Research Article

BIOMEDICAL Journal of Scientific & Technical Research

ISSN: 2574 -1241

DOI: 10.26717/BJSTR.2022.45.007142

A Randomised and Comparative Study to Evaluate the Efficacy of M2-Tone Tablets on Endometrial Lining and Pregnancy Rate in Infertile Females

Mojisola Aderonmu, Adebisi Oyero, Oluwole Ogunsola and Oladapo Ashiru*

Medical Art Center and Institute of Assisted Reproductive Technology, Nigeria

***Corresponding author:** Oladapo Ashiru, Medical Art Center and Institute of Assisted Reproductive Technology, Lagos, Nigeria

ARTICLE INFO

Received: June 24, 2022

Published: July 08, 2022

Citation: Mojisola Aderonmu, Adebisi Oyero, Oluwole Ogunsola, Oladapo Ashiru. A Randomised and Comparative Study to Evaluate the Efficacy of M2-Tone Tablets on Endometrial Lining and Pregnancy Rate in Infertile Females. Biomed J Sci & Tech Res 45(1)-2022. BJSTR. MS.ID.007142.

Keywords: Endometrium; Pregnancy; IVF; Implantation; M2-Tone

Abbreviations: ART: Assisted Reproductive Techniques; IUI: Intrauterine Insemination; IVF: *In-Vitro*-Fertilisation; WOI: Window of Implantation

ABSTRACT

Aim: The main aim of study was to determine the effectiveness of M2-Tone tablet in comparison with standard conventional treatment on endometrial lining and pregnancy rate in infertile women.

Design: Randomised, comparative trial.

Setting: Private Infertility Clinic and Academic Center, Nigeria.

Patients: Fifty patients who failed to develop an endometrial thickness of at least 8mm in previous evaluation cycles were recruited. Twenty five patients were administered Tablet M2- Tone and Progynova (E2v) (Test drug group) and remaining twenty five patients were given Progynova (Control group). Both drugs were given orally in the dose of 2 tablets twice a day for three months prior to the IVF procedure.

Investigational Procedures: HCG test, Transvaginal ultrasonography, IVF.

Main Outcome Measures: Endometrial lining, and pregnancy rates.

Results: In M2-Tone group endometrial thickness increased by 6.6 mm as compared to baseline. There was an increase in endometrial thickness in the M2 - Tone group as compared to the control group (with E2v only). Significant improvement was observed in implantation rate with the use of M2 - Tone Tablets.

Conclusion: The finding of present study suggests that M2-Tone tablets significantly improved endometrial thickness and also showed significant improvement in pregnancy rate. We can conclude that M2-Tone helps endometrium attain a receptive profile that may lead to successful implantation.

Introduction

In today's global scenario infertility is a common clinical problem. As per WHO estimation it affects 13 to 15% of couples worldwide [1], thus affecting 50 to 80 million worldwide, out of which 20 - 35 million couples in Africa experience this problem. This can be extrapolated to 3 - 4 million Nigeria couples suffering from infertility (Thomas 1995).Thus prevalence varies widely, being less in developed countries and more in developing countries where limited resources for investigations and treatment are available [2]. In the recent study on Nigerian Community 50.5% cases of infertility belonged to the age range of 36-45 years [3]. Nowadays, increased numbers of women delay childbearing for educational

and career goals, resulting in dramatic increase in age related infertility. With recent dramatic advances in infertility treatment, age related infertility remains as one of our most difficult challenges. Infertility specialists have known for years that the pregnancy rate is inversely related to the female partner's age. Infertility treatment includes medication (hormone stimulation), surgical correction of anatomical defects, or artificial insemination. Second option is Assisted Reproductive Techniques (ART). Regardless of advances in assisted conception, female infertility remains one of the major challenges for intrauterine insemination (IUI) and *in-vitro*-fertilisation (IVF). The incidence of female infertility is about 10-15%. The majority of these couples opt for IUI and IVF. But the overall success rate of IUI in terms of clinical pregnancy per cycle is about 10-20% and about 25-40% in patients going for IVF [4-7]. With oocyte donation the events of ovarian stimulation and the artificial preparation of endometrium are not completely met.

In IUI and IVF, implantation failure may be the major cause of unsuccessful pregnancies. Implantation is a dynamic process occurring between blastocyst and endometrial layers, the priming of the endometrium towards the window of implantation (WOI) is of pure maternal origin. A reduced endometrial receptivity is found in an increasing number of unexplained infertilities (Lessey et al., 1995). The incidence of unexplained infertility appears to have risen in the past few years. An inadequate endometrium can be considered as a main fertility-determining factor [8]. A healthy pregnancy is like a good plantation. The successful plantation depends on - the season (resembling normal ovarian steroid-dependent status), the good quality viable seeds (resembling healthy gametes i.e. sperm and ovum), the fertile soil (healthy and conducive endometrial structure) and the water (resembling normal endometrial vasculature). Poor quality of any of these four essentials leads to infertility. In the majority of unexplained infertility cases, changes in uterine epithelium (endometrium- soil) could be a major attribute.

Intensive research work has been performed to better understand the regulation of the endometrium and its clinical implications to improve implantation. Thin endometrium can result from intrauterine adhesions, endometritis, curettage, and use of oral contraceptives or clomiphene citrate [9]. The suggestion that women with infertility may have an impaired uterine blood supply was first raised by Goswamy [10,11]. Decline in endometrial receptivity, which is associated with a decrease in uterine perfusion, may play an important role in the decrease of implantation rates. Oestrogen is considered to play an important role as a modulator of uterine vascularity. It is reported that administration of oestrogen improved uterine response in patients with poor uterine perfusion [12]. Because oestrogen receptors have been identified on the walls of uterine arteries, it is likely that the effect of estradiol on the uterine flow wave is a direct one [13]. For couples who do not benefit from the general line of infertility treatments, the remaining options include gamete intrafallopian transfer, intracytoplasmic sperm injection, *in vitro* fertilisation, and zygote intrafallopian transfer. Although all of these therapies can be successful, they are often expensive, invasive, and time consuming. With the introduction and integration of CAM, there has been an increase in the acceptance and use of complementary practices in dealing with infertility [14].The use of traditional Indian medicine as primary or adjuvant therapeutic tool has become very appealing because of proven efficacy, safety and cost effectiveness.

M2-Tone tablet, a polyherbal medicine by M/s Charak Pharma Private Ltd, Mumbai has established its estrogenic effect in female infertility through various studies conducted in the past. M2-Tone has shown significant results in clinical trials on menstrual and infertility problems. Recently a trial conducted on M2-Tone by Dr. Malhotra N et al, on endometrial morphology, vasculature and pregnancy rate, M2-Tone showed significant improvement in endometrial thickness, endometrial lining and pregnancy rate (31%) as compared to placebo [15]. We aimed to establish a treatment approach in infertile women opting for ART where thin endometrial lining is a reason for consecutive failure of IVF. M2-Tone tablet was administered for 3 months (before IVF procedure) as an adjuvant in patients with older age (35-53 years) and persistently thin endometrial lining. We hypothesise that adding M2-Tone tablet prior to the IVF protocol can promote normal endometrial lining, for better implantation in the subjects. Endometrial thickness is important for implantation and there is little data addressing the aetiology of persistently thin endometrium. This study therefore aimed to evaluate the role of orally administered M2-Tone tablets in modifying endometrial thickness and the corresponding pregnancy rate in patients with infertility.

Materials and Methods

The clinical trial was conducted by Dr Oladapo Ashiru and his team on fifty female patients undergoing infertility treatment at Medical Art Center (IVF clinic in Nigeria). A prospective randomised and comparative clinical trial where a total of 50 patients with thin endometrium (<8mm), were included in this study. Informed consent was taken from each subject prior to the study. The subjects between the 35 - 53 years age group were included as per inclusion and exclusion criteria. Patients were randomly assigned to two different groups, test group and control group with 25 patients per group; patients in test group received M2-Tone tablets and Progynova (E2v) twice daily for 3 months and the control group received E2v on similar lines prior to their IVF treatment cycle. Endometrial lining was measured at baseline and end of study was measured using transvaginal scans. The endometrial thickness during the IVF cycle was monitored weekly with the use of a

transvaginal 3.5mHz transducer scan. Endometrial thickness was measured at the thickest point between the two basal layers on the anterior and posterior uterine walls. The endometrial layers were measured in millimeters.

Results

The result obtained showed a good increase in endometrial thickness when compared with the other group treated with Progynova only. There was also a significant increase in implantation rate in patients on M2-tone. The most outstanding observation found in patients in this group was the fact that they had repeated IVF failure prior to use of M2-tone. Pregnancy rate measured was about 24% (Figures 1-3).

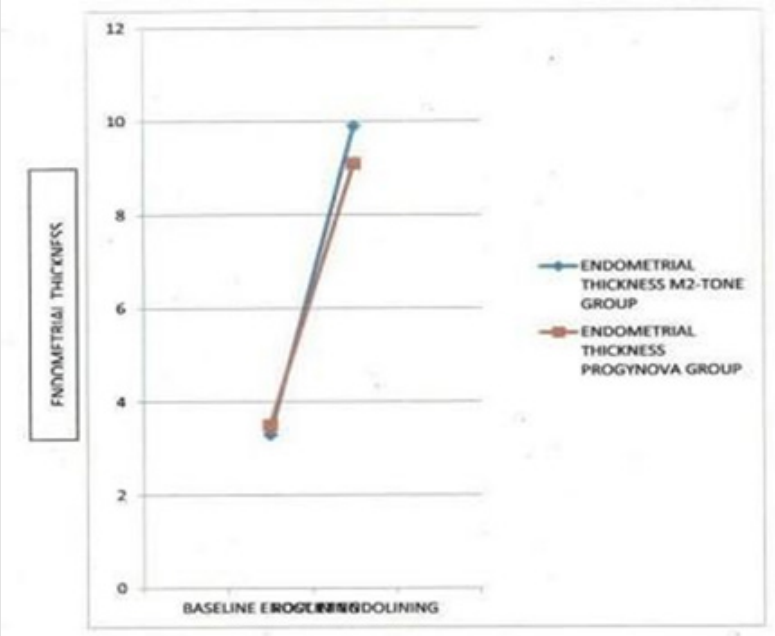

Figure 1: Effect of M2-Tone on Endometrial Thickness.

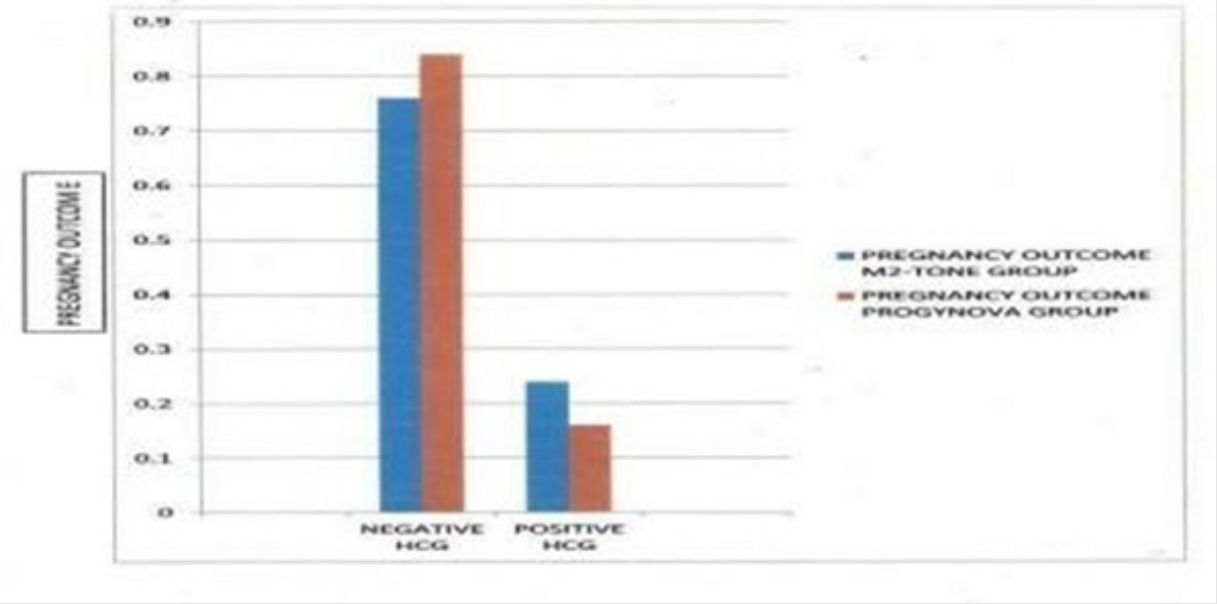

Figure 2: Effect of M2- Tone on Pregnancy Outcome.

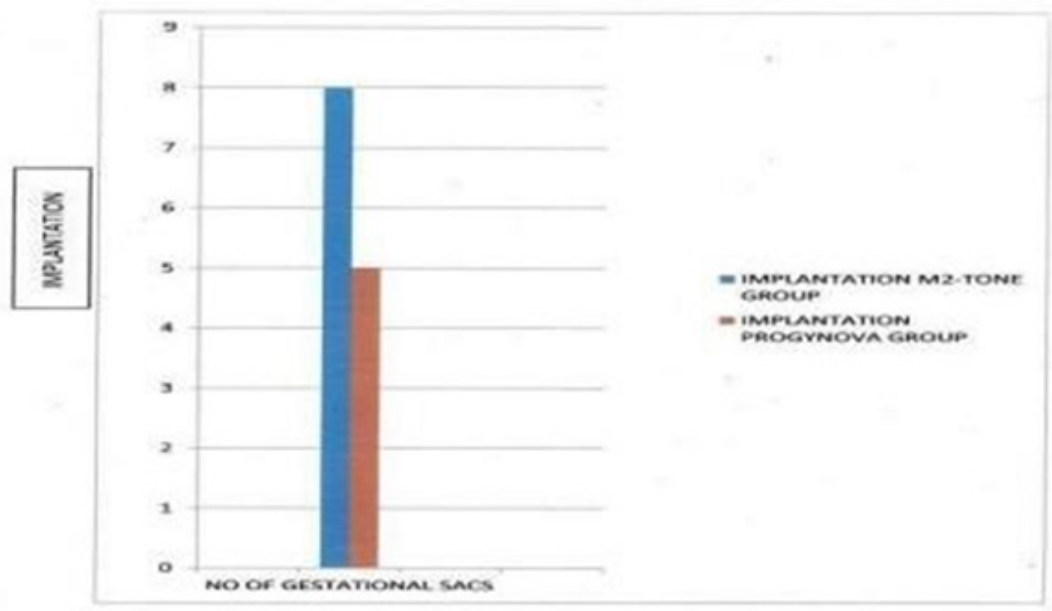

Figure 3: Effect of M2-Tone on Implantation.

Endometrial Morphology and Uterine Health

The test group showed significant improvement in endometrial thickness (Table 1). Positive Human Chorionic Gonadotropin (hCG) was 24% in the Test drug group and 16% in the control group (Tables 2 & 3).

Table 1: Effects of M2-Tone on Endometrial Lining.

Group	Baseline Endometrial Lining	Post Treatment Endometrial Lining
Control	3.5 ± 0.5	9.1± 0.81
Test drug	3.3 ± 0.5	9.9± 0.95

Table 2: Effects of M2-Tone on Pregnancy Outcome.

Pregnancy Outcome	-βHCG +βHCG
Control	84% 16%
Test drug	76% 24%

Table 3: Effects of M2-Tone on Implantation.

Implantation n Rate	NO of Gestational Sacs on Ultrasound
E2V only	5
E2V + M2-Tone	8

Discussion

In population terms, the age range 15-44 years has been chosen to represent the "reproductive age group." A noticeable decline in fertility begins between age 30 and 33 and a much steeper decline begins at 35-38 years of age for the female partner. With advancing age and/or a declining hormonal profile, concomitant with a decline in oocyte quality, the endometrium undergoes gradual involution from proliferative to inactive, eventually becoming atrophic [16]. The widespread use of contraception and many other confounding variables for reproduction in industrialised societies, and poor nutrition in developing countries, has significantly reduced our ability to establish natural fertility rates. With recent dramatic advances in infertility treatment, age related infertility remains as one of our most difficult challenges. To our current knowledge, the endometrium, among many other things, is definitely a fertility determining factor. The human endometrium is a unique tissue that undergoes sequential phases of proliferation, and secretory changes followed by tissue shedding and bleeding during menstruation. Tissue remodelling is a distinct feature of human endometrium in the secretory phase which prepares endometrium for implantation during the "receptive phase" of the cycle. The receptive endometrium is a prerequisite for conception [17].

Treatment options for infertility vary, yet the barriers of invasiveness, cost, and access inhibit treatment use for many women. Alternative medical approaches exist for this indication, and sustain certain popularity. With the introduction and integration of different medical approaches, we tried to establish a safe, effective, realistic and beneficial option for infertility. M2-Tone, a polyherbal formulation, has shown its beneficial effects on the three prerequisites (menstruation, ovum, and endometrium) for conception and the effects on endometrial vasculature are also evaluated. We aimed to evaluate the efficacy of M2-Tone tablet as an adjuvant in modifying endometrial thickness and the corresponding pregnancy rate in patients with infertility. Therefore,

we systematically studied a standard therapeutic formulation (Progynova) with polyherbal formulation (M2-Tone) to treat unexplained infertility in women. A preliminary study by Joglekar SN et al. indicated estrogenic effect of M2-Tone evidenced by increase in weight and glycogen content of uterus [18]. Pattanayak M, et al. compared the efficacy of M2-Tone with low dose aspirin and an antioxidant as an adjuvant in IVF cycles for poor responders. The results of the study demonstrated the advantage of M2-Tone over the others as far as implantation rates and live birth rates are concerned. Parulekar S, et al.; Dubey P, et al; Gupta I, et al.; Kotadawala P, et al. and Urala MS, et al. in five separate clinical studies showed improvement in menstrual irregularities [19-23]. Tanksale VS, et al. in a clinical trial showed that M2- Tone stimulates ovulation in anovulatory females [24]. Chandravati, et al. after a clinical evaluation reported that M2-Tone achieved a very good rate of conception in infertility [25]. In an clinical trial by Malhotra N, et al. M2-Tone showed significant improvement in endometrial morphology and increase in pregnancy rate [26]. An animal study conducted by Bhutani K, et al. to evaluate the efficacy and safety of Lodhra one of the main ingredient of M2-Tone revealed enhanced folliculogenesis, presence of mature follicles and detached oocytes as a result of increased FSH and LH levels [27].

Progynova contains the oestrogen oestradiol valerate, a prodrug of the natural human 17ß oestradiol. Progynova is a synthetic hormone replacement for oestrogen. It is used to build up the lining of the uterus in preparation for embryo transfer during a thaw cycle for women who do not ovulate regularly. Side effects like breast tenderness, gastric upset, nausea, headaches and an increase in body weight have been reported. This study focuses on the endometrial development towards a receptive milieu. In total, 50 patients were selected with the age group 35 - 53 years. The patients were divided in two groups (25 patients per group). First group (test drug group) received M2-Tone tablets and Progynova tablets. Second group was the control group which received Progynova tablets only. Total duration of treatment was 3 months. To measure endometrial thickness transvaginal sonography was performed before and after treatment. Transvaginal ultrasonography is an ideal non-invasive method to determine endometrial thickness in ART. The endometrial thickness during the IVF was monitored weekly with transvaginal 3.5 MHz transducer scan in millimetres.

In the M2-Tone group the endometrial thickness increased to 9.9 mm. There was an increase in pregnancy rate (24%) in the M2-Tone group as against (16%) in the control group. M2-Tone was well tolerated by patients and none of them reported any adverse effects. Thus we can conclude that M2-Tone, as an adjuvant prior to IVF procedures, increases the chances of pregnancy by improving endometrial thickness. The variable morphological response of the endometrium to endogenous hormones in the naturally cycling patients reflects either a unique physiological response to hormones at the target tissue, or an insufficiency in the amount of endogenous hormones at the level of the uterine epithelium, or both [16]. The present study analyses the epithelial response to the polyherbal formulation M2-Tone with conventional line of treatment. In summary, the M2-Tone group demonstrates significant improvement in endometrial lining and pregnancy outcome as compared to the control group. We have started a more extensive retrospective and prospective study to be able to determine the ramifications of M2-Tone on endometrial receptivity and pregnancy outcome as compared to low dose aspirin treatment [28] and vitamin E [29].

Conclusion

This study was conducted by Prof Oladapo Ashiru and his team on fifty female patients undergoing infertility treatment at Medical Art Center, an IVF clinic in Nigeria. The finding of present study suggests that M2-Tone tablets improves endometrial thickness and also shows improvement in pregnancy rate. We can conclude that M2-Tone helps endometrium attain a receptive, stimulated and morphological profile that may lead to successful implantation. Problems associated with the endogenous hormone response, coupled with the tailor regime with M2- Tone may prove beneficial in infertility cases as adjuvant or single drug therapy. Further we highlight the need for priming of the 'window of receptivity'. We suggest further study to evaluate the effect of M2-Tone considering windows of receptivity and uterine endometrium.

Acknowledgement

We would like to acknowledge the contributions of the following doctors for their participation in the earlier parts of this study; Dr Cynthia C. Okeke, Dr Oluwakemi Olukoya, Dr. Oluwakemi Ailojie, and Mrs Biola J. Adewusi.

References

1. (1983) WHO. World Health Organization: Report of the Meeting on the Prevention of Infertility at the Primary Health Care Level. WHO, Geneva 1983, WHO/ MCH/1984.
2. Cates W, Farley TM, Rowe PJ (1985) Worldwide patterns of infertility: is Africa different? Lancet 2(8455): 596-598.
3. Sule JO, P Erigbali, L Eruom (2008) Prevalence of Infertility in Women in a Southwestern Nigerian Community. African Journal of Biomedical Research 11(2008): 225-227.
4. Manners CV (1990) Endometrial assessment in a group of infertile women on stimulated cycles for IVF: immuno histochemical findings. Hum Reprod 5(2): 128-132.
5. Chiang Ch, Hsieh TT, Chang MY, Shiau CS, Hou HC, et al. (2000) Prediction of pregnancy rate of in vitro fertilization and embryo transfer in women aged 40 and over with basal uterine artery pulsatility index. J Assist Reprod Genet 17(8): 409-414.

6. Kdous M, Fadhlaoui A, Boubaker M, Atef Youssef, Anis Chaker, et al. (2007) Intrauterine insemination with conjoint semen. How to increase the success rate? Tunis Med 85(9): 781-787.
7. Jinno M, Ozaki T, Iwashita M, Y Nakamura, A Kudo, et al. (2001) Measurement of endometrium tissue blood flow: a novel way to assess uterine receptivity for implantation. Fertility and sterility 76(6): 1168-1174.
8. Thomas Strowitzki, A Germeyer, R Popovici, M von Wolff (2006) The human endometrium as a fertility-determining factor. Human Reproduction Update 12(5): 617-630.
9. Kwang Rae Kim MD (2010) Efficacy of luteal supplementation of vaginal sildenafil and oral estrogen on pregnancy rate following IVF-ET in women with a history of thin endometrial: A pilot study. Journal of Women's Medicine 3(4): 155.
10. Goswamy RK, Williams G, Steptoe PC (1988) Decreased uterine perfusion-a cause of infertility. Hum Reprod 3(8): 955-959.
11. Groutz A, Wolman I, Jaffa A, I Yovel, A Amit (1997) Influence of ovulation induction with human menopausal gonadotropin on uterine blood flow. Comparison of unexplained and mechanical infertility. J Ultrasound Med 16(7): 455-458.
12. Goswamy RK, Steptoe PC (1988) Doppler ultrasound studies of the uterine artery in spontaneous ovarian cycles. Hum Reprod 3(6): 721-726.
13. Perrot Applanat M, Groyer Picard MT, Garcia E, F Lorenzo, E Milgrom (1988) Immunocytochemical demonstration of oestrogen and progesterone receptors in muscle cells of uterine arteries in rabbits and humans. Endocrinology 123(3): 1511-1519.
14. Jongbae J Park, Myungja Kang, Sangseop Shin, Eunmi Choi, Sukyung Kwon, et al. (2010) Unexplained Infertility Treated with Acupuncture and Herbal Medicine in Korea. J Altern Complement Med 16(2): 193-198.
15. Narendra Malhotra, Jaideep Malhotra, JP Rao, Shobhna Arora, Neharika Malhotra, et al. (2010) Randomized placebo-controlled study to evaluate the role of M2- TONE. as an adjuvant treatment in IUI protocol by assessing its effects on endometrial morphology, vasculature and pregnancy rate using 3 D Colour Doppler in female infertility. SAFOG 2(2): 115-122.
16. Susan M Adams, Vera Terry, Margot J Hosie, Nalini Gayer, Christopher R Murphy (2004) Endometrial response to IVF hormonal manipulation: Comparative analysis of menopausal, down regulated and natural cycles. Reproductive Biology and Endocrinology 2: 21.
17. Siamak Tabibzadeh (2002) Decoding implantation and menstruation: the tale of two opposing signals. Frontiers in Bioscience 7: 1475-1486.
18. Joglekar SN (1982) Estrogenic Activity of 'M2 Tone' An Indigenous Drug preparation. Current Medical Practice 26(3): 108-111.
19. Parulekar VS (2001) Evaluation of M2-Tonesyrup in functional menstrual disorders. Data on file.
20. Dubey P (2001) Clinical Evaluation of M2-Tonein Adolescent Menstrual Disorders. Advances in Obsterics & Gynecology 1(50): 293-298.
21. Gail B Slap (2000) Menstrual disorders in Adolescents. Obs & Gynae 17(1): 75-92.
22. Kotadawala P (2004) Evaluation of M2-Tonein menstrual disorders of adolescents and young adults. Int J Gynecol Obstet India 7(2).
23. Urala Ms (1999) Role of M2 - Tone Syrup In Menstrual Disturbance of Reproductive Age Group Women. Obs & Gyne 4(7): 469-471.
24. Tanksale VS (1999) M2 - TONE Syrup in Treatment of Unexplained Infertility in Females. Obs & Gynae 4(2): 105-110.
25. Chandravati (1999) Clinical Evaluation of M2-TONE in Unexplained Infertility. Obstetrics & Gynecology Communications 1(6): 50-54.
26. Malhotra N (2000) Endometrial scoring for prediction of implantation-Evaluation of drug therapy. Obstetrics & Gynecology Communications 2(1): 51-57.
27. Bhutani KK, Atul Jadhav, Vandana Kalia (2004) Effect of Symplocos racemosa Roxb on gonadotropin release in immature female rats and ovarian histology. Journal of Ethnopharmacology 94(1): 197-200.
28. Zhang X, Guo F, Wang Q, Bai W, Zhao A (2022) Low-dose aspirin treatment improves endometrial receptivity in the midluteal phase in unexplained recurrent implantation failure. International Journal of Gynecology & Obstetrics 156(2): 225-230.
29. Mohd Mutalip SS, Ab Rahim S, Rajikin MH (2018) Vitamin E as an antioxidant in female reproductive health. Antioxidants 7(2): 22.

ISSN: 2574-1241

DOI: 10.26717/BJSTR.2022.45.007142

Oladapo Ashiru. Biomed J Sci & Tech Res