Prevalence of Female Reproductive Dysfunctions in Waged Labourers Employed at Rice Fields of West Bengal

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Abstract: A longitudinal study was carried out on 80 females, 40 engaged in waged labour in rice fields (WAL) and 40 as control (CH) of same socioeconomic status. The WAL females showed menstrual disorders that were supported by their hormonal patterns. Moreover the females had high stress hormone levels that may influence their pituitary ovarian axis Further the stress biomarker was significantly correlated to durations of sleep and physical labour. We account the higher physical labour and less sleep duration as the cause of this stress leading to female reproductive morbidity.

1 Introduction

In third world countries like India, agriculture contributes a lump sum share of the State's economy. In rural India, women show a greater involvement in doing agricultural pasture activities apart from their normal household schedule. They sub serve a vast proportion about 55% to 66% of the total field labour (Yadav, 2009).Compared to a pair of bullocks with an average labour for 1064 hours and a man who spent 1212 hours in the field, a woman shows 3485 hours of labour in a year on a one hectare farm (Shiva FAO, 1991; Shiva *et al*., 2003).

Mainly rural women are engaged in agricultural activities in three different ways depending on the socio-economic status of their family and regional factors. They are work as: (i) Paid Labourers (ii) Cultivator doing labour on their own land and (iii) Managers of certain aspects of agricultural production by way of labour supervision and the participation in post harvest operations.

With the advent of new technologies in crop yield, the human labour investment is subsiding leading to emigration of males from this profession to other better waged jobs and thus the lacuna is filled by the rural females disposing the increase in the involvement of females in such activities. Further farmers and land owners prefer females in such jobs so that they can squeeze their cost of modern technologies in production from the share of wages for these female labourers. This sector of occupational community has to perform manoeuvre like sowing of seeds, transportation of sapling, winnowing, storage of grain etc. In addition these sectors of population are also involved in various forms of processing and marketing of agricultural produce (Aggarwal, 2003).

The rural female are at a stake of severe work load as they are to perform several extension activities like house hold management, cooking, nursing of young and old family members, cattle management, fodder arrangement and milking. This may be a venture for their health hazards in addition to their poor income and improper nutritional status.
Evidences in the available literature suggest that females engaged in this population had anemia and nutritional insufficiency (Uday Lakshmi and Babitha, 2014) but female reproductive illness amongst these populations has not been highlighted. Further occupational exposure to pesticides and insecticides in females has proved to have adverse reproductive outcome (Ibrahim *et al*., 2011; Bretveld *et al*., 2006) but reproductive outcomes in agricultural laboured

females engaged in sowing, plantation, carrying, reaping, sapling, winnowing, and storage of grain in Indian scenario has yet to be revealed.

The ovary is susceptible to the action of glucocorticoids; receptors for glucocorticoids are present and it is well known that the reproductive function may be impaired during periods of adrenal hyperactivity. In addition to a direct effect on the ovaries, glucocorticoids also affect ovarian function indirectly via the adrenal– hypothalamo–pituitary axis. One of the prominent glucocorticoids affecting ovarian function is cortisol. Serum cortisol is a well accepted stress biomarker (Labad *et al*., 2015). Cortisol is not produced de novo by the ovaries (Omura & Morohashi, 1995) but transport takes place from the adrenal glands through the circulation. It exhibits prominent actions in ovaries leading to alterations of hormone production in preovulatory follice and post ovulatory corpus luteum (Andeson, 2002).

The primary aim of the present study was to monitor the socioeconomic and nutritional status followed by incidence of any menstruation disorder of the waged agricultural laboured females. Moreover the present study specifically aimed to observe the alteration of female hormonal patterns as biochemical manifestation of female reproductive morbidity along with estimation of stress hormone as biochemical biomarker for stress in this occupationally challenged population.

2 Methodology

2.1 Selection of Study Population

A longitudinal study tenured for 1 year (since January 2013) in female population who were engaged in rice plantation like sowing, reaping, threshing and winnowing were selected as study population. They were not directly involved in any field management activities like spraying of fertilizers, insecticides and fungicides.
All females were of the child bearing age group (22-30) years. A total of 70 females of this occupational sector (WAL group) were surveyed. Amongst them, females who were ligated (n=18) or taking any medications (n=8), previous history of any disease (n=3) were discarded. Females who were willing to co-operate for invasive study were preferred. One such unwilling subject complaining menstrual trouble was filtered. A total of forty such females were involved in this study.
Control population (CH) involved rural females who were not involved in any waged labourer activities. A willing population of 40 females of the desired age group was monitored.

2.2 Questionnaire

The subjects were made well familiar and well comfortable to the research personnel. The questionnaire involved the following aspects: age, year of marriage, number of offspring, interval of issue, history of any disease, use of intoxicants, duration of education, duration of daily work and other activities, sleep and recreation, period of involvement in the occupation, family income, menstrual irregularities and related disorders for last few months.

2.3 Assessment of nutritional status

BMI of the studied population were calculated using height and weight. The anthropometric measurements viz., height in cm, weight in kg, waist and hip circumference of each respondent were recorded. The nutritional status was assessed by calculating Body mass index (BMI) and Waist to hip ratio (WHR). The respondents were categorized into underweight, normal at risk of obesity and obese categories depending upon the BMI classification given by WHO (2004). Body mass index was calculated by using height and

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weight of the respondents. WH ratio was calculated. Respondents were divided into two categories viz., normal and abdominal as per the classification given below (Bray, 1987).

2.4 Blood collection

5 ml blood samples (follicular phase) were collected from radial vein by the help professional compounder early in the morning at about 5.30 p.m. with disposable syringe from the subjects in labeled vials. Serum was isolated and used for the analysis of FSH, LH, estrogen, and cortisol level.

2.5 Assay of serum hormones

FSH, LH, estrogen, cortisol was estimated by CLIA kits procured from Diagnostic Automation / Cortez Diagnostics, Inc., California, USA. following the underlined principle.
Principle of CLIA: Chemiluminescence Immunoassay (CLIA) detection using microplate luminometers provides a sensitive, high throughput, and economical alternative to conventional colorimetric methodologies, such as Enzyme-linked immunosorbent assays (ELISA)..

The specific hormone EIA is based on the principle of competitive binding between hormone in the test specimen and the same in HRP conjugate for a constant amount of rabbit anti-hormone. In the incubation, goat anti-rabbit IgG coated wells are incubated with 25 µl hormone standards, controls, patient samples, 100 µl hormone-HRP Conjugate Reagent and 50 µl rabbit anti-hormon reagent at room temperature (18-25°C) for 60 - 90 minutes (as specified in the kit). During the incubation, a fixed amount of HRP-labeled hormone competes with the endogenous hormone in the standard, sample, or quality control serum for a fixed number of binding sites of the specific hormone antibody. Thus, the amount of hormone peroxidase conjugate immunologically bound to the well progressively decreases as the concentration of hormone in the specimen increases. Unbound hormone peroxidase conjugate is then removed and the wells washed. A solution of chemiluminescent substrate is then added and read relative light units (RLU) with a Luminometer. The intensity of the emitting light is proportional to the amount of enzyme present and is inversely related to the amount of unlabeled hormone in the sample. By reference to a series of hormone standards assayed in the same way, the concentration of hormone in the unknown sample is quantified.

2.6 Statistical Analysis

The Mean ± SD of the study population, two tailed Students t test was computed using Minitab Statistical Software. Pearsons correlation was computed and the computed r was tested for significance using the mentioned software. $p<0.05$ was considered as biologically significant.

3 Results

The variation in average work hours of WAL at field and at house hold (11.36±0.87) was 35.8% greater than CH (8.36±1.66); while sleep was 18.4% (5.96±0.4 hrs.) less than of the CH population (7.3±0.52 hrs.).

The WAL females reported the problems of menstruation where oligomenorrhea was prevalent in 58% of the females. Another 10% reported dysmenorrhea. 5% females had no menstruation since the last six months; metrorrhagia was reported by 4% of the females. The CH population reported the incidence of oligomenorrhea in 30% of females, dysmenorrhea in 2% and hypomenorrhea in 1% of the population.
The nutritional status of the respondents reported (19.37±0.9, 818.8±0.95; 3.03 %($p>0.05$))

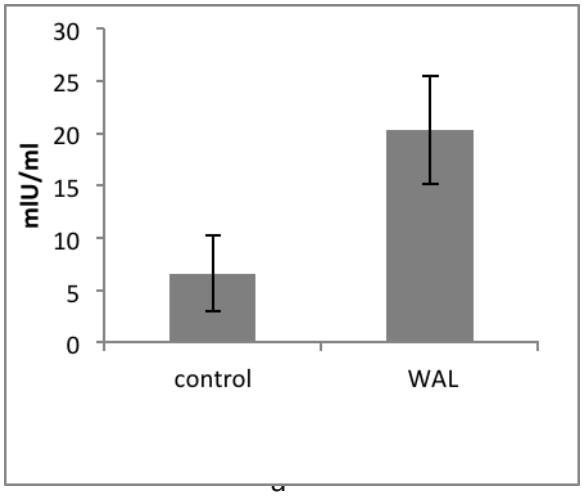
and (0.86±0.027, 0.80±0.028; 7.5 % ($p<0.05$)) increase in mean BMI and WHR of the WAL females respectively compared to the CH (Figure 1 a & b).

The WAL females reported elevated mean serum LH (21.18±4.24mIU/ml, 5.55±2.77mIU/ml) level by 281.7% ($p<0.001$), followed by cortisol level (16.16±3.12 µg/dl, 8.1±1.04µg/dl;(99.5%, $p<0.001$)) and FSH(8.79±1.33 mIU/ml, 6.97±2.20 mIU/ml) by 26.1% ($p< 0.01$). However there was 48.6% ($p<0.001$) fall in serum estrogen level (32.25±5.66 pg/ml, 61.1 ± 1.945 pg/ml) in the occupationally challenged females (Figure 1).

The Pearson's r indicated that cortisol, biochemical stress marker, was adequately correlated to sleep ($r = -0.399$, $p<0.01$); physical labour ($r= 0.625$, $p<0.0001$); FSH ($r=0.596$, $p<0.001$); LH ($r= 0.688$, $p<0.0001$); estrogen ($r = 0.58$, $p<0.001$) in the WAL females as tabulated below (Table 1).

Table 1 Bivariate Correlation coefficient of cortisol with labour, rest, FSH, LH, estrogen in WAL and CH

	Physical labour(hrs)	Sleep(hrs)	FSH (mIU/ml)	LH (mIU/ml)	Estrogen (pg/ml)
Cortisol (WAL) (µg/dl)	r=0.6255 (p<0.0001) 95% CI for ρ (0.39 0.784)	r=-0.399 (p<0.01) 95% CI for ρ (-0.632 -0.101)	r=0.596 (p<0.0001) 95% CI for ρ (0.35 0.765)	r=0.688 (p<0.0001) 95% CI for ρ (0.48 0.823)	r=-0.58 (p<0.0001) 95% CI for ρ (0.757 -0.333)
Cortisol (CH) (µg/dl)	r=0.183 (p=0.42) 95% CI for ρ (-0.423 0.19)	r=-0.574 (p<0.0002) 95% CI for ρ (-0.751 -0.32)	r=0.301 (p=0.58) 95% CI for ρ (-0.01 0.56)	r=0.258 (p=0.10) 95% CI for ρ (0.016 -0.157)	r=-0.153 (p=0.34) 95% CI for ρ (-0.443 0.165)



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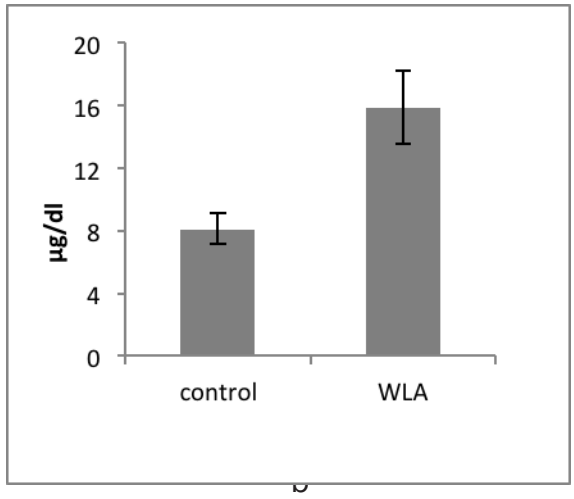

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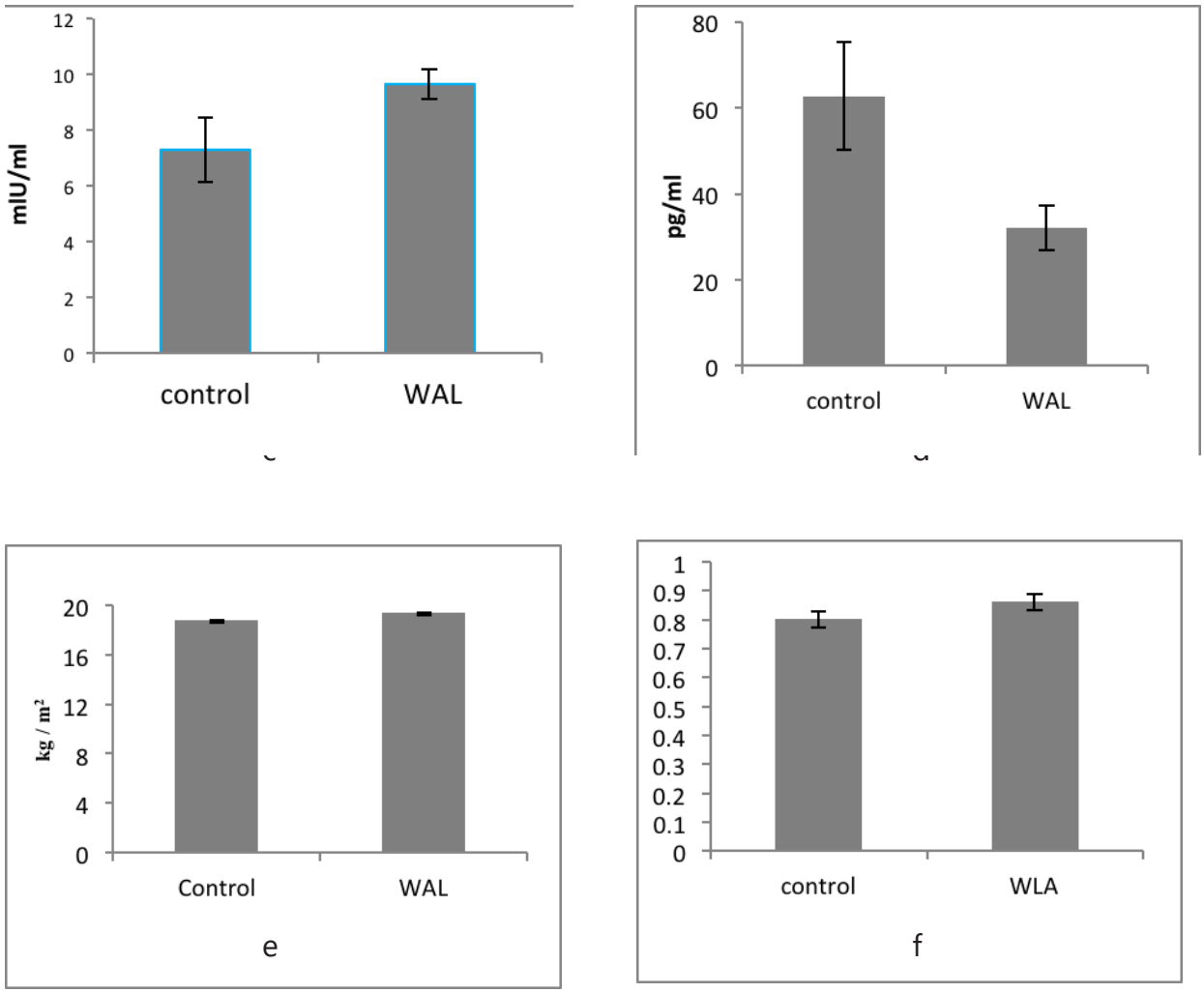

Figure. 2 BMI, WHR, and hormonal changes in the WAL and CH populations: a - BMI ($p>0.05$), b - WHR ($p<0.05$), c - LH ($p<0.001$), d - cortisol ($p<0.001$), e - FSH ($p<0.01$), f - estrogen ($p<0.001$)

4 Discussion

Reproductive function in females has been shown to be overwhelmed with abnormalities in nutritional status (ESHRE Capri Workshop Group, 2006). Surprisingly the BMI of study populations revealed that they were neither underweight nor obese, as formerly figured out in rural females (Uday Lakshmi and Babitha, 2014).Thus the complications of high or low BMI may be taboo for this study.

Conversely the issue of increasing involvement of females in occupations may be a root for hazardous reproductive implications. Stress and occupational exposure to chemicals may have poor reproductive outcomes in females (Burdorf *et al*., 2006).Although monitoring of the present study population did not reveal the direct exposure to harmful work-place chemicals, stress factor could not be overruled.

Serum cortisol is a well accepted stress biomarker (Labad *et al*., 2015). Amplified cortisol response may be tuned to physical work load; long working hours and inadequate sleep. Work-related stressors have been associated with augmented cortisol response (Sluiter *et al*, 2001; Thomas, Hertzman, and Power, 2009; Thayer *et al*., 2010). Moreover stress in

individuals with high work load is associated with elevated morning cortisol level (De Vente *et al*., 2003). The study population showed 24 / 8 hrs fieldwork loads, 5.8 hours of average sleep and 1.2 hrs of leisure emphatically befitting a higher titre of morning serum cortisol than their control counterparts.

Serum FSH and LH are essential gonadotropins for detection of normal ovarian function. Early follicular FSH levels along with estradiol levels are an indicative of ovarian reserve. Usually cohort of follicles released during this phase is dependent on the initially low estradiol (80 pg / ml) and high FSH levels (<10 mIU/L), a higher FSH or estradiol above the desired level may be an indicative of poor ovarian reserve (Broekmans *et al*., 2006; Singh *et al*., 2007).The present study demonstrated significantly elevated FSH levels in agricultural laboured females compared to the control, estrogen levels were significantly decreased. LH levels were in-contrast elevated. Elevated LH can affect follicular maturation, luteinization, fertilization and pregnancy (Paulson *et al*., 1992).

Stress and amplified cortisol in females have been previously shown to dampen the pituitary gonadal axis. Hydroxycortisone administration to human has shown to suppress the mean FSH, LH as well as pulse frequency and amplitude of these gonadotropins (Saketos, Sharma and Santaro, 1993). Our findings unlikely explored an elevated gonadotropin level with higher significant titre of serum LH than FSH suggesting a stress induced over secretion of gonadotropins. These findings are partially supportive of previous study of Nepomnaschy *et al*. where stress induced hypercortisolism was associated with elevated follicular gonadotropins and progestrin levels in follicular phase that might impair reproductive functions of rural Mayan females (Nepomnaschy *et al*., 2004).

Disorders of menstruation seem to be cumulatively determined by factors controlling the follicular phase. Variability in the length of this phase may account for polymenorrhea or oligomenorrhea. Increased gonadotropin levels and low estrogen levels may be a significant marker for oligomenorrhea as observed in the present study. Moreover stress and extended physical activity may be a predisposing factor for the said disorder (Wong, 2011) as evidenced form this study.

5 Conclusion

It may be concluded that low estrogen may be due to excess cortisol formation. High cortisol and elevated FSH may signify ovarian ageing. High LH and comparatively low FSH may further highlight elongated follicular phase with increase in cycle length leading to a decrease in frequency of cycles. This may be a cause for prevalent oligomenorrhea in these females. We thus conclude that physical stress engendered by long work periods (including field and household activities and inadequate sleep) may persuade cortisol secretion in dictating ovarian function of the affected females leading to reproductive morbidity in these waged agricultural female labourers. This should be immediately highlighted to the society so that these females may get adequate social care towards their better wellbeing.

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