

Development Of Design And Dermatological Assessment Of A Unique Herbal Formulation For The Possible Treatment Of Postmenopausal Syndrome

Abbaraju Lakshmi Harini

Research Scholar, School of pharmacy, Sabarmati University, India.

Dr Yashoda Krishna

Professor, School of pharmacy, Sabarmati University, India.

Dr. Sujit Kumar Mohanty

Professor, Department of Pharmaceutical Chemistry, Shri Vishnu College Of Pharmacy, Bhimavaram, India.

Abstract : During the years leading up to, during, and after menopause, levels of reproductive hormones, especially estrogen, drop below the standards that are considered normal. During menopause, the menstrual cycle comes to an end and remains that way permanently. It is important to note that the problems that develop during these times have a substantial influence on the quality of life (QoL) of women. Osteoporosis and postmenopausal obesity are two conditions that may make a variety of metabolic diseases much worse. In addition to enduring the consequences of aging, these folks are also going through hormonal shifts, which makes an already challenging situation much more challenging for them. When it comes to this particular field, nevertheless, new kinds of help are necessary. Hormone therapy is a tried-and-true approach for treating these illnesses; but, it is not without its hazards, especially when it is used for a prolonged length of time. In order for a pharmacological treatment to be successful, it is necessary for patients to be ready to accept the clinical interpretation. This line of thinking is congruent with the manner in which the participants in this research were questioned about the ways by which they planned to receive the drugs. In this particular technique, market research was carried out via the use of a survey that was based on questionnaires. The themes of an organized questionnaire that was produced and piloted included menopausal symptoms, treatment views, sociodemographics, menopausal hormonal therapy, menopause knowledge, information sources, care, and worries. The questionnaire was first designed and piloted. The manifestation of menopausal symptoms may be seen in a woman's bodily health, mental health, and sexual health, as our research has shown. There are a number of elements that may have an effect on the intensity of symptoms, including your nutrition, social circle, educational background, information sources, geographical location, and general health. According to the findings of the study, women were more likely to use herbal medicines and were prepared to agree to delivery techniques that did not include oral administration.

Keywords : postmenopausal obesity, QOL, Osteoporosis, social circle, hormonal shifts.

1. INTRODUCTION

Given that women make up half of the world's population, it is imperative that they be given priority (Gender split by global area, 2021). The subspecialty of medicine known as "women's health" is primarily concerned with the diagnosis and treatment of health disorders that are unique to women, both emotionally and physically. The topics of heart illness, breast cancer, premenstrual syndrome (PMS), pregnancy, birth control, menopause, and a great deal of other topics are discussed in great detail. Menopause is a significant and unavoidable health problem that women go through as they reach the end of their reproductive years. Certain women may experience an improvement in their quality of life (QoL) after they have stopped menstruation, while others may experience a reduction in their QoL. As menopause starts, it is common for a great deal of worry to creep in. It is usual for women to have these problems, and since they are thought to be unavoidable, they generally learn to deal with them for themselves. Whiteley et al. (2013) discovered that women experience a considerable decrease in their productivity after menopause as a result of changes in their health. Considering that the majority of academics, doctors, and people do not pay much attention to the problem because they believe it happens organically, the fact that it is so intriguing is even more fascinating. Countries like ours, where menopause is a source of health concerns and leads women to lose emotional, physical, and financial support, need to give this subject greater attention since it has the potential to produce sudden and serious problems. There are certain countries where it is well received by a huge number of people, while in others it is not observed at all. This element of the condition has not yet gotten sufficient attention, despite the fact that it is very significant and that the implications it may have might be quite severe. It is imperative that research into and treatment of these symptoms continue to be conducted on a worldwide basis even as the average lifespan continues to rise and the percentage of women who are experiencing menopause continues to rise. According to the findings of earlier studies, this will be a significant public health issue by the year 2030, with the severity of the problem being second only to that of ischemic heart disease (Reddy, 2010).

Our healthcare system is now striving toward the goal of increasing quality of life as it transitions from a biological worldview that is centered on illness to one that focuses on the complete person and their health. According to Megari (2013), an improved quality of life is especially crucial for those who are dealing with chronic diseases such as menopause. A remark was issued by Shofany in the year 2017. There are a variety of symptoms that may be experienced by a woman who is going through or has gone through menopause. These symptoms can have a negative impact on our quality of life. In spite of the fact that issues about quality of life that are associated with menopause are a big worry in developed countries, a significant number of people still do not give a hoot about them. In light of this, it is essential to work toward bettering one's health in connection to this matter. In order to improve the quality of life of menopausal women, it is imperative that all parties concerned, not only medical and governmental organizations, make an

effort to improve the situation. Without having a significant amount of medical expertise, it is impossible for anybody to do this, as stated by the World Health Organization (WHO). Quality of life (QoL) is now a topic that is receiving a lot of attention, and there is a lot of literature on the issue.

The widespread lack of understanding and recognition of menopause as a valid medical condition is one of the most significant challenges that must be overcome in order to succeed in the development of an effective instrument to relieve menopausal symptoms. In light of this, it may be deduced that the relevant organizations will get information on public polls in the near future. In this day and age, surveys are a reliable technique for gathering the information that you want, and they are also an effective method for setting the framework for specific study in any sector. A questionnaire-based survey is a method of gathering information on people's habits, attitudes, or behaviors (Preston, 2009). This method makes use of a certain set of questions that are organized. Surveys are used to collect this data, which may be quantitative or qualitative in character, depending on the context. When it comes to conducting surveys, there is a wide variety of alternatives available, such as in-person, online, over the phone, via the mail, and so on. They provide researchers with the opportunity to engage in conversation with a wide range of persons and get insight into the ways in which people's social and physical settings influence the behaviors they exhibit. Through the use of statistical analysis, the goal is to collect information and arrive at a result that is worth considering. In order for the results to give information that can be relied upon on the subject matter that the questionnaire was designed for, it is essential that the process of developing the questionnaire and selecting the respondents be carried out with a great deal of care. After taking into account the research problem, the tools that are available, the number of groups, and the kind of group, the questionnaire should be designed. The fact that responders are allowed to fill out the form in an anonymous manner contributes to the good results that this approach produces. With the help of this tactic, we are able to quickly gather a great deal of information.

A plan to relieve concerns associated to menopause was devised based on the outcomes of an online poll that was conducted throughout the process. As the foundation of this research endeavor, an exceptional methodology that is based on design thinking and places the patient as the primary focus is essential. Despite the fact that the currently available treatment tools have a number of pharmacotechnical and therapeutic problems, the technique that has been offered has the ability to solve a problem that has been disregarded for a long time that is relevant to the health of women. It is essential to have improved targeting since it has a systemic influence, the objective is to have fewer side effects with few or no interactions with other medications, and it has a larger bioavailability without compromising the safety of the patient who is being treated.

2. LITERATURE REVIEW

Recent advances in pharmaceutical formulations have led to the development of innovative delivery systems to improve drug bioavailability and therapeutic outcomes. Ainbinder and Tuitou (2005) developed an ethosomal testosterone formulation that demonstrated superior transdermal absorption compared to conventional patches, offering a promising alternative for hormone replacement therapy. Similarly, Fu et al. (2019) designed ethosomes for macromolecular protein delivery, which not only enhanced percutaneous absorption but also accelerated wound healing in murine models.

For dermatological applications, Sguizzato et al. (2020) formulated Coenzyme Q10-loaded ethosomes that provided significant protection against oxidative stress in human skin fibroblasts and 3D epidermis models. Anjum et al. (2020) developed a naproxen sodium ethosomal gel that exhibited potent anti-inflammatory effects in a rat model of rheumatoid arthritis, suggesting its potential as an improved topical therapy.

Nanostructured lipid carriers (NLCs) have also shown promise in dermatological drug delivery. Imran et al. (2020) created a dual-drug NLC gel containing quercetin and resveratrol, which enhanced epidermal and dermal penetration while demonstrating cytotoxic effects against skin cancer cells.

Polymer-based delivery systems have been explored for sustained drug release. Avinash et al. (2013) developed an inter-polymer complex between Carbopol 971P and PVP K90 for acyclovir delivery, which exhibited excellent mucoadhesive properties and controlled release kinetics. Shivhare et al. (2009) formulated sustained-release aceclofenac tablets using carboxypolyethylene polymers that maintained therapeutic levels for 24 hours.

Ethosomal systems have proven particularly versatile. Jain et al. (2007) demonstrated that lamivudine-loaded ethosomes altered stratum corneum structure to enhance drug permeation, while Prasad et al. (2016) developed a proniosomal transgel system for pioglitazone that significantly improved transdermal delivery and antidiabetic efficacy compared to oral tablets.

Quality by Design (QbD) principles have been applied to optimize formulation development. Waghule et al. (2021) detailed a QbD approach for liquid crystalline nanoparticles, providing a regulatory-compliant framework for reproducible manufacturing. Abdallah et al. (2021) incorporated jojoba oil into brucine-loaded liposomal emulgels, significantly enhancing anti-inflammatory effects for topical applications.

These studies collectively demonstrate the potential of advanced drug delivery systems to overcome bioavailability challenges, enhance therapeutic efficacy, and provide targeted treatment options across various medical conditions. The integration of novel carriers with quality-focused development approaches promises to revolutionize pharmaceutical formulations in coming years.

3. EXPERIMENTAL WORK

The list of instruments used in this facility includes various high-precision equipment from well-known manufacturers worldwide. These include an electronic weighing balance by Mettler Toledo from Langacher, Switzerland, and a refrigerated centrifuge (Tomy MX-305,

High-Speed Micro Centrifuge) from Japan. For particle size analysis, the Zeta Sizer from Malvern Instruments, UK, is utilized, while differential scanning calorimetry (DSC) is performed using a Perkin Elmer Inc. system from Waltham, MA, USA. The facility also employs a UV-Visible spectrophotometer (UV-1601) by Shimadzu, Japan, and a high-performance thin-layer chromatography (HPTLC) system from CAMAG, Muttenz, Switzerland. For mass spectrometry and chromatography, a Waters Corp. UPLC-MS system, with serial numbers F09 UPB 920M and JAA 272, is used, along with an FTIR spectrophotometer from Perkin Elmer, Germany. In microscopy, the laboratory utilizes advanced instruments such as the Tecnai G20 Transmission Electron Microscope from FEI, Netherlands, and a Zeiss Confocal Laser Scanning Microscope from Heidelberg, Germany. A fluorescence microscope from Life Technologies (EVOS FLc) and an Olympus inverted light microscope (ix71) from Tokyo, Japan, are also available. For sample processing, the lab relies on equipment like a mechanical plate shaker (MixMate® by Eppendorf, Germany), an ELISA plate reader (Synergy HT by BioTek, USA), and various laboratory accessories such as a magnetic stirrer (Tarsons Products Pvt Ltd, India) and a CO2 incubator by Thermo Scientific, USA. Other essential equipment includes a refrigerator from Samsung, an autoclave (Biogene, India), a hot air oven (Widsons Scientific Work, India), a texture analyzer from Stable Micro Systems Ltd., UK, and a rheometer (Physica MCR 101 Anton Parr Rheometer). Furthermore, common laboratory consumables and accessories, such as micropipettes (Eppendorf, India), pH meters (Mettler Toledo, Switzerland), vortex mixers (Nirmal International, India), and syringe filters (Axiva-Nylon, AxivaScichem Biotech Pvt, USA), are readily available. Dialysis membranes (MWCO 10-12 kDa, Himedia, India), ELISA plates (Thermo Fisher Scientific, USA), TLC silica gel (Merck KGaA, Germany), and centrifugal filter devices (Millipore, USA) are also part of the equipment inventory, alongside various syringes, sutures, and local vendors' supplies.

A. To evaluate the health status and needs of adult women, we developed a quality of life questionnaire that is particular to menopause.

Prelude: Our healthcare system is now striving toward the goal of increasing quality of life as it transitions from a biological worldview that is centered on illness to one that focuses on the complete person and their health. There is a pressing need to improve quality of life, particularly in the case of protracted illnesses such as menopausal disease. There are a variety of symptoms that may be experienced by a woman who is going through or has gone through menopause. These symptoms can have a negative impact on our quality of life. Therefore, it is essential for individuals to make preparations for climate change and to ensure that they are cognizant of this era of transition. In order to better manage these changes, it is beneficial for individuals to have a thorough understanding of menopause, its symptoms, and the several diets and treatments that have the potential to reduce those problems. When one takes into account the age group in issue as well as the health risks that are linked with it, it seems that effective health management is essential. In order to improve the quality of life of menopausal women, it is imperative that all parties concerned, not only medical and governmental organizations, make an effort to improve the situation. This could only be accomplished by someone who has a substantial amount of medical understanding. We wanted to know how women in a specific area of Delhi, India, felt about menopause, how common symptoms were, what they liked and didn't like about different methods of dealing with it, if there were any connections or issues, and how to bring attention to this neglected health issue through this questionnaire study. This survey was developed with the purpose of bringing attention to concerns that are associated with menopause and have an impact on quality of life.

B. Quality of life evaluation using menopausal symptoms as measured by

questionnaire responders

Within the scope of the assessment, an extra focused inquiry was included in order to ascertain whether or not the individual was self-aware of the symptoms. Everyone did their best to come up with any sign that may possibly have even a little adverse effect on the quality of life the individual is experiencing. Most women experience hot flashes (118/59%), sleep loss (151/75.5%), depression (149/74.5%), anxiety or nervousness (138/69%), mood swings (162/81%), weight gain 156/78%, irritation 153 (76.5%), decrease in physical strength 173 (86.5%), muscle pain 168 (84%), joint pain 169 (84.5%), vaginal dryness 133 (66.5%), vaginal itching 131 (63.5%), and less sexual desire than usual (127/63.5%). Both Table 1 and Table 2 provide more information on the descriptive data and the Spearman's association matrix that were obtained from the MENQOL.

Table 1. Personality traits related to MENQOL domains.

*Statistically significant, p-value calculated by one-way analysis of variance (ANOVA).

Domain	Perimenopausal Mean (SD) 95% CI	Postmenopausal Mean (SD) 95% CI	MENQOL Mean (SD)	%	p- Value
Vasomotor	2.08 (1.12) (1.97-2.19)	1.94 (0.97) (1.80-2.08)	2.03 (1.07)	59.6	0.141
Psychosocial	2.34 (1.07) (2.28-2.41)	2.20 (1.02) (2.10-2.29)	2.29 (1.01)	76	0.915
Physical	2.29 (1.01) (2.25-2.34)	2.16 (2.03) (2.09-2.23)	2.25 (1.02)	72.9 6	0.001 *
Sexual	2.07 (0.96) (2.01-2.13)	1.85 (1.0) (1.75-1.94)	2.0 (0.98)	62.4 3	0.023 *
All domains	2.81 (0.86) (2.78-2.84)	2.68 (0.86) (2.62-2.73)	2.78 (0.86)	69.8 7	0.019 *

Table 2. Spearman's correlation matrix: symptom presence/absence served as the basis for the calculation.

Domain	Vasomotor	Psychosocial	Physical	Sexual
Vasomotor	1.000	0.682**	0.414**	0.786**
Psychosocial	0.682**	1.000	0.588**	0.849**
Physical	0.414**	0.588**	1.000	0.517**
Sexual	0.786	0.849**	0.517**	1.000

C. Patients' present treatment status and outlook on therapy

The data showed that 44 respondents, or 22% of the total, were receiving treatment. A majority of these individuals were using herbal medicines to address their issues, accounting for 33% or 55%. In addition, 26 ladies (12.68%) seemed unaware of the various therapy options. Herbal remedies for menopause were, however, well-received by 108 (52.68%) of the responding women. In addition, 79 participants (38.54%) had bad things to say about hormone therapy. A total of 78 respondents (38.05%) said they would consume the goods orally, while 90 respondents (43.90%) indicated transdermal application would be their preferred method of administration. Many respondents cited their spouses, close friends and family, and other loved ones as the primary sources of support during this challenging period.

D. Key Findings

According to our study, the onset of menopause occurred at an average age of 51.96 years, with a standard deviation of 8.23 years. Changes occur in women's bodies, brains, and sexuality during the menopause (also known as the transitional period). However, the severity of the symptoms may also be affected by other factors, such as the individual's culture, level of education, information levels, geography, and level of physical fitness. Research should be conducted to determine the reasons why perimenopausal women feel a little higher level of pain compared to postmenopausal women. Only a small percentage of women who were sick were really going to therapy sessions. It is becoming increasingly common for women to look for natural remedies, and this research lends support to that trend. When asked about hormone replacement therapy (HRT), respondents provided responses that were contradictory to one another. Transdermal dosage forms were also more popular among women than oral dose forms, perhaps because the former were believed to be easier to administer than the latter.

E. In vitro drug release study

Due to the fact that it proves the effectiveness of a product in delivering a drug (TEF in this particular case) via the utilization of a certain carrier system, this study is of great significance. For the purpose of determining the total quantity of TEF, samples

were taken from the release medium at defined intervals ranging from 0 to 28 hours. These samples were then compared to a control sample consisting of pure release media at a particular wavelength. Plots were created utilizing the drug release rates for both the traditional gel and the ethosomal gel. These plots were used to determine the relationship between the time gap and the overall percentage of drug released. As demonstrated in Figure 4.23, the releases are slower and continue for a longer period of time for ethosomal gel after an initial burst release of 22.7%, but the releases for conventional gel persist longer after 7.8% has been released. It is likely that this two-stage release pattern may be explained by the fact that surface-bound TEF releases rapidly, but ethosome-bound TEF travels slowly across the lipid bilayer, which impedes and limits the release of the TEF. A regulated release profile, with an initial fast release, is required for the proper distribution of topical drugs into the circulation. This is done in order to maintain a high concentration gradient between the two solutions. Additionally, after a period of twenty-four hours, the ethosomal gel released TEF at an average rate of 83%, whereas the conventional gel only released it at a rate of 51%. By aiding the ethosomal vesicles' penetration into the gel matrix, the presence of ethosomal vesicles in the gel matrix may be responsible for the greater rate of ethosomal gel release. An additional finding was the discovery of a correlation coefficient (R^2) between the release rates of the mixture and the Peppas model.

Prasad et al. (2016) and Paliwal et al. (2019) reported that the values for normal gel and ethosomal gel were 0.933 and 0.950, respectively, and that the release curve was in conformity with the Fickian diffusion model. Both of these studies were published in the United States. There were multiple release kinetic models for ethosomal gel that were depicted in Figure 4.24. These models included zero-order, first-order, Higuchi, and Korsmeyer Peppas models, all of which provided support for the findings.

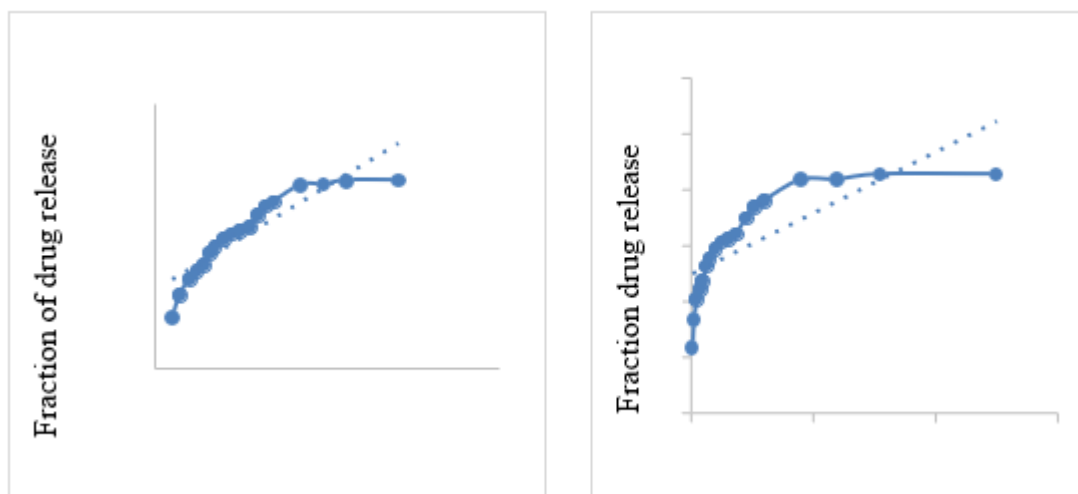


Figure 1. In vitro release study profile of TEF-loaded conventional gel and TEF-loaded ethosomal gel. Each study was performed in triplicate and data are shown as mean \pm SD. A comparison of the release profile of both gels is shown with an initial burst release followed by a sustained release pattern

4. Findings and analysis

➤ Evaluation of data

The outcomes consist of the averages together with the standard deviations (SDs) that correspond to each of them. In this particular investigation, we examined the results by using a one-way analysis of variance (ANOVA) and, if appropriate, Tukey's post hoc test for multiple comparisons. A p-value that was less than 0.05 indicated that there was a change that was statistically significant.

➤ The pathology report

Figure 4.31 shows a collection of photomicrographs of rat vaginal smears that depict the four phases of the estrous cycle in a Wistar rat that is in good condition. During the course of our investigation, we were able to recognize four unique stages: the proestrous, the estrous, the metestrous, and the diestrous phases. As can be seen in Figure 2, the vaginal smear images that were collected from day one to day four demonstrate that the rats who had their ovaries removed (OVX) did not have an estrous cycle and only had the diestrus phase remaining. According to Lotz and Zangrossi (2021), the areas of the cell smear that were examined indicated a significant quantity of mucus cells, white blood cells, and a limited number of nucleated epithelial cells. Figure 3 presents a comparison of the results of the vaginal smear performed on the participants in the varied training groups. An increased number of pus cells and white blood cells were seen in the OVX group, despite the fact that the number of nucleated and cornified cells was lower. In contrast, the hypothetical control group demonstrated the metestrous phase of the reproductive cycle. When it came to the CRE group, which showed a completely different pattern, the predominant cells were cornified cells that were devoid of leucocytes and pus cells. On the other hand, the pattern that was seen in the SHAM group was observed in both the CRO and CRT groups. According to Montes and Luque (1988), one of the probable explanations is that the estrogen levels in the rats were found to be too high.

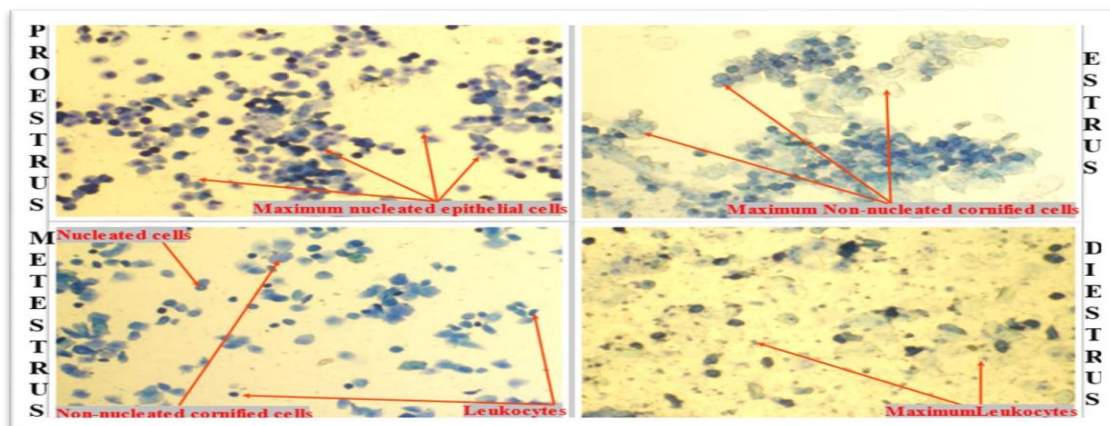


Figure 2. Images captured under a 40X microscope of a rat's vaginal smear reveal the four stages of the estrous cycle in a robust Wistar rat. Nucleated epithelial cells predominate during the proestrous phase. Estrous phase cells: cornified, non-nucleated, During the menstrual

period, neutrophils, non-nucleated cornified cells, and nucleated epithelial cells Death stage: white blood cells

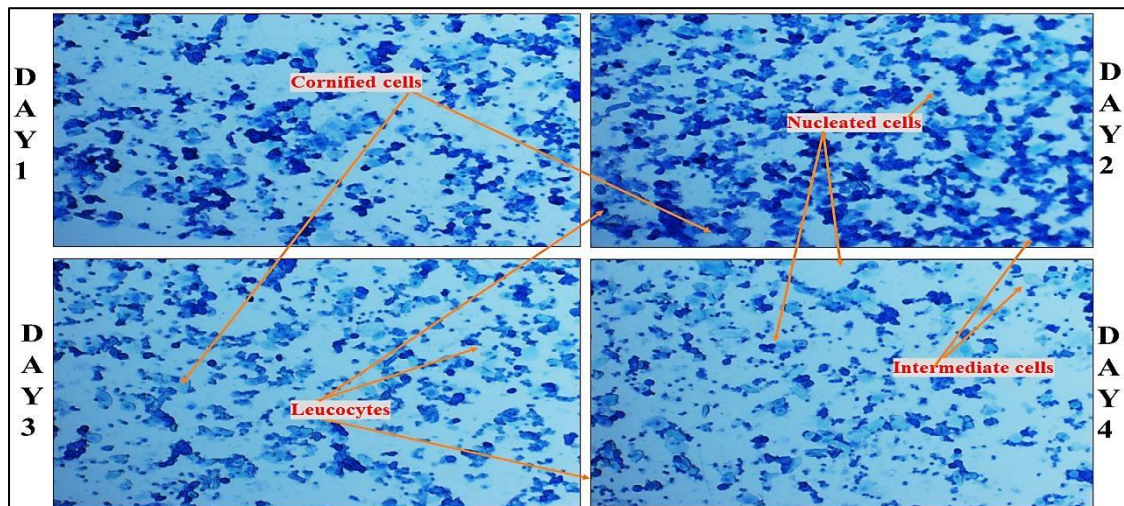


Figure 3. The photomicrographs show the vaginal smear of the OVX rats from the first day of the five-day trial, which included vaginal cell smear. Despite the lack of an apparent estrous cycle after ovariectomy, the cell smears revealed an abundance of leukocytes, mucus cells, and a small number of nucleated epithelial cells, indicating the rat's diestrus phase during all four days.

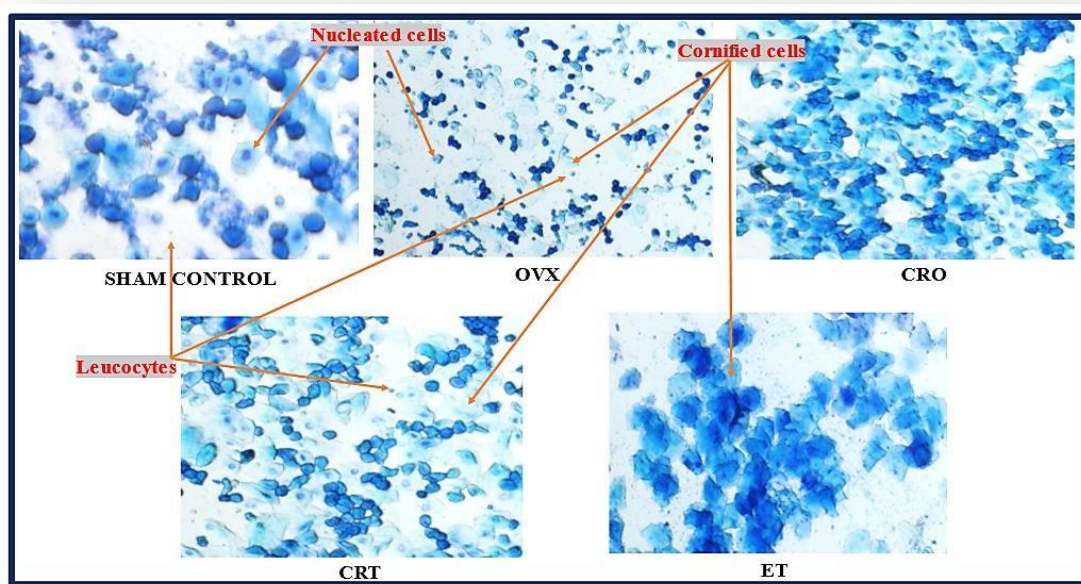


Figure 4. Images of vaginal smears taken under a microscope showing the pattern of vaginal cell smears in various intervention groups.

In comparison to the fake control group, the OVX group exhibited a lower number of nucleated and cornified cells (diestrous phase), as well as a greater number of pus cells and leucocytes for examination. The CRE gr, on the other hand, showed a high number of cornified cells during the whole estrous phase, but leucocytes and pus cells were not present inside the sample. In addition to the CRO gr and CRT groups, the sham group also exhibited the metaestrous phase of the reproductive process.

The photomicrographs of animal vaginal smears showed that the vaginal cells had become more cornified as a result of transdermal estrogen treatment. This was proven by the lack of pus cells and white blood cells. There is a possibility that the increased estrogen levels in the rat led to the development of vaginal walls that were bigger and more cornified. On the other hand, the cornification was slightly less severe in the groups who were given CR either oral or transdermal administration method. In light of this, it is possible that CR does not have an estrogenic effect.

Conclusion

According to her, "Communities, countries, and ultimately the world are only as strong as the health of their women." Because women make up half of the world's population, their health must be a top priority. When women go through menopause and the health problems that come with it, they are less able to help out in society. Because this problem happens so often, the general public, schools, and doctors and nurses nearly never ignore it. Women in our area experience a loss in mental and physical health due to menopause; hence, further study on this subject is required. There must be a way to alleviate menopausal symptoms so that women may take greater pleasure in this normal life transition, as they make daily living more difficult for women.

Menopausal symptoms may be effectively treated with MHT. But a lot of damage will still be done by this. With the use of nanotechnology, we set out to make a plant gel that may ease menopausal symptoms throughout their several stages.

The public at large often assumes that a treatment's efficacy is heavily influenced by factors including its cost, availability, and public perception of it. The product has been evaluated and approved by well-known professionals, in this case gynecologists, so you can be sure it is of high quality. We continued along this line of thinking by asking doctors and patients how they planned to treat the problem using medicine, because this was seen to be the most likely solution. Questionnaires provided the bulk of the data used in this process.

References

- 1) Abdallah, M.H., Elsewedy, H.S., Abulila, A.S., Almansour, K., Unissa, R., Elghamry, H.A., Soliman, M.S., 2021. Quality by design for optimizing a novel liposomal jojoba oil-based emulgel to ameliorate the anti-inflammatory effect of brucine. Gels. <https://doi.org/10.3390/gels7040219>
- 2) Abdulbaqi, I.M., Darwis, Y., Khan, N.A.K., Assi, R.A., Khan, A.A., 2016. Ethosomal nanocarriers: The impact of constituents and formulation techniques on ethosomal properties, in vivo studies, and clinical trials. International Journal of Nanomedicine. <https://doi.org/10.2147/IJN.S105016>

- 3) Aghamiri, V., Mirghafourvand, M., Mohammad-Alizadeh-Charandabi, S., Nazemiyeh, H., 2016. The effect of Hop (*Humulus lupulus* L.) on early menopausal symptoms and hot flashes: A randomized placebo-controlled trial. *Complementary Therapies in Clinical Practice*. <https://doi.org/10.1016/j.ctcp.2015.05.001>
- 4) Ahmed, M.G., Kapoor, C., A, S., 2017. FORMULATION AND EVALUATION OF ORAL SUSTAINED IN SITU GELLING SYSTEM OF ROXATIDINE. *INDONESIAN JOURNAL OF PHARMACY*. <https://doi.org/10.14499/indonesianjpharm28iss3pp178>
- 5) Ahuja, M., 2016. Age of menopause and determinants of menopause age: A PAN India survey by IMS. *Journal of Mid-Life Health*. <https://doi.org/10.4103/0976-7800.191012>
- 6) Ainbinder, D., Touitou, E., 2005. Testosterone ethosomes for enhanced transdermal delivery. *Drug Delivery: Journal of Delivery and Targeting of Therapeutic Agents*. <https://doi.org/10.1080/10717540500176910>
- 7) Akter, M.J., Shirin, E., 2018. Latest Evidence on Using Hormone Replacement Therapy in the Menopause. *Journal of Bangladesh College of Physicians and Surgeons*. <https://doi.org/10.3329/jbcps.v36i1.35508>
- 8) Albertson, A.J., Skinner, D.C., 2009. A novel animal model to study hot flashes: No effect of gonadotropin-releasing hormone. *Menopause*. <https://doi.org/10.1097/gme.0b013e3181a9f60b>
- 9) Alshaer, W., Nsairat, H., Lafi, Z., Hourani, O.M., Al-Kadash, A., Esawi, E., Alkilany, A.M., 2023. Quality by Design Approach in Liposomal Formulations: Robust Product Development. *Molecules*. <https://doi.org/10.3390/molecules28010010>
- 10) Alves, D.L., Lima, S.M.R.R., da Silva, C.R., Galvão, M.A.L., Shanaider, A., de Almeida Prado, R.A., Aoki, T., 2008. Effects of *Trifolium pratense* and *Cimicifuga racemosa* on the endometrium of wistar rats. *Maturitas*. <https://doi.org/10.1016/j.maturitas.2008.08.006>
- 11) Anjum, F., Zakir, F., Verma, D., Aqil, M., Singh, M., Jain, P., Mirza, M.A., Anwer, M.K., Iqbal, Z., 2020. Exploration of Nanoethosomal Transgel of Naproxen Sodium for the Treatment of Arthritis. *Current Drug Delivery*. <https://doi.org/10.2174/1567201817666200724170203>
- 12) Ankli, A., Reich, E., Steiner, M., 2008. Rapid high-performance thin- layer chromatographic method for detection of 5% adulteration of black cohosh with *Cimicifuga foetida*, *C. heracleifolia*, *C. dahurica*, or *C. americana*. *Journal of AOAC International*. <https://doi.org/10.1093/jaoac/91.6.1257>.