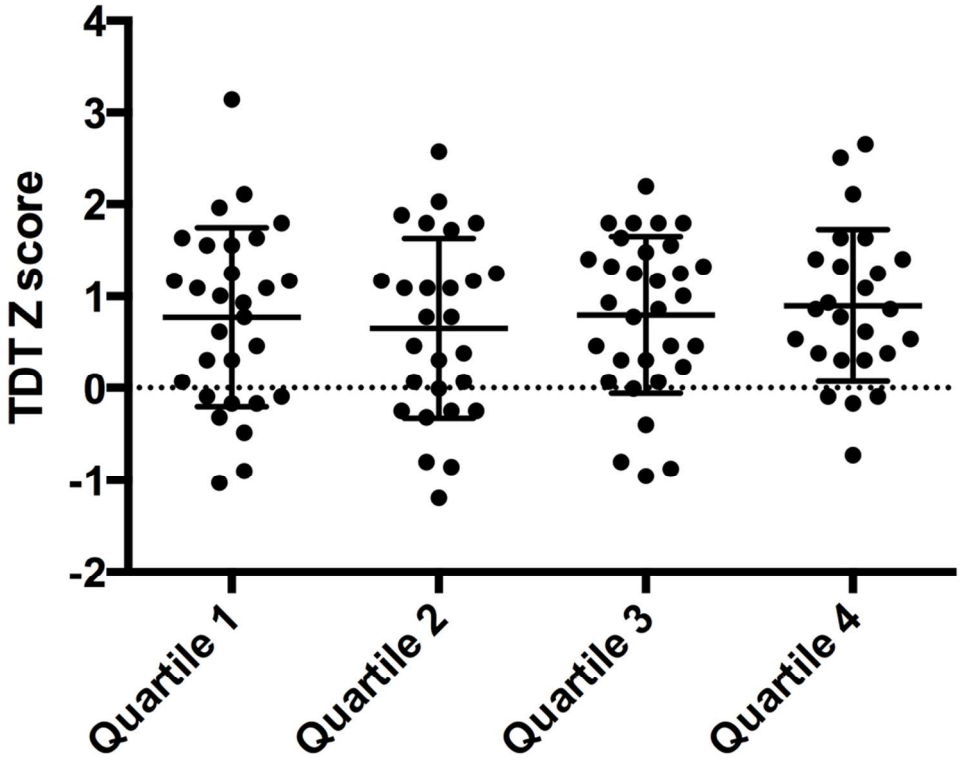
EJN European Journal of Neuroscience FENS Federation of European Neuroscience Societies

Menstrual cycle and the visual temporal discrimination threshold

Journal:	*European Journal of Neuroscience*
Manuscript ID	EJN-2016-02-23442
Manuscript Type:	Short Communication
Date Submitted by the Author:	21-Feb-2016
Key Words:	Temporal discrimination, Menstrual quartile, Oestrogen, Sexual dimorphism

Complete List of Authors:
McGovern, Eavan; St. Vincent's University Hospital, Department of Neurology ; University College Dublin, School of Medicine; University of Dublin Trinity College, Trinity Centre for Bioengineering
O Connor, Emer; St Vincents University Hospital, Department of Neurology
Beiser, Ines; St. Vincent's University Hospital, Department of Neurology; University College Dublin, School of Medicine
Williams, Laura; St. Vincent's University Hospital, Department of Neurology; University College Dublin, School of Medicine
Butler, John; University of Dublin Trinity College, School Centre for Bioengineering; University of Dublin Trinity College, School of Engineering; Dublin Institute of Technology, School of Mathematical Sciences
Quinlivan, Brendan; University of Dublin Trinity College, Trinity Centre for Bioengineering; University of Dublin Trinity College, School of Engineering
Reilly, Richard; University of Dublin Trinity College, Trinity Centre for Bioengineering; Trinity College Dublin, School of Engineering ; University of Dublin Trinity College, School of Medicine
O'Riordan, Sean; St. Vincent's University Hospital, Department of Neurology; University College Dublin, School of Medicine
Hutchinson, Michael; St. Vincent's University Hospital, Department of Neurology; University College Dublin, School of Medicine



Temporal discrimination threshold (TDT) Z-scores in 14 female healthy controls during each of eight quartiles of two consecutive menstrual cycles. Quartile One is the quartile of interest. The filled circles in Quartile 1 represents the TDT from this quartile during two consecutive menstrual cycles in controls ; similarly Quartile 2 represents TDT Z-scores from two consecutive menstrual cycles. Figure 1
92x74mm (300 x 300 DPI)

European Journal of Neuroscience: Short Communication

Title: Menstrual cycle and the visual temporal discrimination threshold

Authors: Eavan M Mc Govern[1,2, 3*], Emer O'Connor[1,], Ines Beiser[1,2], Laura W Williams[1,2], John B Butler[3,4, 6], Brendan Quinlivan[3,4] , Richard B Reilly[3,4, 5] Sean O'Riordan[1,2], Michael Hutchinson[1,2]

1. Department of Neurology, St Vincent's University Hospital Dublin, Ireland
2. School of Medicine & Medical Science, University College Dublin, Ireland
3. Trinity Centre for Bioengineering, Trinity College, The University of Dublin, Dublin, Ireland
4. School of Engineering, Trinity College, The University of Dublin, Dublin Ireland
5. School of Medicine, Trinity College, The University of Dublin, Dublin, Ireland
6. School of Mathematical Sciences, Dublin Institute of Technology, Kevin St, Dublin Ireland.

* Corresponding Author: Email: eavanmcgov@hotmail.com

Telephone: 01-2774033 Fax: 01 - 2213842

Running Header: Temporal discrimination threshold and the menstrual cycle

Journal section: Short communications

Word count: Abstract:168 Introduction: 209 Manuscript: 2738

Page count: 14. Figures: 1. Tables: 1

Keywords: Temporal discrimination, menstrual quartile, oestrogen, sexual dimorphism

Abstract: The temporal discrimination threshold represents the point at which an individual determines two sequential sensory stimuli to be asynchronous. Women, aged 20–40 years, have faster temporal discrimination than men. However, with age, temporal discrimination in women worsens at a faster rate than in men so that, after 40 years of age women have slower temporal discrimination than men; it is not clear whether this is an hormonal or chromosomal effect. The aim of this study was to examine visual temporal discrimination thresholds at weekly intervals during two consecutive menstrual cycles in 14 healthy female volunteers to determine whether physiological changes in oestrogen or progesterone affected temporal discrimination. There was no evidence of any significant differences in weekly temporal discrimination thresholds during the menstrual cycles and no significant correlation with the menstrual cycle stage. This observed stability of temporal discrimination during cyclical physiological hormonal change supports the hypothesis that the age-related worsening of temporal discrimination in women is more probably chromosomally determined and not due to menopausal-hormonal change.

Introduction: The temporal discrimination threshold (TDT) represents the point at which an individual determines two sequential sensory stimuli to be asynchronous [normally about 30-50ms] (Kimmich *et al.*, 2011). Mean TDT scores increase with age in a sexually dimorphic pattern; after 40 years of age women's TDT scores deteriorate at approximately three times the rate of their male peers (Williams *et al.*, 2015). Before this age, women have faster temporal discrimination than men. It is unclear as to whether this age-related sexual dimorphism in temporal discrimination is hormonally or sex-chromosome mediated. Menopause is the female transition into reproductive senescence and is accompanied by reduced oestradiol levels and a depletion of ovarian follicles (Speroff *et al.*, 1999). The mean age of menopause occurs at 51.3 years, ten years after this observed decline in female TDT score (McKinlay *et al.*, 1992). The menstrual cycle is a physiological, monthly occurrence regulated by cyclical-hormonal changes (Mihm *et al.*, 2011). This cycle provides an opportunity to examine TDT scores during monthly fluctuations in sex-hormone levels. The purpose of this study, therefore, was to examine weekly TDT scores during two consecutive menstrual cycles and to correlate these scores with individual menstrual cycle stage. We hypothesised that TDT scores would be unaffected by monthly fluctuations in sex-hormone levels.

Participants and Methods

Participants: Fifteen healthy, nulliparous female volunteers were recruited to the study; all were medical students. Exclusion criteria were current or previous use of hormonal contraception and a history of neurological disease. Fourteen participants completed the study. Study participants ranged in age from 19 - 23 years with a mean age of 19 years (SD: ± 0.99 years). In accordance with the Declaration of Helsinki,

written informed consent was obtained from all participants. The study was approved by the Medical Research Ethics Committee at St Vincent's University Hospital.

Methods: Visual TDT testing was carried out as described previously (Molloy et al.,2014). Briefly, testing was carried out in a single session in a dimly-lit room. Testing was performed using a portable-TDT headset, which ensured participants had a consistent position relative to the stimulus. TDT was measured using paired-visual stimuli presented to the participant in a single session. Two white LED lights were positioned seven degrees into the participant's peripheral visual field on the side of the body being tested. The participant was instructed to fixate on a centrally presented red LED. Initially, the pair of white LEDs flashed synchronously and were progressively illuminate but out of sync in 5-ms steps. The trial ended when the participants reported on three consecutive occasions that the pairs of white LEDs flashed asynchronously. The first of three asynchronous responses was taken as the TDT for that trial. This procedure was repeated four times on the left and right side of the body resulting in a total of eight runs per participant. The order was randomised to minimise practice effects. TDT testing was performed weekly for nine consecutive weeks. The first week of recording was discarded to allow for practice effect.

Menstrual cycle diary. Participants were asked to keep a menstrual diary. The onset and duration of the menstrual flow and the total-cycle length was noted. The first day of the menstrual flow corresponded to day one of the menstrual cycle. The textbook menstrual cycle is 28 days (interval between day one of menses to the next menses) (Speroff *et al.*, 1999). However, menstrual cycle is highly variable between women and ranges from 24 to 35 days (Mihm *et al.*, 2011). Due to this cycle-length variability, we expressed an individual's menstrual cycle as a quartile of the total-cycle length. Typically, the first four days of menstruation (or the first quartile of a

cycle with variable-cycle length) corresponds to the early-follicular phase. It is associated with low oestradiol levels, sometimes within post-menopausal levels, low progesterone levels and an elevation in FSH (Sundström Poromaa & Gingnell, 2014). This phase was selected as our quartile of interest given its hormonal similarities to menopause (Ycaza Herrera & Mather, 2015). Participants were instructed to keep a menstrual diary throughout the testing period and one month either side of this time. Participants entered the study on the same date, thus their testing commenced at varying stages of their menstrual cycle. Four participants commenced the study in the first quartile, five in the second quartile, four in the third quartile and one in the fourth quartile.

Statistical Analysis

Visual Temporal Discrimination Threshold: The median of four trials on each side was averaged to obtain a summary visual TDT score. All TDT results (in milliseconds) were converted to standardised Z-scores to enable comparison of individual results using the formula:

$$\text{Z – Score} = \frac{\text{Actual TDT - Age-related control mean TDT}}{\text{Age-related control standard deviation}}$$

The control mean and standard deviation used in the formula depends on the age of the participant; the age-related control mean TDT (and standard deviation) values have been published (Molloy *et al.*, 2014).

Variance analysis: A one-way repeated measure ANOVA was used to compare any variance in mean TDT Z-score across each menstrual quartile (quartile 1 – 4) and each week of testing (week 1 – 9). The independent variables used for this analysis were menstrual quartile and week of testing; the dependent variable was the TDT Z-

score. Given the small sample size, a non-parametric Kruskall-Wallis test was also applied to our data. All ANOVAs were carried out twice, with and without inclusion of the first week of testing. The first week of recording was discarded to allow for practice effect and this did not alter the significance of the results.

Correlation analysis: Correlation analysis was carried out to explore the relationship between TDT Z-score and menstrual quartile. The menstrual quartile was expressed as a dichotomous variable (1 = quartile of interest and 2 = other quartiles). The first quartile was the quartile of interest and it corresponded to the early-follicular phase. Quartiles two to four accounted for the other quartiles. A coefficient of determination was calculated to assess how much variance the two variables shared. As with the ANOVA, all correlation tests were carried out twice, once with inclusion of week one of testing and once without. The first week of recording was discarded to allow for practice effect and this did not alter the significance of the results.

Results

Fourteen participants completed the study. The mean age of study participants was 19 years (*SD* ± 0.99 years). Menstrual cycle length varied between participants (mean 32 days, *SD* ± 6.93 days). The mean TDT Z-score was 0.78 (*SD* 0.90). A one-way repeated measure ANOVA was conducted to compare TDT Z-scores at Time 1 (quartile 1), Time 2 (quartile 2), Time 3 (quartile 3) and Time 4 (quartile 4). The means and standard deviations are presented in Table 1. There was no significant effect for menstrual quartile on TDT Z-score [Wilks' Lambda = 0.95, F(3, 55) = 0.376, p = 0.771, multivariate partial eta squared = 0.049]. Mean TDT Z-score did not vary across menstrual cycle quartiles (Figure 1). A one-way repeated measures ANOVA was repeated to compare TDT Z-scores across each week of testing. There

was no significant effect for week of testing on TDT Z-score [Wilks' Lambda = 0.36, F(7, 111) = 1.75, p = 0.239, multivariate partial eta squared = 0.636]. We applied a non-parametric Kruskall-Wallis Test to our data. This confirmed that there was no statistically significant difference in mean TDT scores across menstrual cycle stages of testing (chi-square 0.650, *df* 3, $p < 0.885$). The relationship between the TDT Z-score and menstrual quartile (1 = quartile of interest, 2 = other quartile) was investigated using a point bisserial correlation. Preliminary analyses was performed to ensure no violation of the assumption of normality, linearity and homoscedasticity. There was a lack of correlation observed between the two variables. [r_{pb}=0.004, n =112, p <0.968). The dichotomous variable, menstrual quartile, demonstrated a lack of variability in mean TDT score. The coefficient of determination for the two variables (TDT Z-score and menstrual quartile) was low (r^2 = 0.0000016), therefore demonstrating a lack of overlap between the two variables.

Discussion

In this study we have observed no statistical significant variation in weekly visual TDT Z-scores during two consecutive menstrual cycles. Importantly our results demonstrate no significant difference in visual TDT Z-scores during our quartile of interest compared with other quartiles. Visual TDT Z-score was independent of menstrual quartile and its mean did not vary between menstrual cycle quartiles. Our findings suggest that the visual TDT Z-score is unaffected by the cyclical-hormonal fluctuations during two consecutive menstrual cycles. A previous study demonstrated that mean temporal discrimination in women increases (worsens) significantly with age when compared with men (Williams *et al.*, 2015). After the age of 40 years temporal discrimination in women deteriorates at almost three-times the rate of their

age-matched male peers. The lack of hormonal influence on visual TDT Z-scores observed in our study is important when considering the pathogenesis of this sexually-dimorphic, age-related deterioration in temporal discrimination. Menopause is defined as the permanent cessation of menstruation which reflects cessation of ovulation due to the loss of ovarian activity (Speroff *et al.*, 1999). This phase is accompanied by high gonadotropin, reduced oestrogen and progesterone levels and a depletion of ovarian follicles (Burger *et al.*, 1995). The mean age of menopause is 50 - 51 years in women from industrialised countries (Gold, 2011). This is 10 years after the sexually-dimorphic age-related decline observed in temporal discrimination (Williams *et al.*, 2015). Oestradiol, the most biologically active form of oestrogen, remains within normal range [or slightly elevated] in the perimenopausal transition [mean age 45.1 years] (Rannevik *et al.*, 2008). Therefore, we propose that this age-related deterioration in women's visual TDT scores is unlikely to be related to menopausal hormonal change. The findings from our study, that the visual TDT Z-score is unaffected by cyclical-hormonal fluctuations, further corroborates this hypothesis. Our results lead us to consider an alternative explanation for this age-related sexual dimorphic deterioration in temporal discrimination. Menstrual cycle studies have, for the most part, failed to demonstrate an influence of menstrual cycle on cognitive function (Sundström Poromaa & Gingnell, 2014). Previous studies have however demonstrated that hormonal fluctuations in oestrogen levels can influence processing of emotional information (Pompili *et al.*, 2016). This may be due, in part, to the expression of oestrogen receptors [ERα, ERβ] in pertinent brain regions such as the hippocampal formation which encodes new declarative memories and the amygdala which is particularly involved in encoding arousing stimuli (McEwen & Alves, 1999) (Frick, 2013). Although evidence is limited, it is suggested that emotion

recognition, consolidation of emotional memory and fear extinction may be modulated by the menstrual cycle. Unlike emotional memory, the TDT represents an individual's ability to detect environmental change. The midbrain network for covert attentional network plays a key role in the detection of salient stimuli in our environment (Fecteau & Munoz, 2006) (Redgrave *et al.*, 2010). We consider an abnormal TDT is due to a disorder of this network for covert attentional orienting caused by reduced gamma-aminobutyric acid (GABA) inhibition (Hutchinson *et al.*, 2014). A magnetic resonance spectroscopy study in healthy participants demonstrated a sexually-dimorphic, age-related decline in frontal GABA (Gao *et al.*, 2013). Above the age of 40 years women exhibited a more marked reduction in frontal GABA compared with men. This decline in frontal GABA mirrors the age-related deterioration in TDT scores observed in women. We suggest that the sexually-dimorphic differences observed in TDT above the age of 40 results from age- and sex-chromosome-related alterations in GABA levels and are not due to hormonal influences (Butler *et al.*, 2015). Our study, which demonstrates a lack of variation in the visual TDT Z-scores during cyclical hormonal fluctuations, supports our hypothesis, that the age-related deterioration in temporal discrimination is unrelated to changes in hormonal status and is more probably related to sex-chromosome effects.

Acknowledgements

This research was supported by grants from Dystonia Ireland, the Irish Institute for Clinical Neuroscience and the Foundation for Dystonia Research (Belgium) and the Trinity Centre for Bioengineering.

References:

Burger, H.G., Dudley, E.C., Hopper, J.L., Shelley, J.M., Green, A., Smith, A., Dennerstein, L., & Morse, C. (1995) The endocrinology of the menopausal transition: a cross-sectional study of a population-based sample. *J. Clin. Endocrinol. Metab.*, **80**, 3537–3545.

Butler, J.S., Beiser, I.M., Williams, L., McGovern, E., Molloy, F., Lynch, T., Healy, D.G., Moore, H., Walsh, R., Reilly, R.B., O'Riordan, S., Walsh, C., & Hutchinson, M. (2015) Age-Related Sexual Dimorphism in Temporal Discrimination and in Adult-Onset Dystonia Suggests GABAergic Mechanisms. *Front. Neurol.*, **6**, 258.

Fecteau, J.H. & Munoz, D.P. (2006) Salience, relevance, and firing: a priority map for target selection. *Trends Cogn. Sci.*, **10**, 382–390.

Frick, K.M. (2013) Epigenetics, oestradiol and hippocampal memory consolidation. *J. Neuroendocrinol.*, **25**, 1151–1162.

Gao, F., Edden, R.A.E., Li, M., Puts, N.A.J., Wang, G., Liu, C., Zhao, B., Wang, H., Bai, X., Zhao, C., Wang, X., & Barker, P.B. (2013) Edited magnetic resonance spectroscopy detects an age-related decline in brain GABA levels. *Neuroimage*, **78**, 75–82.

Gold, E.B. (2011) The Timing of the Age at Which Natural Menopause Occurs. *Obstet. Gynecol. Clin. North Am.*, **38**, 425–440.

Hutchinson, M., Isa, T., Molloy, A., Kimmich, O., Williams, L., Molloy, F., Moore, H., Healy, D.G., Lynch, T., Walsh, C., Butler, J., Reilly, R.B., Walsh, R., & O'Riordan, S. (2014) Cervical dystonia: A disorder of the midbrain network for covert attentional orienting. *Front. Neurol.*, **5 APR**, 1–10.

Kimmich, O., Bradley, D., Whelan, R., Mulrooney, N., Reilly, R.B., Hutchinson, S., O'Riordan, S., & Hutchinson, M. (2011) Sporadic adult onset primary torsion dystonia is a genetic disorder by the temporal discrimination test. *Brain*, **134**, 2656–2663.

McEwen, B.S. & Alves, S.E. (1999) Estrogen actions in the nervous system. *Endocr. Rev.*, **20**, 279–307.

McKinlay, S.M., Brambilla, D.J., & Posner, J.G. (1992) The normal menopause transition. *Maturitas*, **14**, 103–115.

Mihm, M., Gangooly, S., & Muttukrishna, S. (2011) The normal menstrual cycle in

women. *Anim. Reprod. Sci.*, **124**, 229–236.

Molloy, A., Kimmich, O., Williams, L., Quinlivan, B., Dabacan, A., Fanning, A., Butler, J.S., O'Riordan, S., Reilly, R.B., & Hutchinson, M. (2014) A headset method for measuring the visual temporal discrimination threshold in cervical dystonia. *Tremor Other Hyperkinet. Mov. (N. Y).*, **4**, 249.

Pompili, A., Arnone, B., Amico, M.D., Federico, P., & Gasbarri, A. (2016) Psychoneuroendocrinology Evidence of estrogen modulation on memory processes for emotional content in healthy young women. *Psychoneuroendocrinology*, **65**, 94–101.

Rannevik, G., Jeppsson, S., Johnell, O., Bjerre, B., Laurell-Borulf, Y., & Svanberg, L. (2008) A longitudinal study of the perimenopausal transition: altered profiles of steroid and pituitary hormones, SHBG and bone mineral density. *Maturitas*, **61**, 67–77.

Redgrave, P., Coizet, V., Comoli, E., McHaffie, J.G., Leriche, M., Vautrelle, N., Hayes, L.M., & Overton, P. (2010) Interactions between the Midbrain Superior Colliculus and the Basal Ganglia. *Front. Neuroanat.*, **4**, 1–8.

Speroff, L., Glass, R.H., & Kase, N.G. (1999) *Clinical Gynecologic Endocrinology and Infertility 6th Edition*. Lippincott Williams & Wilkins.

Sundström Poromaa, I. & Gingnell, M. (2014) Menstrual cycle influence on cognitive function and emotion processing-from a reproductive perspective. *Front. Neurosci.*, **8**, 380.

Williams, L.J., Butler, J.S., Molloy, A., McGovern, E., Beiser, I., Kimmich, O., Quinlivan, B., O'Riordan, S., Hutchinson, M., & Reilly, R.B. (2015) Young Women do it Better: Sexual Dimorphism in Temporal Discrimination. *Front. Neurol.*, **6**, 1–8.

Ycaza Herrera, A. & Mather, M. (2015) Actions and interactions of estradiol and glucocorticoids in cognition and the brain: Implications for aging women. *Neurosci. Biobehav. Rev.*, **55**, 36–52.

Table

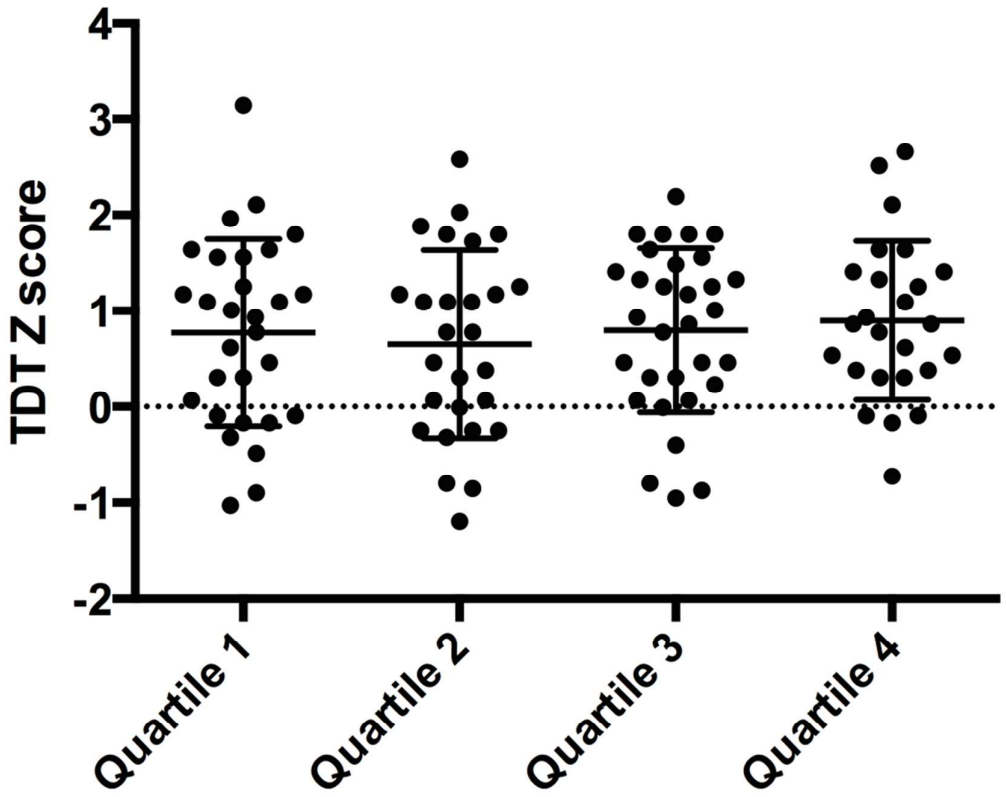
	Quartile 1	Quartile 2	Quartile 3	Quartile 4
Mean TDT (SD) (milliseconds)	43.7 (15.5)	41.8 (15.5)	43.9 (13.6)	45.7 (13.2)
Mean TDT Z-score	0.77	0.65	0.79	0.89

Legends for Figure & Table

Figure: Temporal discrimination threshold (TDT) Z-scores in 14 female healthy controls during each of eight quartiles of two consecutive menstrual cycles. Quartile One is the quartile of interest. The filled circles in Quartile 1 represents the TDT from this quartile during two consecutive menstrual cycles in controls ; similarly Quartile 2 represents TDT Z-scores from two consecutive menstrual cycles.

Table: The visual temporal discrimination thresholds (TDT) (in milliseconds and Z-scores) in 14 healthy control female participants recorded at weekly intervals over the eight weeks of two menstrual cycles. Quartile one results were obtained in the first quartile of the menstrual cycle.

Figure


TDT Z score
4
3
2
1
0
-1
-2
Quartile 1
Quartile 2
Quartile 3
Quartile 4