



Research Article

BIOCHEMICAL EVALUATION OF SALIVARY SIALIC ACID IN WOMEN UNDER DIFFERENT REPRODUCTIVE PHYSIOLOGICAL CONDITIONS

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ABSTRACT

In human female reproductive physiology predicting ovulation is the basis on which the fertile period is determined. The present study was designed to investigate the level of salivary Sialic acid to find the possibility to develop a biomarker for detection of ovulation by non-invasive method. 42 females of different age group of different reproductive physiological conditions (i.e. prepubertal, parous, non-parous, menopausal, and diabetogenic) were selected for this study. It was observed significant ($p < 0.001$) increase in ovulatory, menopause and diabetic condition and highly significant decrease in non-parous ovulatory and post ovulatory condition in women. The study showed that salivary Sialic acid concentration was elevated during ovulation.

Keywords: Diabetogenic, Menopausal, Non-Parous, Parous, Pre-pubertal, Sialic Acid.

INTRODUCTION

Sialic Acid is a constituent of many salivary glycoproteins. Elevated salivary Sialic acid was observed in pregnancy, Down's syndrome and in Diabetes mellitus (Dharapur, 1999; Yarat *et al.*, 1999). Earlier report of Dabelsteen *et al.*, 1991 indicated elevated level of salivary Sialic acid in various type of cancer in comparison to healthy human subjects. Some reports shows that the saliva is a very good source of both hormones and biomolecules and that their level changes in accordance with the reproductive cycle in women (Weidong *et al.*, 1995) indicated that estrogen (ovarian steroid) stimulated the breakdown of glycogen and other material into glucose. Recent reports shows that composition of human saliva altered during pregnancy, menstruation and menopause due to changes in steroid hormones level whereas increase in glucose level in Diabetic patients diminished flow of saliva which causes dry mouth (Twetman *et al.*, 2002). Sialic acid is a constituent of human salivary mucin. It is obtained by precipitating the mucin from saliva with ammonium sulphate, weak acids, acetone or alcohol. Dried salivary mucin contains 2 percent Sialic acid (Berggård & Werner, 1958). It comprises of 9 carbon α - keto acids with a nine

carbon backbone (Reuter & Gabius, 1996; Varki, 2008) and is located at the non-reducing end of glycans (Schauer *et al.*, 1995). An important function of host sialic acid is to regulate innate immunity. In Human, brain has the highest Sialic acid concentration due to which disorder of Sialic acid primarily affects the nervous system (Schnaar *et al.*, 2014). Various studies have reported the significance of Sialic acid as a tumor marker in cancerous conditions (Malati, 2007). Metastatic breast cancer cell often express a high density of Sialic Acid rich glycoprotein (Jin & Mu, 2015). Serum sialic acid is increased during pregnancy (Crook *et al.*, 1997).

N-acetyl neuraminic acid (commonly referred to as sialic acid), is a negatively charged Nine carbon monosaccharide commonly attached by α -glycosidic linkage. Sialic acids are important terminal sugars of the carbohydrate chains of cell membrane glycoproteins and glycolipids (Schauer, 1982). More than 50 kinds of sialic acid are known, all of which can be obtained from a molecule of neuraminic acid by substituting its amino group of one of its hydroxyl groups (Varki, 2008). Generally plants do not contain sialic acid (Freeze *et al.*, 2009). It plays a vital role in a variety of physiological and pathological processes (Angata & Varki,

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2002). It indicates that salivary hypofunctions to oral infections such as caries and periodontitis might be responsible for concentration of Sialic acid in saliva of diabetic women (Twetman *et al.*, 2002). Lemay *et al.*, 1982 reported that a significant increase in circulating estradiol concentration occur in normal menstruating women one week before the LH surge. Sialic acid causes an impairment of cell maturation (Annunziata *et al.*, 1983). Elevated plasma Sialic acid concentration is strongly related to the presence of complications in Type 1 diabetes (Crook *et al.*, 1993). In Type 2 diabetes the circulating Sialic acid concentration is elevated (Crook *et al.*, 1993).

MATERIALS AND METHODS

Collection of Samples

Unstimulated whole saliva was collected from 42 women and ovulation was detected. Samples were divided into five categories; as prepubertal (7-9 years), pre-ovulatory phase (6-12 days), ovulatory phase (13-14 days), post-ovulatory phase (15-26 days), and menopause (above 45 years). Saliva was collected in morning. Subjects were requested not to consume food or drink at least 2 hrs. prior to the

collection. Saliva was collected by spitting method into a sterile labeled container placed on ice and then stored at -20°C and centrifuged at 4°C at 4000rpm for 45 mins. The supernatant was transferred into tube and sealed with paraffin dried in evacuator and stored at -20°C.

Biochemical assay

Each sample was subjected to evaluation of the Sialic Acid. Salivary Total protein was estimated by means of (Lowry *et al.*, 1951). The data obtained was subjected to a statistical analysis.

RESULTS AND DISCUSSION

The results revealed that a highly significant decreased level of sialic acid is observed in parous pre-ovulatory phase and highly significant increased level of Sialic Acid in saliva of Ovulatory and decreased in saliva of postovulatory phase in comparison to prepubertal condition might due to lower activity of gonadotropin hormones in prepubertal condition and active ovarian steroidogenesis activity due to active gonadotropic effect (Figure 1).

Table 1. Parous and Non-parous ovulatory might be due to its estrogen depending in saliva.

S. No.	Name of different conditions with symbols	Level of sialic Acid (mg/dl) mean \pm SE of 5 samples	P-Value
1	<i>Pre pubertal</i> –(a)	11.31 \pm 0.07	
2	<i>Parous</i>		a to b – (p<0.02) S, a to c – (p<0.001) HS, a to d – (p<0.001)HS a to e –(p<0.001)HS a to f – (p<0.001)HS a to g –(p<0.001)HS
	I. <i>Pre ovulatory</i> –(b)	10.73 \pm 0.15	
	II. <i>Ovulatory</i> – (c)	42.57 \pm 0.24	
	III. <i>Post ovulatory</i> –(d)	8.64 \pm 0.10	
3	<i>Non- parous</i>		b to e–(p<0.001)HS c to f–(p<0.05)S d to g –(p<0.001)HS
	I. <i>Pre ovulatory</i> –(e)	9.57 \pm 0.19	
	II. <i>Ovulatory</i> –(f)	31.30 \pm 0.22	
	III. <i>Post ovulatory</i> –(g)	7.27 \pm 0.09	
4	<i>Menopausal</i> –(h)	19.23 \pm 0.52	a to h – (p<0.001)HS b to h –(p<0.001)HS c to h –(p<0.001)HS d to h – (p<0.001)HS e to h –(p<0.001)HS f to h – (p<0.001)HS g to h –(p<0.001)HS
5	<i>Diabetogenic</i> –(i)	16.57 \pm 0.15	a to i –(p<0.001)HS, , b to i –(p<0.001)HS. c to i –(p<0.001)HS d to i –(p<0.001)HS e to i – (p<0.001)HS f to i –(p<0.02)S g to i – (p<0.02)S h to i – (p<0.01)HS

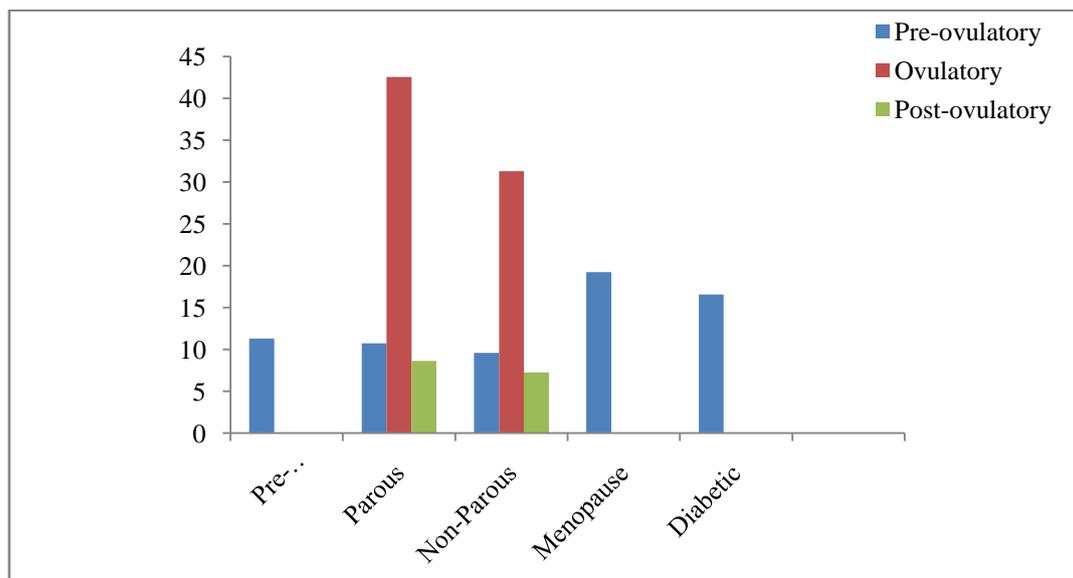


Figure 1. Hormones in prepubertal condition and active ovarian steroidogenesis activity due to active gonadotropic effects.

The decrease in Sialic acid concentration during the preovulatory phase has been observed both in human cervical remarkable cyclic variations usually with the high concentration around ovulatory period. Decrease in Sialic acid concentration during the pre-ovulatory phase has been observed both in human cervical mucus and human whole saliva (Oster & Yang, 1972). But highly significant decreased Sialic acid concentration in saliva of non-parous, preovulatory, ovulatory and postovulatory phase of menstrual cycle than parous phase of menstrual cycle might be an indication of disturbed gonadal hormone release. In menopausal condition the saliva concentration of Sialic acid showed a highly significant increase than prepubertal condition but highly increased concentration than parous and non-parous preovulatory and decreased concentration than parous & non-parous ovulatory might be due to its estrogen depending in saliva (Table1). In diabetogenic women highly significant increased concentration than menopausal but increased and decreased concentration in comparison to parous & non-parous phase of menstrual cycle might have been due to the deregulation of metabolic activity during diabetic condition.

CONCLUSION

This study concluded that saliva –specific carbohydrates (i.e. Sialic acid) in the ovulatory saliva makes the possibility to develop a biomarker for detection of ovulation by non-invasive method.

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