Enhancement of Conception Rate by EveCare after Ovulation Induction by Clomiphene Citrate followed by Intrauterine Insemination

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SUMMARY

Objective: In this comparative clinical trial the efficacy of EveCare and placebo was tried on 50 patients in the age group of 25-32 years, who underwent ovulation induction by clomiphene citrate and hCG followed by intrauterine insemination. Methods: The patients received EveCare or an identically prepared placebo and were advised to take the medication at a dose of 2 teaspoonfuls for 16 days after intrauterine insemination. The investigations included TSH, FSH, LH, Prolactin and E2 levels. Results: The results of these investigations showed that except for prolactin and E2 levels all the other values increased. The conception rates were high in the EveCare-treated group. Out of the 25 patients who took placebo only 7 conceived (28%) when compared to women who were on EveCare therapy. The rate of conception in EveCare therapy was 64% (16 out of 25). Conclusion: These observations indicate that EveCare can be an useful therapy in women with infertility. No untoward incidences were seen in any of the patients.

Key words: EveCare, clomiphene citrate, infertility, ovulation, intrauterine insemination

Clomiphene citrate is an effective agent to treat several disorders that induce multiple follicular maturation for in vitro fertilization and embryo transfer. Even though ovulation rate is high with clomiphene citrate, pregnancy incidence is relatively low. Clomiphene citrate inhibits fertility in female rats when administered during the tubal passage of fertilized embryo. It has been shown that clomiphene causes failure of implantation in rats during experimentally produced delayed implantation or during lactational delay. It has also been suggested that the failure of implantation after clomiphene administration may be due to inhibition of postovulatory surge, suppression of decidual response and expulsion of eggs or blastocysts from the fallopian tube into the uterus.

EveCare is an herbal preparation formulated by The Himalaya Drug Company, Bangalore, with different aqueous extracts of herbs, which are useful in various menstrual disorders such as puberty, menorrhagia, dysmenorrhea, pre-menstrual syndrome, abnormal bleeding and
threatened abortion. EveCare syrup contains *Saraca indica* (10%), *Symlocos racemosa* (6.6%), *Adhatoda vasica* (4%), *Aloe vera* (5%), *Asparagus racemosus* (6.4%), *Boerhaavia diffusa* (6.4%), *Bombax malabaricum* (2.4%), *Cocos nucifera* (6.4%) and *Tinospora cordifolia* (6.6%) as its main constituents.

The regulation of embryo transport and uterine differentiation, depends on the level of progesterone and estrogen. Thus, compounds that interfere with or augment the action of endogenous steroids could also affect implantation process in animals. Several reviews on the subject have been mentioned elsewhere. Alterations in estrogen and progesterone levels affect reproductive outcome and the two hormones do not act independently\(^1\). Among these is the inhibition of implantation in animals and humans by high levels of estrogen\(^1\). Birkenfed et al.\(^1\), reported that clomiphene citrate adversely affects the endometrium and impairs implantation. The assessment of endometrium by transvaginal ultrasound is a useful and non-invasive method to observe the endometrial conditions.

Estrogen and progesterone receptor concentrations in the endometrium, which show peak on the preovulatory day\(^1\) are induced mainly by estrogen during proliferative phase, play an important role in the endometrial growth and maturation\(^1\). Clomiphene citrate is known to suppress the endometrial estrogen and progesterone receptor concentrations\(^1\).

The present study was designed to confirm whether EveCare can overcome antiestrogenic activity/inhibitory effects of clomiphene citrate and enhance endometrium receptivity, in protecting the process of implantation and subsequent pregnancy, since EveCare is known for its use as a stimulant to the endometrium and ovarian tissue and also possesses anti-oxytocic activity.

**PATIENTS AND METHODS**

Fifty infertile women were enrolled in the study. Blood was taken for routine thyroid stimulating hormone (TSH), follicle stimulating hormone (FSH), leutinizing hormone (LH), prolactin (PRL) and estrogen (estradiol) hormonal assay. All the patients were given 50 mg clomiphene citrate for 5 days starting from day 3 to day 7 of the menstrual cycle (day 1 - day of menstruation). From day 8 follicular and endometrium developments were monitored with the use of transvaginal probe (7.5 MHz). When the follicle size reached 18 mm or >18 mm diameter, 10,000 IU human chorionic gonadotropin (hCG) was injected intramuscularly. After 36 hours of hCG injection intrauterine insemination was performed after sperm wash by swim-up technique with normozoospermic husband/donor semen. The patients were divided into two groups of 25 patients each. Group I received EveCare 2 tablespoonfuls, twice daily for a period of 16 days after intrauterine insemination and Group II was placebo group. In both the groups, 10 mg of progesterone was given twice daily for luteal support. Pregnancy test was performed on 15\(^{th}\) or 17\(^{th}\) day after intrauterine insemination to confirm pregnancy.
Hormone Assays
Blood samples were collected in the early follicular phase (day 3-5 of the menstrual cycle of spontaneous bleeding or withdrawal bleeding induced by medroxyprogesterone) through vein puncture and centrifuged within 2 hours after withdrawal. Serum was stored at −20°C until assay for hormones such as Thyroid stimulating hormone (TSH), Follicle stimulating hormone (FSH), Leutienizing hormone (LH), Prolactin (PRL) and Estradiol (E₂). All assays were performed by immunometric assay with commercially available kits (United Biotech Inc., CA, USA). To avoid interassay interference, blood samples of each subject were analysed in the same assay.

RESULTS
The hormonal profiles of the placebo group and EveCare group before treatment are shown in Table 1. The hormonal profiles of the placebo and EveCare group treatment showed no significant difference in the hormonal values. The conception rate after EveCare treatment for a period of 16 days undergoing ovulation induction by clomiphene citrate and hCG, followed by intrauterine insemination is shown in Table 2. A significant increase in the conception rate (64%) was observed compared to the placebo group (28%). None of the patients in the EveCare or placebo group had early pregnancy loss or abortion.

DISCUSSION
The treatment with clomiphene citrate has shown to inhibit fertility when administered during the first 4 days of pregnancy when the developing eggs are in the oviduct rat, but had no adverse effect when treatment was given later. Several investigators also reported the antifecundity effects of clomiphene and suggested that the possibility that failure of implantation may be due to a direct blastotoxic action of the compound. The failure of implantation in clomiphene citrate treated animals is also reported to be due to expulsion of blastocysts.

In the present study, there was a significant (p<0.05) increase in the percentage of conception rate (64 %), i.e. 16 out of 25 patients when the patients were given EveCare after intrauterine insemination for a period of 16 days compared to placebo group (28%), i.e. 7 out of 25 patients. Clomiphene citrate is a widely used nonprotein drug that induces human ovulation. By careful selection of anovulatory patients, clomiphene citrate induces ovulation in 70% of patients.

Table 1: Hormonal indices in patients with unexplained infertility before EveCare treatment

<table>
<thead>
<tr>
<th></th>
<th>Placebo Group</th>
<th>EveCare Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (Years)</td>
<td>28.64 ± 1.09</td>
<td>28.84 ± 1.06</td>
</tr>
<tr>
<td>TSH (mIU/ml)</td>
<td>1.03 ± 0.12</td>
<td>1.32 ± 0.15</td>
</tr>
<tr>
<td>FSH level (mIU/ml)</td>
<td>8.08 ± 0.74</td>
<td>9.12 ± 0.52</td>
</tr>
<tr>
<td>LH level (mIU/ml)</td>
<td>6.05 ± 0.74</td>
<td>9.37 ± 2.26</td>
</tr>
<tr>
<td>Prolactin (mIU/ml)</td>
<td>16.04 ± 1.58</td>
<td>14.19 ± 1.23</td>
</tr>
<tr>
<td>E₂ level (pg/ml)</td>
<td>59.79 ± 4.30</td>
<td>57.13 ± 3.70</td>
</tr>
</tbody>
</table>

All values are means ± SEM; NS –Not statistically significant.

Table 2: Conception rate after EveCare treatment for 16 days in patients undergoing ovulation induction by clomiphene citrate and hCG followed by intrauterine insemination

<table>
<thead>
<tr>
<th>Group</th>
<th>Total No. of patients</th>
<th>No. of patients pregnant</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Placebo</td>
<td>25</td>
<td>7</td>
<td>28.0%</td>
</tr>
<tr>
<td>EveCare</td>
<td>25</td>
<td>16*</td>
<td>64%</td>
</tr>
</tbody>
</table>

* p<0.05 significantly as compared to placebo group
women but results in only 27-40% pregnancy rate with an associated 25% abortion rate. However, in this study, none of the patients had abortion in the treatment or placebo group.

The low pregnancy and high abortion rates are also attributed to impairment of endometrium receptivity perhaps due to inhibitory actions of clomiphene citrate affecting expulsion of preimplantation embryos, increased embryo transport and uterine preparation for pregnancy due to direct or indirect hormonal actions. The increase in pregnancy rate with EveCare treatment after IUI may be attributed to some of the constituents present in EveCare such as Saraca indica known for its use as a stimulant to the endometrium, ovarian tissue and saponin glycoside obtained from Asparagus racemosus which exhibits antioxytocic activity.

In conclusion, the results of the preliminary study indicate that infertile patients undergoing follicular stimulation followed by hCG administration and subsequently undergoing intrauterine insemination followed by EveCare treatment indicate that the conception rate can be significantly increased in infertile patients undergoing assisted conception. Further studies are required to identify the specific constituents present in EveCare syrup and its effects on the endometrium during follicular and luteal phase of the menstrual cycle.

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REFERENCES


