Effect of Infant Massage on Salivary Oxytocin Level of Mothers and Infants with Normal and Disordered Bonding

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Abstract
Maternal-infant bonding is an affective maternal-driven process that occurs primarily to her infant. Prophylactic interventions or treatment of disordered bonding include infant massage. Evidence suggests that oxytocin plays an important role in facilitation of mother–infant bonding. Main objective is to assess the effect of infant massage on salivary oxytocin level of mothers and their infant during postpartum period. And to assess the difference of oxytocin level in normal and disordered maternal-infant bonding. This study is a quasi-experimental study, carried out on 37 pairs of mothers and their infants from second to sixth month postpartum, attending Basateen Gharb primary health care center (PHC) in Albasateen district, Cairo, Egypt. Postpartum Bonding Questionnaire (PBQ) was used to differentiate between mothers with normal and disordered bonding. Pre and post massage salivary samples were taken from mothers and their infants. Tappan’s technique of infant massage was used. Results showed that 48.6% (N = 18) of mothers had disordered maternal infant bonding. Mothers and infants with normal bonding showed a positive relationship with their salivary oxytocin level post massage. On the other hand, mothers and infants with disordered bonding showed no change in their salivary oxytocin level post massage. Salivary oxytocin level in male infants has decreased post massage, while oxytocin level in female infants has increased post massage in mothers with normal bonding. We concluded that infant massage increases salivary oxytocin level in mothers and infants with normal bonding and it has no effect on salivary oxytocin level of mothers and infants with disordered bonding.

Keywords
salivary oxytocin, maternal infant bonding, infant massage, PBQ

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Introduction
Postpartum period is a critical phase for mothers and infants where major physiological, psychological and social changes occur that can influence their health.¹

Postpartum maternal-infant bonding refers to the mother’s attachment to her infant and is defined as a unique relationship between 2 people that endures through time.² Maternal-infant bonding has a positive impact on the physical and emotional health of the infant during his life.³

Infant massage is particularly empowering and can improve the maternal-infant attachment process.⁴ Early and extensive contact between mother and her infant enables her to become acquainted with her infant. Feeding, embracing, rocking, maintaining prolonged visual contact, and actively seeking these opportunities for interaction with the infant all foster the development of an affective tie.⁵ Most of the abovementioned behaviors occur in the process of infant massage, especially one that is as structured as the technique instructed to the mothers in the study.

The effect of infant massage on oxytocin release is bidirectional, since both massage deliverers and receivers are affected.⁶ Oxytocin, known informally as the “love hormone,” is a small peptide neurohypophysial hormone

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that was first known for its role in lactation and delivery. Then milk ejection property of oxytocin was described.7-11 After that, many researches have showed the prosocial role of oxytocin. It has a role in social and emotional regulation, and the facilitation of maternal infant bonding.12-16 Salivary oxytocin, the measurement used in this study, is a reliable biomarker of peripheral oxytocin.17

Studying the effect of infant massage on oxytocin levels in infants and mothers is compelling, especially when the analysis draws a distinction between mothers and infants who have normal bonding and others who have disordered bonding. We hypothesized that salivary oxytocin level will increase in both mother and infant with normal bonding following infant massage.

The main objective of the study was to assess the effect of infant massage on salivary oxytocin levels of mothers and their infants during the postpartum period, and to assess the difference of the oxytocin level in normal and disordered maternal-infant bonding.

Material and Methods

Subjects and Participants

This study is a quasi-experimental study conducted on mothers and their infants from second to sixth months postpartum, attending PHC Basateen Gharb, Cairo, Egypt, for routine follow up visits, during the period from September to November 2015.

Exclusion criteria included mothers with twin infants, taking psychiatric medications or hormonal therapy and infants with contagious skin disease or unstable medical condition.

All mothers and infant with inclusion criteria were asked to participate in the study during a period of 2 months. Forty five (45) pairs of mother and infant (90 participants) were asked to participate in the current study with total collection of 180 samples of salivary oxytocin, as each pair gave 2 samples of salivary oxytocin, pre- and post-massage.

After oxytocin analysis, 8 pairs were excluded from statistical analysis due to undetectable oxytocin levels or due to laboratory errors. Accordingly, the number of participants included in the statistical analysis was 37 pairs of mother and infant with 148 samples of salivary oxytocin.

Design and Procedures

Informed consent, socio-demographic data, and Postpartum Bonding Questionnaire (PBQ) were administrated to participant mothers. Pre massage salivary samples were collected from mothers and their infants. Infant massage was then performed; the researcher taught the mothers how to massage their infants by demonstration on a soft doll (according to Infant massage technique described in Tappan’s Handbook of Healing Massage Techniques).18

Mothers performed the massage technique on their infants for 7 min. Post massage salivary samples were collected from mothers and their infants. To avoid the rising peak of salivary oxytocin in relation to breastfeeding, assessments were arranged to take place at least half an hour after breastfeeding and to end at least half an hour before breastfeeding.19 The whole process took between 1 and 1.5 h/s with each participant depending on the time spent on infant soothing.

Postpartum Bonding Questionnaire PBQ

It is a 25-items screening questionnaire; it reflects the mother’s feelings and attitudes toward her infant and records early indications of disorders in mother infant bonding and relationship. Mothers respond to statements on a 6-point scale—always, very often, quite often, sometimes, rarely, or never.20,21

PBQ is scored on 4 factors—a general factor (scale 1; items no. 1, 2, 6, 7, 8, 9, 10, 12, 13, 15, 16, 17), rejection and pathological anger (scale 2; items no. 3, 4, 5, 11, 14, 21, 23), anxiety about the infant (scale 3; items no. 19, 20, 22, 25), and incipient abuse (scale 4; items no. 18 and 24). Scale 1 cutoff score ≥12 denoted pathological bonding.20

Salivary Oxytocin Techniques and Measurements

Sample collection from the mothers: Mothers were asked to reserve the saliva in their mouths without swallowing for 1 min, and then they were asked to spit in a small plastic cup. If the amount was not sufficient, they were asked to spit another time. The saliva was then poured into a plastic tube of 1.8 ml, labeled, and immediately stored in an ice bag.

Sample collection from infants: Infants were laid supine on bed, their head were tilted to one side and a plastic disposable pastier tube was used to collect the saliva from the side of the mouth, dispensed in the plastic tube of 1.8 ml and immediately stored in the ice bag.

Seven minutes after massage sessions, post-massage saliva was collected from mothers and their infants using the same procedure.

The date, time, and sample number for each sample were recorded. The saliva samples were transported in ice bags and stored at −80°C in the Biochemistry department, Cairo University, until measured.

The oxytocin level in saliva was assessed by Enzyme-linked immunosorbent assay (ELISA) technique following the method of Carter et al.17
Data were statistically described in terms of mean, standard deviation (SD), or frequencies (number of cases) and percentages when appropriate. Comparison of numerical variables between the study groups was done using Student t test for independent samples. For comparing categorical data, Chi-square test was performed. Correlation between various variables was done using Pearson moment correlation equation for linear relation in normally distributed variables. P value less than .05 was considered statistically significant. All statistical calculations were done using computer program SPSS (Statistical Package for the Social Science; SPSS Inc., Chicago, IL, USA) release 15 for Microsoft Windows (2006).

After oxytocin analysis, participants were divided, according to their results on PBQ, into normal and disordered bonding and their salivary oxytocin results were correlated using Pearson correlation equation.

**Results**

**Socio-demographic Characteristics**

Age of mothers range is 20 to 37 years with a mean 27 years and Standard deviation (SD) ± 5.0. The majority of mothers were 30 years and below (78.3%), and 97.3% were house wives. Sixty five (65%) of participants had educational level higher than primary school. Age of infants range is 2 to 6 months with a mean of 4 months and SD ± 1.3. Regarding infant feeding; 51.4% of infants were exclusively breast fed, 27% were bottle fed, and 21.6% mixed feeding.

**Massage Experience**

Majority of mothers didn’t have a previous massage experience 75.7% (n = 28). While 19% (n = 7) have a cultural massage experience, as they were guided by their grandmothers to flex and extend their infant knees and arms (as step 9 and 10 in Tappan’s massage technique).

**Maternal-infant Postpartum Bonding**

Participants were divided according to their response to PBQ into normal distorted bonding (General factor score equal or above 12). Accordingly, 48.6% (N = 18) of participants had disordered maternal infant bonding. Two mothers (5.4%) experienced infant focused anxiety. None of the participants experienced rejection, pathological anger, or incipient abuse.

**Salivary Oxytocin Results**

Salivary oxytocin levels of participants are demonstrated in Table 1. Table 2 shows a significant positive correlation between pre and post massage maternal salivary oxytocin level (r = 0.372, P = .023) and a negative significant correlation between maternal and infant salivary oxytocin level post massage (r = -0.336, P = .042). We compared salivary oxytocin pre and post massage in mothers and infants with normal and disordered bonding. There was a significant positive correlations between salivary oxytocin level pre and post massage in mothers and infants with normal bonding (P = .05 and P = .041, respectively). On the other hand, there were insignificant correlations in mothers and infants with disordered bonding (Table 3).

According to Table 4, male infant has a significant higher basal (pre massage) oxytocin level than female infant. Salivary oxytocin level in male infants decreased significantly following massage while oxytocin level in female infant increased significantly following massage as reflected in rate of change (Figure 1).

It was observed that participants with normal bonding, salivary oxytocin level significantly decreased following massage in male infants, and significantly increased in female infants, as reflected in rate of change. While participant with disordered bonding, there was insignificant difference in maternal and infant oxytocin level or change pre and post massage (Table 5).

**Discussion**

The current study hypothesized that salivary oxytocin levels in both mother and infant with normal bonding will increase following infant massage. This hypothesis was proved by the significant correlation between maternal salivary oxytocin level pre- and post-massage (P = .023). By comparing oxytocin levels in participants with normal and disordered bonding, there was a significant positive increase in maternal oxytocin levels in
Table 2. Correlations between Maternal and Infant's Salivary Oxytocin Levels Pre and Post Massage.

|                          | Maternal oxytocin level (pre massage) | Maternal oxytocin level (post massage) | Infant oxytocin level (pre massage) | Infant oxytocin level (post massage) |
|--------------------------|---------------------------------------|----------------------------------------|-------------------------------------|--------------------------------------|
| Maternal oxytocin level (pre massage) | R 1 0.372                             | 0.113                                  | 0.098                               |
|                           | P value – .023                         | –                                      | .507                                | .566                                 |
| Maternal oxytocin level (post massage) | R 0.372                               | 1                                      | −0.141                               | −0.336                               |
|                           | P value .023                           | –                                      | .407                                | .042                                 |
| Infant oxytocin level (pre massage)    | R 0.113                               | −0.141                                 | 1                                   | 0.175                                |
|                           | P value 0.507                          | .407                                   | –                                   | 0.301                                |
| Infant oxytocin level (post massage)    | R 0.098                               | −0.336                                 | 0.175                               | 1                                    |
|                           | P value .566                           | .042                                   | .301                                | –                                    |

Table 3. Correlations of Salivary Oxytocin Levels Pre and Post Massage in Participants with Normal Bonding and Disordered Bonding.

In participants with normal bonding N= 19

|                          | Maternal oxytocin level (pre massage) | Maternal oxytocin level (post massage) | Infant oxytocin level (pre massage) | Infant oxytocin level (post massage) |
|--------------------------|---------------------------------------|----------------------------------------|-------------------------------------|--------------------------------------|
| Maternal oxytocin level (pre massage) | R 1 0.451                             | 0.160                                  | 0.061                               |
|                           | P value – .05                          | .512                                   | .805                                |
| Maternal oxytocin level (post massage) | R 0.451                               | 1                                      | −0.421                               | −0.343                               |
|                           | P value .05                            | –                                      | .072                                | .151                                 |
| Infant oxytocin level (pre massage)    | R 0.160                               | −0.421                                 | 1                                   | 0.474                                |
|                           | P value .512                           | .072                                   | –                                   | .041                                 |
| Infant oxytocin level (post massage)    | R 0.061                               | −0.343                                 | 0.474                               | 1                                    |
|                           | P value .805                           | .151                                   | .041                                | –                                    |

In participants with disordered bonding N= 18

|                          | Maternal oxytocin level (pre massage) | Maternal oxytocin level (post massage) | Infant oxytocin level (pre massage) | Infant oxytocin level (post massage) |
|--------------------------|---------------------------------------|----------------------------------------|-------------------------------------|--------------------------------------|
| Maternal oxytocin level (pre massage) | R 1 0.212                             | 0.009                                  | 0.168                               |
|                           | P value – .398                         | .972                                   | .506                                |
| Maternal oxytocin level (post massage) | R 0.212                               | 1                                      | 0.182                               | −0.340                               |
|                           | P value .398                           | –                                      | .471                                | .168                                 |
| Infant oxytocin level (pre massage)    | R 0.009                               | 0.182                                 | 1                                   | −0.208                               |
|                           | P value .972                           | .471                                   | –                                   | .409                                 |
| Infant oxytocin level (post massage)    | R 0.168                               | −0.340                                 | −0.208                              | 1                                    |
|                           | P value .506                           | .409                                   | –                                   | –                                    |
| Maternal oxytocin level (pre massage) | R 1 0.212                             | 0.009                                  | 0.168                               |
|                           | P value .506                           | .168                                   | .409                                | –                                    |
mothers with normal bonding, whereas there was no significant increase in salivary oxytocin levels in mothers with disordered bonding.

The aforementioned findings highlight that normal maternal infant bonding is associated with bidirectional response to massage, which was expressed in terms of oxytocin change. On the other hand, disordered bonding is associated with maternal and infant unresponsiveness to massage and consequently no change in oxytocin level. This finding is in agreement with another study done in 2010 that concluded that mothers who provided high levels of affectionate contact with their infants showed an oxytocin increase following this contact.22 It is also in agreement with the systematic review conducted in 2011 and the study conducted by Feldman et al, in 2007 who concluded that significant and strong associations between levels or patterns of oxytocin and aspects of mother-infant relations or attachment.23,24 Another study conducted in 2010 reported that significant correlations emerged between parental and infant oxytocin levels at assessments before and after tactile stimulation, and that higher oxytocin levels in parent and child were related to greater affect synchrony and infant social engagement.24

One of the new findings in the current research is the difference between male and female infants in their oxytocin levels pre- and post-massage and their correlation with the oxytocin level of mothers with normal and disordered bonding. When analyzing oxytocin level in all infants, it was found that male infants have a significantly higher basal (pre-massage) oxytocin level than that of female infants. The oxytocin level in male infants of the normal bonding group decreased significantly following massage, while the oxytocin level in female infants with normal bonding increased significantly following massage. This finding is explained in many studies which found that oxytocin and oxytocin receptor expression is usually higher in females, and that the central roles of oxytocin in behaviors and physiology are strongly dependent on steroid hormones and gender.17,25

Gao et al.,26 supports this finding by reporting that accumulating behavioral evidence suggests that oxytocin may have evolved sex-specific functional roles in the domain of human social cognition, and that oxytocin may act differentially via the amygdala to enhance the salience of positive social attributes in women but negative ones in men. Also, in intranasal oxytocin studies, males and

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**Table 4. Salivary Oxytocin Level in Male and Female Infants Pre and Post Massage.**

|                        | Mean | SD   | Mean | SD   | P value |
|------------------------|------|------|------|------|---------|
| Infant oxytocin level (pre massage) Male infant N = 20 | 324.65 | 69.215| 272.88 | 98.287 | .039    |
| Infant oxytocin level (post massage) Male infant N = 20 | 296.25 | 74.725| 299.41 | 77.817 | .624    |
| Rate of change of infant oxytocin level Male infant N = 20 | −28.40 | 114.792| 26.53 | 84.134 | .029    |
| Infant oxytocin level (pre massage) Female infant N = 17 | 272.88 | 98.287| 272.88 | 98.287 | .039    |
| Infant oxytocin level (post massage) Female infant N = 17 | 299.41 | 77.817| 299.41 | 77.817 | .624    |
| Rate of change of infant oxytocin level Female infant N = 17 | 26.53 | 84.134| 26.53 | 84.134 | .029    |

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**Figure 1.** Changes in oxytocin level in male and female infant pre and post massage.
females tend to respond differentially to oxytocin administration; males showed attenuated amygdala reactivity, and women increased reactivity of the amygdala to similar emotional stimuli.27-29

Differentiating participants into those with normal and disordered bonding revealed that oxytocin levels in male infants with normal bonding decreased significantly following massage. On the other hand, oxytocin levels in female infants significantly increased following massage, but only in participants with normal bonding. One thing that male and female infants in the disordered bonding group had in common was that their oxytocin levels post-massage remained, as was without any increase or decrease.

In our study, structured tactile stimulation to the infants failed to increase oxytocin level in mothers and infants with disordered bonding. This could indicate that the routine infant handling and caring will not lead to an increase in maternal-infant oxytocin levels, highlighting the importance of early identification and management of disordered maternal-infant bonding during the postpartum period. Much like postpartum depression is assessed during routine postpartum care in primary health care, infant-maternal bonding also needs to be routinely assessed. This would enable primary care physicians to swiftly discover bonding issues and attempt to resolve them as soon as possible.30

**Future Direction**

To our knowledge, studies that tackled the relation of maternal-infant bonding and infant massage on salivary oxytocin are scarce. Further studies are needed to bridge the current gap in knowledge in gender difference in relation to oxytocin and bonding. The above findings emphasize those changes in oxytocin level, whether up or down, occur in the presence of normal maternal infant bonding and are hindered in disordered bonding.

**Limitation of the Study**

The small sample size and its consequences in being non-representative, however, this study could pave the way for future researches in area of maternal infant bonding.

**Strength of the Study**

1. To our knowledge, this is the first study to:
   - Correlate maternal and infant salivary oxytocin levels in mothers and their infants with normal and disordered bonding.
   - Compare oxytocin level in male and female infants and correlate this finding with level of mothers with normal and disordered bonding.
   - Use Tappan’s technique of infant massage and measure salivary oxytocin in mother and infant before and after infant massage.

2. Findings of this study highlight the issue of disordered maternal infant bonding.

3. This study draws attention to the physiological changes that occur in oxytocin in infancy.

**Declaration of Conflicting Interests**

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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### Table 5. Level of Salivary Oxytocin Pre and Post Massage in Male and Female Infants Whose Mothers Have Normal Bonding and Disordered Bonding.

|                         | Male infant |            | Female infant |            | P value |
|-------------------------|-------------|------------|---------------|------------|---------|
|                         | Mean        | SD         | Mean          | SD         |         |
| In participants with normal bonding (male infants: 9, female infants: 10) |            |            |               |            |         |
| Maternal oxytocin level (pre massage) | 188.67      | 69.75      | 159           | 94.949     | .568    |
| Maternal oxytocin level (post massage) | 199.11      | 33.602     | 187.40        | 82.349     | .567    |
| Rate of change of maternal oxytocin level | 10.44       | 63.842     | 28.40         | 92.086     | .713    |
| Infant oxytocin level (pre massage) | 332.22      | 55.081     | 267.00        | 111.774    | .094    |
| Infant oxytocin level (post massage) | 297.00      | 78.804     | 296.70        | 81.791     | .837    |
| Rate of change of infant oxytocin level | −35.22      | 106.01     | 29.70         | 60.135     | .05     |
| In participants with disordered bonding (male infants: 11, female infants: 7) |            |            |               |            |         |
| Maternal oxytocin level (pre massage) | 217.82      | 50.378     | 183.57        | 51.588     | .297    |
| Maternal oxytocin level (post massage) | 213.27      | 62.368     | 207.14        | 69.127     | .928    |
| Rate of change of maternal oxytocin level | −4.55       | 74.978     | 23.57         | 71.549     | .298    |
| Infant oxytocin level (pre massage) | 318.45      | 81.125     | 281.29        | 82.951     | .294    |
| Infant oxytocin level (post massage) | 295.64      | 75.103     | 303.29        | 77.997     | .555    |
| Rate of change of infant oxytocin level | −22.82      | 126.372    | 22.00         | 115.806    | .297    |
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Research Involving Human Participants
All procedures performed in the current study were in accordance with the ethical standards of the national research committee of the Ministry of health, Egypt and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. Ethical approvals were obtained from The Research Committee in Cairo University and Ethical Research Committee in Ministry of Health, Egypt.

Informed Consent
Informed consent was obtained from all participants included in the study.

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Data Availability Statement
The data that support the findings of this study are available from the corresponding author upon reasonable request.

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