et al., 2008). This conclusion is supported by

literature reporting that testosterone and verbal memory abilities are negatively correlated in women (Mordecai et al., 2008; Phillips & Sherwin, 1992).

Overall, a notable proportion of the limited literature on the cognitive effects of COCP use suggests that verbal memory may improve as a result of COCPs, with no available studies suggesting that it declines. Moreover, the studies that have reported no significant effects tended to have small samples (i.e., 16-34 COCP-users per study), raising questions about statistical power necessary to support the conclusion of a null finding. Unfortunately, further research examining the potential effects of COCPs on other verbal and language abilities is notably lacking.

Visuospatial Functions

Another domain of interest for researchers examining COCP effects on cognition is visuospatial functioning. Similar to the literature on COCPs and verbal memory, the current evidence for COCPs on visuospatial abilities is also mixed, with some studies finding no significant effects of COCPs (Gogos, 2013; Gordon & Lee, 1993; Islam et al., 2008; Mordecai et al., 2008; Wuttke et al., 1975), some revealing variable effects (McFadden, 2000), and one even reporting negative effects due to COCP use (Griksiene & Ruskenas, 2011). However, many studies have found that COCPs *improve* spatial and visuospatial abilities, assessed using a range of tasks (Beltz, Hampson, & Berenbaum, 2015; Cicinelli et al., 2011; Wright & Badia, 1999). For instance, COCP-users performed significantly better on tasks of both visuospatial calculation and spatial processing in comparison to women during the follicular phase of the natural MC (Becker, Creutzfeldt, Schwibbe, & Wuttke, 1982; Wright & Badia, 1999). A visual attention (i.e., line bisection) task also revealed a similar pattern of findings for women who were tested

while on COCPs and off COCPs; women performed best during active pill use (Cicinelli et al., 2011). Accordingly, despite relatively mixed findings, much of the existing research literature reports considerable improvements on a range of visuospatial tasks with traditional COCP use.

Other Cognitive Functions

In comparison to the literature on visuospatial and verbal abilities, research into the effects of COCP use on other cognitive processes has not yielded consistent patterns. For example, within domains of attention, information processing speed, and executive functions (e.g., working memory, mental flexibility, inhibitory control, verbal fluency), there is no clear consensus on the likely impact of COCPs (Warren et al., 2014). While some studies have suggested that COCPs may improve performance in these domains (Gogos et al., 2014; Griksiene & Ruksenas, 2011; Wright & Badia, 1999), the majority of studies have observed no significant differences as a result of COCP use (Gordon & Lee, 1993; Islam et al., 2008; Komenich, Lane, Dickey, & Stone, 1978; Mordecai et al., 2008; Nielsen, Ertman, Lakhani, & Cahill, 2011), and a few studies have suggested that COCPs may impair these functions (Becker et al., 1982; Garrett & Elder, 1984; Wuttke et al., 1975). Therefore, there is little evidence to suggest directional effects within these domains as a result of COCPs.

The Menstrual Cycle

A typical menstrual cycle (MC) lasts approximately 25-34 days – with an average of 28 days – and may be divided into three parts: the follicular phase, ovulation, and the luteal phase. The follicular phase begins on the first day of menstruation and lasts until ovulation (i.e., about 14 days, or halfway through the MC). The luteal phase follows ovulation and lasts until menses

begins again (i.e., approximately the final 14 days). Across the MC, there are systematic variations in serum sex hormones. At the beginning of the cycle, estrogen remains relatively low, hovering between 25 and 75 pg/ml until it reaches its first peak during the late follicular phase, immediately prior to ovulation (Wuttke et al., 1975). Estrogen levels then increase substantially (to approximately 200 pg/ml), before starting to decline following ovulation. Estrogen levels then rise towards a second peak that occurs during the middle-to-late luteal phase, just prior to menstruation; at this point, estrogen levels reach around 150 pg/ml, before falling back to their baseline as the cycle begins anew (Wuttke et al., 1975). Similarly, serum progesterone levels begin the cycle at their lowest levels (i.e., around 1 ng/ml) and remain relatively stable until ovulation, at which point they begin to increase (alongside estrogen's peak) until they reach their height (of approximately 7-9 ng/ml) during the mid-to-late luteal phase; progesterone levels decline towards baseline in the days before the cycle ends (Mihm, Gangooly, & Muttukrishna, 2011; Wuttke et al., 1975). Testosterone levels are believed to remain relatively low and stable across the typical MC, excluding a brief peak during ovulation; as such, testosterone levels do not appear to differ between luteal and follicular phases (Judd & Yen, 1973).

Hormone Effects on Cognition during the Menstrual Cycle

Evidence from an emerging literature suggests that variations in these hormones across the MC impact performance on varied neurocognitive tasks. Some of the earliest findings in the field assessed "sexually dimorphic differences" (i.e., termed "masculine" visuospatial vs. "feminine" verbal by these authors) associated with fluctuating levels of hormones – particularly estrogen – across the MC (Hampson, 1990; McCormick and Teillon, 2001). Several studies found that periods of *higher* estrogen (i.e., late luteal) were associated with improved

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performance on "feminine" tasks measuring verbal memory, while periods of *lower* estrogen (i.e., early follicular) were associated with poorer verbal memory task performance (Hampson, 1990; Maki, Rich, & Rosenbaum, 2002). In contrast, *lower* estrogen levels during the MC (i.e., early follicular) were found to be related to higher scores on visuospatial tasks (e.g., mental rotation, spatial processing, visual reproduction, and perceptual priming) and tests of arithmetic (Becker et al., 1982; Pletzer, Kronbichler, Ladurner, Nuerk, & Kerschbaum, 2011), while *higher* estrogen levels (i.e., late luteal) were shown to be related to lower scores on visuospatial tasks (Maki et al., 2002; Silverman & Phillips, 1993; Wright & Badia, 1999). For instance, one study demonstrated that women performed better on a spatial integration and memory task during their "non-menstrual phase" (in which estrogen and progesterone levels are lower) than during their "menstrual phase," with its higher levels of hormones (Postma, Winkel, Tuiten, & van Honk, 1999). In an attempt to further explain these findings, some researchers have sought to identify neural correlates for these visuospatial tasks. For example, a pair of studies found that women during their luteal phase (i.e., high estrogen) demonstrated increased reactivity in the angular gyrus – a region often implicated in spatial judgment – and theorized that this greater reactivity was associated with increased need to recruit this area to solve the visuospatial task, suggesting that they are having greater difficulty during this higher estrogen period (Dietrich et al., 2001; Schoning et al., 2007).

Other studies have revealed that performance in other cognitive domains may also be associated with changes in hormones across the MC, though findings are often mixed or inconsistent. Nevertheless, memory and attention tasks appear to be positively associated with periods of increased estrogen during the MC (Gogos, 2013; Otero Dadín, C., Rodríguez Salgado, & Andrade Fernández, 2009; Rosenberg & Park, 2002). In addition, tasks requiring fine motor

skills may also improve during periods of higher estrogen (Hampson, 1990). Overall, problems with research examining MC effects on cognition are similar to those associated with COCP effects on cognition. Further, a number of concerns still exist including: small sample sizes causing lack of power, variability in types of tasks used (i.e., poorly validated), as well as other design considerations.

Summary and Rationale

In contrast to a relatively strong body of evidence on the effects of sex hormones on cognitive function in older adults (e.g., during HRT), additional research is needed to understand patterns that are emerging from the limited literature on cognitive effects of sex hormones in premenopausal women. Verbal memory and visuospatial processing have shown some of the largest and most consistent effects in prior studies examining cognition as a function of COCP use and along phases of the natural MC (Gogos et al., 2014; Warren et al., 2014).

In naturally-cycling women, it has been shown that periods of high estrogen and progesterone – *such as in the luteal phase* – are associated with improved performance on tasks assessing more typically "feminine" domains such as verbal memory. In contrast, periods of lower estrogen and progesterone, such as the follicular phase, has been linked to higher scores on historically "masculine" tasks such as visuospatial processing. COCP use leads to lower overall levels of estrogen, progesterone, and testosterone, and is associated with improved performance on visuospatial and verbal memory measures.

Although the idea that higher estrogen levels facilitate typically "feminine" functions (e.g., verbal memory) and lower estrogen levels facilitate more "masculine" functions (e.g., visuospatial processing) is relatively straightforward, interpretation of these findings in the

context of findings from COCP studies is not. Specifically, the pattern of COCPs improving

performance on verbal memory tasks – despite leading to lower estrogen levels than seen during

any phase of the MC – seems to contradict studies demonstrating the opposite (i.e., that higher

estrogen levels improves verbal memory) in naturally-cycling women. While no explicit models

for this mechanism have yet been established, some researchers have theorized that the improved

verbal performance during COCP use is due to the COCP-induced reduction in testosterone,

which has been supported by negative correlations observed between testosterone and verbal

memory performance in women (Mordecai et al., 2008; Phillips & Sherwin, 1992). Thus, despite

overall lower estrogen levels due to COCPs (in comparison to the natural MC), the COCP-

associated reduction in testosterone may drive these verbal memory effects above and beyond

the effects from estrogen. Therefore, different mechanisms likely exist to clarify how periods of

high estrogen (during the MC) and periods of lower estrogen (resulting from COCPs) can each

be associated with improved verbal memory. Given this pattern, it would follow that

performance on verbal tasks would be best during periods of lowest testosterone (i.e., on COCPs

in comparison to *either* phase of the natural MC), with further distinction being made based on

estrogen levels (i.e., better performance during luteal vs. follicular phases). For visuospatial

abilities, one would assume that lower testosterone due to COCP use would aid task performance

when compared to *either* phase of the MC, but that the additional consideration of reduced

estrogen and progesterone during the follicular phase would further facilitate visuospatial

processing in comparison to higher levels during the luteal phase.

  The inconsistency of findings in this emerging literature is also due to practical reasons.

In addition to an overall lack of research in this area, there are substantial methodological

problems. First, although there is a growing body of literature examining how sex hormones

affect cognition in *naturally-cycling* women, there is a distinct lack of research on the effects of *synthetic* hormones in healthy, pre-menopausal women, particularly as a result of COCPs (Gogos et al., 2014; Warren et al., 2014). Second, there are notable methodological weaknesses in the current literature, including small sample sizes (i.e., mean COCP-users per study of 24 according to a recent systematic review; Warren et al., 2014), which negatively impacts power to detect true effects, as well as the use of unmatched between-subjects designs that were not be able to control for potential confounding variables (e.g., education, income, relationship status). Further, researchers have been generally unable to conduct randomly controlled trials given ethical concerns related to administering placebo or "sham" contraceptives to young women (Warren et al., 2014). Thus, there is a need to carefully examine the relationship between COCPs and cognition using a design and sample that better accounts for these factors.

This study aimed to provide a better understanding of the potential effects that COCPs may have on women's cognition given the widespread usage of COCPs throughout the past several decades (Mosher & Jones, 2010). Given the typical makeup of most human social sciences research (i.e., undergraduates in psychology courses from western, industrialized nations) and the high rates of COCP use in this population, we believed it was important to identify potential variables that may differentially affect research samples (Henrich, Heine, & Norenzayan, 2010). Another research benefit was the use of publicly-available cognitive tests that are validated and can be easily replicated in later studies, which may help provide potential comparison groups for future research. It was also important that cognitive effects of natural and synthetic sex hormones be identified and disseminated so to better inform women's beliefs and expectations regarding potential cognitive changes across their MC or as a result of COCP use. Further, we believed that this information may allow women to make more informed decisions

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about whether to begin, continue, or terminate COCP use given their personal needs and situation, particularly as it relates to cognitive strengths and weaknesses.

Aims

The overall purpose of the current study was to evaluate whether variations in sex hormones due to COCP use and MC phase were associated with differences in cognitive task performance. There were two specific aims. The first aim was to identify broad effects of COCP use on cognition with regards to verbal memory and visuospatial functioning in discordant sibling (i.e., sister) pairs. The second aim was to compare women using COCPs to those who are naturally-cycling at distinct phases (i.e., early follicular and late luteal) to determine how cognitive performance differs across verbal and visuospatial domains.

We proposed to address gaps in prior literature by examining the effects of COCP and MC phase as a function of cognitive domain using two specific study designs. First, we used a novel discordant sibling design (i.e., on/off COCPs) to examine cognitive performance within a relatively large sample of women in which the potential confounds due to environmental and genetic factors may be partially-controlled. Our second study design consisted of a between-subjects examination of hormone status (i.e., COCP users vs. luteal phase vs. follicular phase) on cognition using demographics-matched groups.

Overall, we predicted that COCP-users would perform better on tasks in both verbal and visuospatial domains than naturally-cycling women (see Hypothesis 1). We also predicted that luteal-phase women (i.e. high estrogen/progesterone) would perform better than women in the follicular phase (i.e., low estrogen/progesterone) on a verbal memory task, but that this pattern

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would reverse on a visuospatial task, with follicular women outscoring luteal women (see Hypothesis 2).

Hypotheses

1. *Hypothesis 1:* Within the COCP-discordant sibling design, we predicted that women using COCPs would demonstrate higher scores on the verbal memory and visuospatial tasks than their naturally-cycling sisters.

2. *Hypothesis 2*: We predicted that luteal-phase women would outperform follicular-phase women on the verbal memory task, and follicular-phase women would outperform luteal-phase women on the visuospatial task. We also expected that women using COCPs would outperform naturally-cycling women on both cognitive tasks, regardless of MC phase.

Design Considerations

Throughout the study design process, several other analyses were considered. An ideal study would assess the same individuals on and off COCPs using a double-blinded, randomly-selected, within-subjects design (i.e., on/off COCPs); however, this is not easily feasible given (a) the ethical issues associated with administering a placebo or "sham" contraceptive condition, and (b) the threats to internal and external validity associated with using alternate forms of contraception. Therefore, a discordant sibling pair design was considered an effective approach so to best match participants on a range of biological (e.g., genetics) and environmental variables (e.g., family history, childhood socioeconomic status), while avoiding notable threats to internal, external, and ecological validity (Hypothesis 1). Our second aim was to examine whether cognitive performance differed by MC phase on tasks of verbal memory and visuospatial

functioning. Our initial plan was to continue utilizing this discordant sibling sample to conduct

further analyses contrasting the naturally-cycling women in each the (a) luteal and (b) follicular

phases to their COCP-discordant siblings; however, it was determined that this plan would leave

us with a sample size too small to ensure adequate power (i.e., 11 and 17 pairs, respectively),

despite the sibling-matched design. Thus, we decided to develop a second study with a sample of

women separated into three groups (i.e., COCP, luteal, follicular) that were matched on specific

demographic variables, rather than the inherent sibling relationship. We chose this second design

to allow us to explore the possible effects of MC phase (in addition to COCP use) on verbal

memory and visuospatial functioning.

CHAPTER 2

METHODS

Participants

Participants were selected from the WU-Minn Human Connectome Project (HCP), an open-access initiative seeking to facilitate the examination of brain function and behavior in young adult subjects (aged 22-35) from families with twin and non-twin siblings (Van Essen et al., 2013). All subjects provided informed consent prior to study participation. The sample for the present study was from the March 2017 data release comprising 1206 subjects (http://www.humanconnectome.org).

HCP Exclusion Criteria

Per Van Essen and colleagues (2013), the overall HCP study exclusionary criteria included: (a) past or present psychiatric diagnosis, including substance abuse; (b) major neurological disorder (e.g., epilepsy, multiple sclerosis) or previous brain injury; (c) cardiovascular disease or genetic disorder (e.g., cystic fibrosis, sickle cell disease); (d) premature birth; (e) history of cancer treated by radiation or chemotherapy; (f) treatment of thyroid hormone, diabetes, or migraines lasting one month or more; (g) a score below 26 (of 30) on the Mini Mental Status Exam (MMSE; Folstein, Folstein, & McHugh, 1975); and (h) standard MRI research contraindications (e.g., metal in body, claustrophobia, pregnancy).

Study Exclusion Criteria

Only women with complete data from the full HCP sample (1206 participants) were considered for inclusion. Additional study-specific exclusionary criteria included: (a) recent or current breastfeeding, and (b) diagnosis of endometriosis, polycystic ovary syndrome (PCOS), or any other disease significantly affecting the MC. For naturally-cycling women, participants were required to have regular MCs (i.e., 25-35 days) and not currently be using any form of hormonal contraceptive (e.g., COCP, birth control shot, implant) or fertility drug. Among women currently using COCPs, use of any other hormonal contraceptive or fertility medication, and/or history of major abnormalities with their natural MC (e.g., amenorrhea, oligomenorrhea) were exclusionary. In addition, women endorsing use of an intrauterine device (IUD) were excluded due to our inability to differentiate those women using non-hormonal (i.e., copper) from hormonal IUDs. For the second analysis requiring naturally-cycling women to be in a specific phase of their MC, subjects were assigned to a group according their self-report of the number of days since the start of their last menstruation. Specifically, women were labeled as *follicular* if they were within days 2-8 of their cycle onset, and *luteal* if they were within days 19-25. These relatively narrow ranges were used as they are frequently considered reliable estimates of the high and low points of the MC in terms of estrogen and progesterone levels (Gogos, 2013). Despite notably reducing the number of participants eligible for these analyses, this restriction allowed us to increase sensitivity and prevent extraneous findings as a result of potential variations in hormone levels throughout the MC.

We used G*Power (Faul, Erdfelder, Lang, & Buchner, 2007) to perform a sensitivity

analysis with the specified parameters of power = 0.80 and two-tailed alpha = .05, to determine

the minimum detectable effect (MDE) given our *set* sample of 59 discordant female sibling pairs.

Results from the power analysis indicate that our sample should provide us with adequate power

to detect at least a medium effect ($\Lambda = 0.41$) using a paired Hotelling's T^2 test – a multivariate

analysis technique generalized from the univariate *t* test. Given recent literature reporting a

medium-to-large effect size for the impact of COCP use on overall cognition ($\eta^2 = 0.25$, per

Gogos et al., 2013), it was determined that our estimated sample of 59 discordant pairs ($N = 118$)

should be sufficient to address Hypothesis 1.

A one-way MANOVA with one IV (group) with three factors (COCP vs. Luteal vs.

Follicular) and two DVs (verbal memory and visuospatial functioning) was planned to test

Hypothesis 2. Using the same parameters as above (power = 0.80 and two-tailed alpha = .05), an

a priori power analysis using G*Power indicated that a sample size of 99 was required to detect

a medium effect (i.e., $f^2 = .065$). Therefore, the estimated sample size of 135 women (divided

into three groups of 45) should provide sufficient power for this analysis.

Procedures

HCP study procedures were reviewed and approved by the Washington University

Human Research Protection Office and HIPAA Privacy Offices. Participants completed the full

HCP protocol over a two-day visit during the data collection period from 2012-2015 (Van Essen

et al., 2013). Subjects were administered behavioral measures assessing a range of motor,

sensory, cognitive, and emotional functions. Some of these behavioral measures were taken from

the NIH Toolbox Assessment of Neurological and Behavioral Function, while several other measures were included to assess domains not covered by the NIH Toolbox, including the primary variables for the current study – the Variable Short Penn Line Orientation Test and the Penn Word Memory Test (Gur et al., 2001; Gur et al., 2010). All behavioral and cognitive measures were completed over a 3-4 hour period. All assessments were selected to be reliable and well-validated (Van Essen et al., 2013).

Sample Selection

Two separate samples were selected from the full dataset corresponding to each of the two aims. For the first analysis examining sibling-matched pairs discordant for COCP use (Hypothesis 1), our final sample included 118 participants (59 sibling pairs). Decision rules for families with more than one sibling eligible for each group included: (a) selection of closest sibling relationship available (i.e., monozygotic twin > dizygotic twin > non-twin sibling), and (b) selection of sibling closest in age to match. For the second analyses comparing three groups of demographically-matched women (naturally-cycling women during the follicular phase, naturally-cycling women during the luteal phase, and women using COCPs), we expected our final sample size to total 135 subjects (Hypothesis 2). Each of the three groups comprised 45 women matched for relevant demographic characteristics – including age, race, and education level – to control for potential confounding effects.

For all participants, quality control was ensured by excluding participants with missing data on the primary variables, as well as by identifying and appropriately managing outliers. All variables were evaluated to determine whether parametric statistical assumptions were met (e.g., normalcy via skewness, kurtosis, heteroscedasticity). If the data was not to meet these

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assumptions, appropriate steps were implemented to address the problem (e.g., data transformations, exclusion or replacement of missing data, use of non-parametric statistics).

Measures

Demographic

Participants reported relevant demographic variables, including gender, age, and family information (e.g., sibling relationship, twin status) during a structured interview on their first visit. Self-reported sibling status (zygosity) was verified through genotyping. Other variables collected included self-reported educational attainment, relationship status, and race. Several of these variables were used to match participants by group for the Hypothesis 2 analysis, which allowed us to increase internal validity and account for potential pre-existing group differences. Sample characteristics were also reported to provide a basis for readers evaluate generalizability.

Health and Menstrual Cycle

Women were asked several questions about their MC during their initial intake interview on day one of the HCP assessment. Participants reported their contraceptive use, cycle regularity, cycle length, and days since last menstruation.

Cognitive

Participants were administered the Penn Word Memory Test (Form A) to assess verbal episodic memory (Gur et al., 2001; Gur et al., 2010). On this task, participants were asked to learn a list of 20 words for a subsequent memory test. They were then shown 40 words (comprising the 20 previously presented words and 20 related, novel words) and asked to

determine whether they had previously seen each word by choosing: "definitely yes," "probably yes," "probably no," or "definitely no." Spatial orientation was assessed via the Variable Short Penn Line Orientation Test (Gur et al., 2001; Gur et al., 2010). During this task, participants were shown two lines with different orientations and asked to manually rotate one of the lines (via keyboard button pressing) so that it was parallel to a target line that varied in length and rotation across 24 trials. These tasks were selected due to existing research (reviewed previously), which suggested that the greatest hormone-related effects were likely to occur in these cognitive domains (Gogos et al., 2014; Warren et al., 2014). In addition, participants completed the Penn Progressive Matrices test (Bilker et al., 2012) and the NIH Toolbox Picture Vocabulary Test (Gershon et al., 2014) – estimates of fluid (i.e., nonverbal IQ) and crystallized (i.e., verbal IQ) intelligence, respectively – as a check for internal validity (i.e., so to ensure that matched-groups did not systemically differ on potentially confounding variables). All scores were non-unadjusted raw or standard scores, as available.

Data Analysis

Statistical analyses were performed using SPSS 23. Given the nature of our designs comprising two correlated, continuous outcome variables (i.e., verbal and visuospatial functioning), we used multivariate analyses of variance (MANOVA). By using MANOVA (rather than individual ANOVAs), we were able to increase our power given the correlation between outcome variables, as well as control the family error rate. These analyses also allowed us to detect potential factors that may be influencing the relationship between outcome variables. Thus, to address Hypothesis 1 – comparing sibling pairs discordant for COCP use – a paired Hotelling's T^2 test was used to evaluate the effect of COCPs on cognitive functioning, with

COCP use (on/off) as the IV and verbal memory and visuospatial functioning as the two DVs. This analysis was intended to determine whether the naturally-cycling women exhibit worse cognitive function (verbal memory and visuospatial) than their COCP-discordant siblings across variables, as was initially predicted. For Hypothesis 2, we used a one-way MANOVA to assess differences in cognitive performance among demographics-matched groups, using the same DVs (verbal memory and visuospatial functioning) across three levels of our IV of group (luteal, follicular, COCP). This analysis was chosen to determine the effect of MC phase (in addition to COCP-use) on cognitive functioning. Per our hypothesis, we expected that COCP-users would continue to demonstrate the strongest performance (compared to the naturally-cycling women), but that luteal-phase women would outperform follicular-phase women on the verbal memory task, while follicular-phase women would outperform luteal-phase women on the visuospatial task.

CHAPTER 3

RESULTS

Data analyses yielded two sets of results. The first set addressed Hypothesis 1 via a

multivariate Hotelling's T^2 analysis comparing differences in cognitive performance in two

domains (i.e., visuospatial and verbal memory) between sibling pairs discordant for COCP use.

The second set of results addressed Hypothesis 2 via a one-way MANOVA that examined group

differences in cognitive performance among: (a) naturally-cycling women during the luteal

phase; (b) naturally-cycling women during the follicular phase; and (c) women on COCPs, using

a demographics-matched, between-groups design. All statistical thresholds were two-tailed at $p <$

.05, unless otherwise noted.

Discordant Sibling Study

Sample Characteristics

A total of 118 participants (59 discordant sibling pairs) were selected for the initial

analyses. One participant was found to have incomplete cognitive performance data, which

necessitated removal of the sibling pair from subsequent analyses. Thus, the final sample for

Hypothesis 1 consisted of 116 women (58 sibling pairs) discordant for use of COCPs. Sample

characteristics for the discordant sibling analysis are reported in Table 1. Most women attended

at least some college, and approximately 16% were students at the time of assessment. A small

majority of women were not currently married or cohabitating. The racial makeup of this sample

was relatively consistent with recent census findings (U.S. Census Bureau, 2016), with over a

quarter of participants identifying as non-white. Approximately half of all sibling pairs were monozygotic or dizygotic twins (28.8% and 23.7%, respectively), while the remaining 47.5% of women were classified as non-twin siblings. Group differences were assessed in an attempt to reveal potentially confounding effects of systematic differences in baseline characteristics, with no significant differences observed (see Table 1).

Table 1

Discordant Sibling Study: Sample Characteristics by Group

Variable	Group		t or χ^2	p-value
	Natural Cycle	COCP		
Age, M (SD)	29.8 (3.3)	29.5 (2.9)	.980	.331
Education, M (SD)	15.0 (1.9)	15.2 (1.7)	-1.130	.263
Relationship status, %			.000	1.000
Single	55.2	53.4		
In a relationship	43.1	46.6		
Not reported	1.7	0		
Race, %			.500	.500
White	72.4	75.8		
Black	19.0	17.2		
Asian/Pacific-Islander	3.4	3.4		
More than one	1.7	1.7		
Unknown/Not reported	3.4	1.7		
Nonverbal IQ, M (SD)	15.9 (4.9)	16.7 (4.7)	-1.213	.230
Verbal IQ, M (SD)	114.9 (9.1)	116.1 (8.2)	-1.599	.116

Note. $N = 116$, with equal groups of $n = 58$. Variables are presented as means and standard deviations or percentages. Nonverbal IQ score is *number correct* (out of 24) on the Penn Progressive Matrices Test (range: 6-24). Verbal IQ is *age-unadjusted standard score* on the NIH Toolbox Picture Vocabulary Test (range: 93-143). Paired-samples t tests ($df = 57$) were performed on continuous variables and McNemar tests ($df = 1$) were performed on nominal variables; to facilitate analysis, race was collapsed into a dichotomous variable of white/non-white.

Table 2

Discordant Sibling Study: Cognitive Performance by Group

| Task | Group | | t | p-value | Cohen's d |
	Natural Cycle	COCP			
Visuospatial Function	14.47 (4.68)	13.28 (4.43)	1.84	.071	.487
Verbal Memory	35.57 (2.97)	35.67 (2.40)	.072	.943	-.019

Note. Data presented in format of Mean (SD). Visuospatial Function is *number correct* (out of 40) on the Penn Word Memory Test (range: 27-40). Verbal Memory is *number correct* (out of 24) on the Variable Short Penn Line Orientation Test (range: 3-24). Analyses were conducted via paired-samples *t* tests (*df* = 57).

Data Analyses

We used a multivariate approach to examine the impact of COCPs on tasks of verbal memory and visuospatial processing. A paired-samples Hotelling's T^2 test was chosen so that we could examine effects of related dependent variables (DVs) while minimizing the risk of Type I error. Our independent variable (IV) was COCP use (on/off), and we examined differences in cognitive performance across two DVs: verbal memory and visuospatial functioning. DVs were moderately correlated ($r = .378$, $p = .003$) and free from outliers. Shapiro-Wilk tests indicated that while visuospatial scores were normally distributed ($p = .220$), the distribution of verbal memory scores violated the normality assumption ($p < .001$). Visual inspection of the graphed data revealed a negatively skewed distribution. To examine skewness more thoroughly, we calculated *z* scores using the following equation: $Z_{Skewness} = \text{Skewness} / SE_{Skewness}$ (Ghasemi & Zahediasl, 2012). With Skewness = .719 and SE = .225, our calculated *z* score of 2.20 was found to significantly depart from normality at $p < .05$ (i.e., ± 1.96). Thus, to correct for the left skewed distribution, we transformed the data using a cubed transformation (DeCoster, 2001). Updated Shapiro-Wilk tests revealed that our transformed data no longer violated the normality assumption at $p < .01$ ($W = .949$, $p = .017$), which was further confirmed by re-calculated

$Z_{Skewness} = 0.81$. Distribution of the data shared a common variance-covariance matrix per Box's $M = 4.010$, $p = .269$. Thus, it was determined that the planned parametric analyses were appropriate and would proceed. The omnibus Hotelling's T^2 test failed to reveal a significant multivariate effect of COCP use on cognition, $T^2 = 52.808$, $F(2, 56) = 1.690$, $p = .194$, $\eta_p^2 = .057$. Although the omnibus test was not significant, we proceeded with our planned hypothesis-driven comparisons and conducted paired-samples t tests for both DVs (see Table 2 for summary of results). Despite our prediction that COCP-users would perform better than their naturally-cycling siblings on tasks of verbal memory and visuospatial functioning, neither analysis revealed significant differences. Of note, naturally-cycling participants outperformed COCP-users on the visuospatial task at a level that approached significance ($p = .071$), in an apparent reversal of our hypothesis. The relationship between cognitive variables and group membership is depicted in Figure 1.
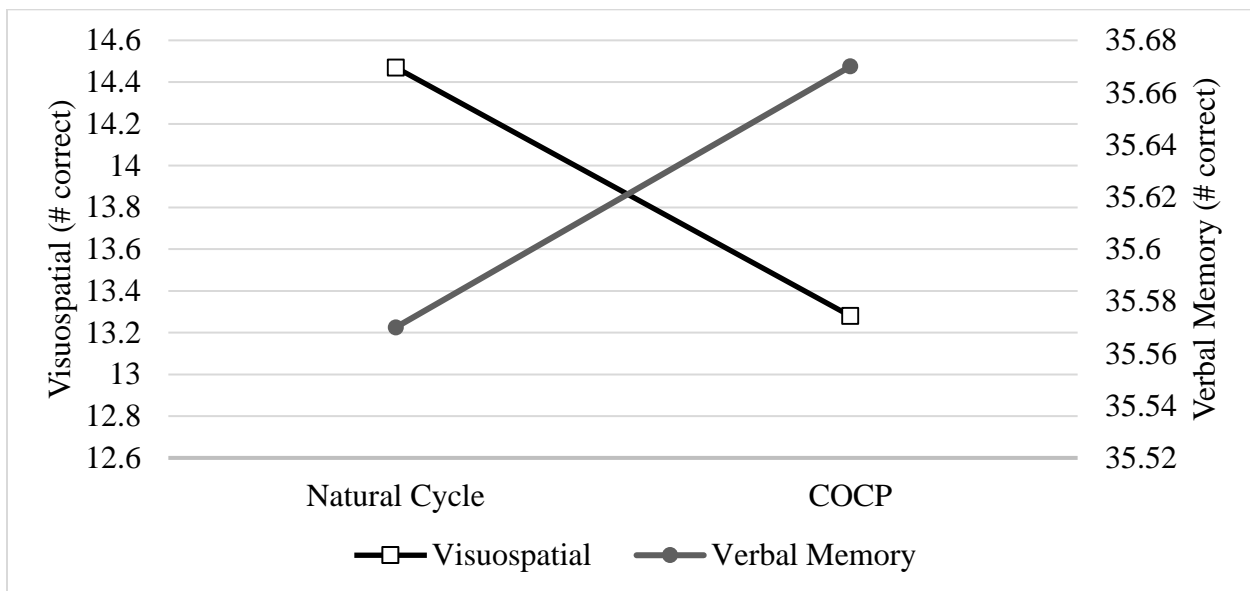

Figure 1. Cognitive performance in siblings discordant for COCP use.

Matched Groups Study

Sample Characteristics

The final sample for our second analysis consisted of 135 participants divided into three

groups with $n = 45$. Each group differed by hormone status (Follicular, Luteal, COCP), but were

matched on age, education level, and race. Overall, baseline sample characteristics were similar

to that of the discordant siblings analysis and can be further reviewed in Table 3. To rule out

potentially confounding effects of group differences, one-way ANOVAs were performed, with

none of the variables found to be significantly different (see Table 3).

Table 3

Matched Groups Study: Sample Characteristics by Group

Variable	Group			F	p-value
	Follicular	Luteal	COCP		
Age, M (SD)	29.8 (3.2)	30.0 (3.0)	30.0 (3.0)	.097	.907
Education, M (SD)	14.6 (1.9)	14.5 (2.0)	14.6 (1.9)	.051	.950
Relationship status, %				.087	.917
Single	46.7	51.1	51.1		
In a relationship	53.3	48.9	48.9		
Race, %				.000	1.000
White	77.8	77.8	77.8		
Black	20.0	20.0	20.0		
Unknown/Not reported	2.2	2.2	2.2		
Nonverbal IQ, M (SD)	15.4 (4.3)	14.9 (4.9)	15.4 (5.3)	.133	.876
Verbal IQ, M (SD)	113.6 (8.4)	113.9 (11.3)	114.2 (7.6)	.047	.954

Note. $N = 135$ with equal groups of $n = 45$. Variables are presented as means and standard deviations or percentages. Nonverbal IQ score is *number correct* (out of 24) on the Penn Progressive Matrices Test (range: 6-23). Verbal IQ is *age-unadjusted standard score* on the NIH Toolbox Picture Vocabulary Test (range: 96-150). One-way ANOVAs were performed, $F(2, 55)$.

Table 4

Matched Groups Study: Cognitive Performance by Group

Task	Group			F	p-value	η^2
	Follicular	Luteal	COCP			
Visuospatial Function	14.13 (5.79)	13.44 (4.43)	12.24 (4.65)	1.920	.151	.028
Verbal Memory	36.80 (2.35)	35.38 (3.86)	34.96 (3.21)	3.871	.023	.055

Note. Data presented in format of Mean (SD). Visuospatial Function is *number correct* (out of 40) on the Penn Word Memory Test (range: 24-40). Verbal Memory is number correct (out of 24) on the Variable Short Penn Line Orientation Test (range: 1-22). Test statistic represents one-way ANOVAs, $F(2, 132)$ with effect size, η^2. After Bonferroni correction, significance at $p < .025$.

Data Analyses

In addition to assessing the impact of COCPs on task performance, we also sought to examine how different *phases* of the natural MC may affect cognition on verbal and visuospatial tasks. For this analysis, a one-way MANOVA was performed to compare participants at three levels of our IV (COCP, Follicular, Luteal) across our two DVs (verbal memory, visuospatial functioning). DVs were positively correlated ($r = .227$, $p = .008$). The assumption of homogeneity was not violated per a nonsignificant Box's $M = 11.27$, $p = .088$. As in the discordant pair analysis, Shapiro-Wilk tests revealed that the verbal memory distribution was in violation of the normality assumption (p < .001). Further examination revealed that the distribution was negatively skewed *(Z*$_{Skewness}$ = -4.75). Although we initially attempted to correct for normality by using a cubed transformation, this did not sufficiently adjust for skewness ($Z_{Skewness} = 2.58$) and we instead used a square root transformation on the inverted data; this seemed to appropriately manage the skewness of the distribution ($Z_{Skewness} = 1.421$) such that skewness was no longer significant. Despite this transformation, an updated Shapiro-Wilk test revealed the data likely continued to violate the assumption of normality at the level of $p < .05$ ($p = .003$). However, per Ghasemi and Zahediasl (2012), "…violation of the normality assumption

should not cause major problems" with "…large enough sample sizes (> 30 or 40)," implying that parametric procedures are still often appropriate even when distributions are not normally distributed (owing in part to the central limit theorem). Given the overall robustness of the MANOVA, our relatively large sample size, and our successful use of transformation to adjust for skewness, it was determined that our transformed distribution (with a Shapiro-Wilk p-value of .003) would be sufficient to allow for planned parametric analyses to proceed. The omnibus test statistic (Wilk's λ) is a measure of the variance not explained by differences in level of the IV (i.e., ratio of within-group variance to total variance). Results of the omnibus MANOVA were significant and indicative of a multivariate effect of group differences on cognitive performance, Wilks' $\lambda = .930$, $F(4, 264) = 2.554$, $p = .048$, $\eta_p^2 = .036$.

Given that our hypotheses involved planned comparisons, we proceeded with our *a priori* univariate analysis approach (i.e., one-way ANOVAs) to assess the main effect of group on each domain of cognitive functioning. To control for multiple comparisons and protect against inflated Type I error, we used a Bonferroni correction and halved the original alpha level of $p < .05$ such that an alpha level of $p < .025$ was required for significance (Abdi, 2007). Results of the ANOVA examining the main effect of group differences on verbal memory performance were statistically significant (see Table 4). In contrast to our predictions, however, women in the follicular phase had the highest verbal memory scores, followed by women in the luteal phase, and then finally COCP-users (see Figure 2).

Following the significant ANOVA, we were interested in pairwise comparisons to identify which group differences were driving the significant results for verbal memory. Analyses were conducted using Tukey HSD tests to control for Type I error resulting from multiple comparisons (see Table 5). Women in the follicular phase were found to have

significantly higher verbal memory scores than women using COCPs. We found no differences

in verbal memory performance between women in the luteal phase and those in the follicular

phase or using COCPs. For the one-way ANOVA examining group membership on visuospatial

functioning, differences in task performance were not significant (see Table 4 and Figure 2).

Accordingly, we found no effect of COCP use or MC phase on visuospatial task performance.
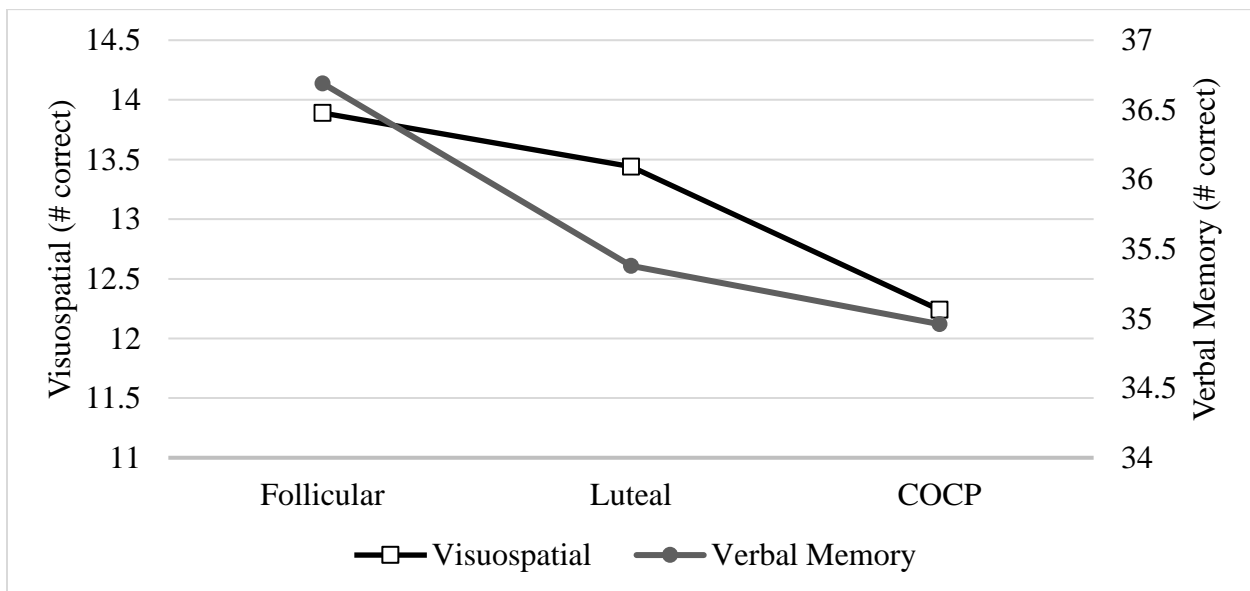

Figure 2. Cognitive performance in demographics-matched groups.

Table 5

Matched Groups Study: Pairwise Comparisons for Verbal Memory Performance

Comparisons	Mean Difference	Standard Error	*t*	*p*-value
Follicular vs. Luteal	1.422	.145	1.801	.167
Follicular vs. COCP	1.844	.145	3.018	.019
Luteal vs. COCP	0.422	.145	0.848	.633

Note. Post hoc comparisons performed using Tukey HSD tests with significance at *p* < .05. Mean Difference values based on estimated marginal means and represent *number correct*.

CHAPTER 4

DISCUSSION

The present study examined the hormonal effects of COCPs and MC variability on verbal memory and visuospatial functioning in healthy, premenopausal women. In our first analysis, we examined group differences in cognitive performance across pairs of COCP-discordant siblings. Despite our predictions that COCP-users would have higher scores than naturally-cycling women on both tasks , we found no group differences in performance. In our second set of analyses, we attempted to better understand hormonal variation across the MC by comparing women in distinct phases to COCP-users using demographics-matched groups. In this case, we found that naturally-cycling women during the follicular phase significantly outperformed women using COCPs on a verbal memory task. Notably, these findings are in direct contrast to our initial hypothesis predicting that COCP-users would have the highest verbal memory scores, followed by women in the luteal phase, and then finally women in the follicular phase. In terms of visuospatial functioning, despite our predictions that COCP-users would outperform follicular-phase women, who would then in turn outperform luteal-phase women, we failed to detect any significant differences between groups across studies.

There are a number of scenarios that may help explain why our findings did not match our hypotheses or the existing literature. One possibility is that our groups may have systematically differed on some variable that we did not account for. For example, the COCP-users performed overall below expectations. One explanation may be that women in the COCP group significantly differed along a particular psychological variable not measured. Although the

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literature is considerably mixed with regards to directionality of effects, a number of researchers have shown that COCP use can significantly impact emotional functioning. In 2002, Oinonen and Mazmanian conducted a review of the existing literature and determined that COCPs tend to have an overall net positive impact on mood; specifically, in comparison to naturally-cycling women, COCP-users experienced less *variability* in affect throughout the entire MC, as well as less *negative* affect during the "menstruation" week (i.e., withdrawal bleed) in particular. However, several studies that have been published since then have seemingly contradicted some of these findings. For instance, a double-blind, randomized control trial by Gingnell et al. (2013) suggested that in comparison to a placebo group, COCPs led to *increases* in depressed mood, mood swings, and fatigue. A recent study found that *positive affect* (measured daily throughout the cycle) was significantly higher for naturally-cycling women than for women using COCPs (Ocampo Rebollar, Menéndez Balaña, & Conde Pastor, 2017). Given these somewhat contradictory findings, additional research in this domain is warranted. Future studies should also explore individual differences in response to COCPs, as well as trait variability in affect and personality, as it relates to COCP use and cognition.

In addition to the COCP-users potentially performing below expectations, luteal-phase women also seemed to be at least trending worse than predicted, suggesting that it may be useful to consider variables that may have affected performance for this group, as well. One possibility is that women may have been experiencing (relatively common) physical and psychological symptoms during the mid-to-late luteal (i.e., premenstrual) phase, and that this may be negatively impacting performance. This seems particularly relevant given reports that 75-80% of reproductive-age women experience some type of physical (e.g., headaches, fatigue) and/or psychological (e.g., anxiety, depression, irritability) MC-related symptoms (Pearlstein & Steiner,

2008; Storck, 2012). Subsequent research has attempted to evaluate the potential impact that these common MC-related symptoms – both within the general population, as well as when they are often at their most severe, such as pre-menstrual syndrome (PMS) or pre-menstrual dysphoric disorder (PMDD) – may have on various domains of functioning. For instance, Keenan, Stern, Janowsky, and Pedersen (1992) reported that PMS symptoms were associated with verbal encoding difficulties, and Slyepchenko et al. (2017) found that women with moderate to severe PMS symptoms showed deficits on tasks of working memory and selective attention. Conversely, many other studies report that women with MC-associated symptoms and diagnoses show no significant differences on a variety of cognitive tasks (Man, MacMillan, Scott, & Young, 1999; Resnick, Perry, Parry, Mostofi, & Udell, 1998).

Verbal Memory

Although findings within the literature have historically been mixed, a general pattern has emerged with regards to verbal memory functioning and the MC such that women typically perform better during periods of high estrogen and progesterone (i.e., luteal phase) and worse during periods of low estrogen and progesterone (i.e., follicular phase; Hampson, 1990; Maki et al., 2002). Our findings from the matched groups analysis almost directly contradict this. Specifically, we found that verbal memory scores were significantly higher for women in the follicular phase (with low estrogen and progesterone) than for women in the luteal phase (with high estrogen and progesterone) or for women using COCPs. In contrast to the relatively direct relationship seen between estrogen and progesterone levels and verbal memory across the natural MC (Hampson, 1990; Maki et al., 2002), it appears that the association between verbal memory and COCP may be much more complex.

It is well established that COCPs normally reduce overall levels of estrogen and progesterone in comparison to the natural MC, and that levels remain relatively stable throughout an entire pill cycle. Despite this reduction in estrogen and progesterone, however, COCP use has actually been linked to *higher* scores on verbal memory tasks (Gogos, 2013; Mordecai et al., 2008). This apparent discrepancy is theorized to relate to the decline in testosterone associated with COCPs, with an apparently inverse relationship at play – i.e., lower testosterone levels are associated with higher verbal memory scores, and vice versa (Mordecai et al., 2008; Phillips & Sherwin, 1992). Based on the prior literature, we hypothesized that there would be an additive effect of COCPs on verbal memory resulting from the COCP-related decline in testosterone, that would act *above and beyond* the effect caused by the corresponding COCP-related reduction in estrogen and progesterone. Despite our prediction, we found no evidence that verbal memory performance improved for women using COCPs; this may indicate that the expected COCP-related changes in testosterone failed to have any effect. However, it does appear that our findings regarding verbal memory performance in COCP-users may fit with the more straightforward (i.e., testosterone-free) model describing a relatively direct, positive relationship between estrogen/progesterone and verbal memory in naturally-cycling women. Specifically, we found that women using COCPs (during which estrogen and progesterone are at their lowest) had poorer verbal memory scores than naturally-cycling women in their follicular phase (during which estrogen and progesterone are known to be higher than in COCPs, but lower than in the luteal phase). These findings suggest that the mechanism of COCPs as they relate to the proposed effects of testosterone on verbal memory performance warrants further examination.

There are several potential reasons why our results are not consistent with our predictions and failed to replicate findings from the literature. Examining the characteristics of the measure

we used to assess verbal memory (i.e., Penn Word Memory Test) is a relevant first step. Although the task was chosen for a number of reasons (i.e., computerized format, availability for use in large-scale research), it was also selected for its psychometric properties (Van Essen et al., 2013). However, in the present study, we found that one of our sample distributions was negatively skewed, with participants performing above expectations. This raises concerns about whether this particular task was able to sufficiently differentiate performance at the upper end of the distribution (i.e., ceiling effects being present). A similar consideration relates to the components of verbal memory that our measure was able to assess; our task was limited to the relatively uncomplicated recognition memory domain and lacked a more difficult recall component. This may have hindered our ability to identify differences in performance on a more fine-grained level (e.g., encoding vs. retrieval-based performance). In the future, researchers may benefit from choosing a verbal memory task with a higher ceiling, a larger range of scores, and multiple dimensions (i.e., repeated learning trials, free recall, cued recall, and recognition).

Visuospatial Functioning

Like much of the existing research looking at hormonal effects on verbal memory, the literature examining hormonal effects on visuospatial functioning is also quite mixed. However, general patterns suggest that low levels of estrogen and progesterone are associated with improved visuospatial functioning, while high levels of estrogen and progesterone are linked to poorer performance on visuospatial tasks (Beltz et al., 2015; Cicinelli et al., 2011; Wright & Badia, 1999). In the present study, we failed to detect any significant differences for either design (i.e., discordant siblings and matched groups). There are several potential explanations for our lack of significant findings. One possibility concerns the measure itself (i.e., Variable Short

Penn Line Orientation Test), which assesses visuospatial functioning via a line orientation task. Given the relative simplicity of the measure, perhaps it lacked the sensitivity to detect hormone-related changes in *higher-level* visuospatial processing and integration functions. Although several studies have attempted to look at similarly straight-forward visuospatial tasks with mixed success, it is possible that a more complex visuospatial task (e.g., spatial problem-solving, mental manipulation) that requires more complex cognitive abilities may be more appropriate for detecting these hypothesized differences.

Another potentially confounding factor pertains to the variability in COCP compositions. There are many different "brands" of COCPs available, and although most contain the same synthetic estrogen compound (i.e., ethinyl estradiol), COCPs often differ on the progestin used (Amy & Tripathi, 2009; Batur, Elder, & Mayer, 2003). Further, progestins in COCPs can generally be divided into four generations; first-generation progestins are no longer used in contraceptives, but current COCPs may contain progestins from the second, third, or fourth generations (Benagiano, Carrara, & Filippi, 2009; Glasier, 2006). Broadly, second- and third-generation progestins (e.g., levonorgestrel, norgestimate) are testosterone-derived and demonstrate more androgenic activity, while the "newer" fourth-generation progestins (e.g., drospirenone) are spironolactone-based and have anti-androgenic properties (Batur et al., 2003; Rowlands, 2003). Overall, emerging literature seems to suggest that cognitive performance patterns likely parallel their opposing androgenic-based actions, with women on fourth-generation COCPs doing worse than women on second- or third-generation brands for a mental rotation task (Griksiene & Ruksenas, 2011). Because the archival nature of the present study prevented us from obtaining information about the specific brands of COCP used, the degree to which this variability potentially impacted task performance is unknown. In sum, researchers

have only begun studying how these progestin-related variations in COCP composition differentially affect hormones and task performance, with findings still inconclusive; it will be important for future studies to better account for progestin types and further explore what other domains may be impacted.

Limitations and Future Directions

Several potential limitations of the current study – as well as relevant suggestions for future research – have already been discussed throughout this manuscript (e.g., imprecision of measures, variability in COCP brands, impact of mood or affect). However, there are a number of additional factors that may have affected our analyses and can be addressed further. For example, because we had very little information on participant demographics beyond the basics (e.g., education, age, race), it is possible that there were group differences in other variables that could have had important effects on our analyses. For instance, one potentially relevant domain that we were unable to address is sexual history, including information about sexual orientation and sexual behavior, both of which may have had meaningful influence on the relationship between COCPs and task performance. On a similar note, although there was an established screening process used to determine eligibility for the present study, most of the decision rules relied on participant self-report in response to brief and sometimes vague questions (e.g., "*Does the participant report having regular menstrual cycles?*"). Consequently, only the few women who mentioned situations such as a recent pregnancy or ongoing breastfeeding during an open-ended question (that was not specifically asking about these topics) were able to be identified and ultimately excluded from the study. Other potentially relevant information that was unavailable to us and may have impacted our findings includes: number of previous pregnancies,

length of time on current COCP, and history of prior COCP use (along with reason for quitting, if relevant). Thus, we can only speculate about how elements such as these may have meaningfully impacted hormone levels, which in turn may have confounded our findings. On a related note, another potential limitation pertains to the reliance on self-report data for details about MC and COCP status in particular. Although there is precedent in prior literature, it is preferred that hormonal assay be used to confirm group membership (e.g., MC phase) or exclude those women with atypical hormonal patterns that are likely to muddle our data. As a way to prevent this from unduly affecting the current study, we attempted to thwart this concern by adopting relatively strict decision rules for study inclusion and group membership. In addition, we chose to try to mitigate some of the individual variance through our selection of relatively robust study designs (i.e., discordant sibling pairs and demographically-matched groups).

Conclusions

This study builds upon the existing literature examining hormonal effects of COCPs and MC variability on cognition via larger sample sizes than typically seen in the literature, as well as relatively robust study designs (i.e., demographics-matched groups and discordant sibling pairs). Although our findings were not consistent with our predictions or much of the prior research, this study does demonstrate that differences across the MC and with use of COCPs may significantly alter verbal memory task performance. Further, it also brings up important questions for future research, including whether mood or affective variables may play a role in task performance (either positively or negatively), and whether specific types of COCPs (or even other types of birth control methods such as hormonal IUDs, implants, injections, etc.) may differentially affect hormones and subsequent cognitive performance. Given the continued

inconsistency in the literature, additional research is needed to better examine how COCPs affect cognition, particularly with their continued widespread usage worldwide. Overall, the discrepant findings in the present study relative to prior research underscores the need for future research on cognitive performance on COCPs and across the MC, particularly if their study designs and cognitive assessment choices address some of the limitations present in much of the literature.

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