Organoleptic, pH Stability, Viscosity, and Sterility of Sonohysterosalpingography hydroxy propyl cellulose-based gel (As an alternative media for examination of *Hystero-Foam Sonosalpingography)*

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ABSTRACT

Tubal evaluation is an important step in female infertillity investigation. Sonohysterosalpingography is a method to evaluate tubal patenct by using ultrasonography. This method requires media distention. Saline is commonly use but have several limitation. Gel is an alternative to saline. Technique that use gel as media distention is known as Hystero-Foam Salpingography (HyFoSy), a technique that recently developed. Gel is the best media distention because its more stabile and last longer in the tube. Sterile gel that available now developed from hydroxyetil celullose, but this product still have to be imported with high cost. Hydroxypropil celulose is an alternative for it, it is much common in Indonesia and the price is cheaper. In preliminary study 3 formulations of hydroxypropil celulose was designed. Comparison between formulas concerning physical appereance, pH level, viscosity and sterility was anallized.

This research was designed as a laboratoric experiment to compare three different gel formulation. Evaluation concerning changes in physical appereance , pH level and viscocity was observed for 42 days. Sterile condition was observed for 12 days. The differences between data is anallized by ANAVA and correlation between variable was studied.

In 42 days of observation there were no changes in consistency, color and odor of all formulas. pH level was different among formulas ($p<0,05$) and pH level decreased during the observation but still in the range of original product pH level. The viscosity also different among formula ($p<0,05$) and decrease during observation. There is no different in sterilization result.

Among three formulas, the first formula that consist of 3,5 % hydroxypropil cellulose, 5 % gliserol and aquabidestilata is the best formula close to inovator gel.

Key Words : Hydroxypropil cellulosa, Sonohysterosalpingography, Sterile gel

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INTRODUCTION

Tubal abnormality is one of the main causes of infertility due to relatively high incidence in women subfertil, which is about 30-35%.[1] Therefore, the evaluation of tubal patency is an important part of a screening protocol for the cause of infertility in women. There are a wide variety of tube inspection techniques are constantly being developed to find a standard diagnostic tests that can provide accurate results, reliable, easy to do and at a low cost.[1, 2]

Tubal evaluation techniques available today considered quite accurate but each still has many lacks. For example chromopertubation at laparoscopy is regarded as the gold standard for evaluation of tubal still constrained due to be carried out in a facility that has specialized laparoscopic equipment, specially trained personnel for laparoscopy, requiring general or regional anesthesia, the risk of surgery or complications and the cost is quite expensive to be used first-line evaluation of tubal abnormalities.[3] As a second alternative is Hysterosalpingography (HSG), the procedure is considered as an effective screening method for the evaluation of tubal patency and the internal architecture of the cavum uteri. The drawback is the HSG does not provide information about the condition of the myometrium and ovarian morphology, making it less than ideal as the basis of a comprehensive evaluation procedure regarding the causes of female infertility. Although HSG is considered safe, but this procedure makes the patients had radiation and the possibility of allergy to contrast media used.[4]

Hysterosalpingo-contrast sonography (HyCoSy) or also known as saline-infusion sonohysterosalpingography (SIS) is a technique that was developed by combining the basic techniques of HSG with ultrasound imaging.[5] This technique was developed in the last 10 years, including in Indonesia and has been regarded as a technique that can be used as first-line evaluation of the basic causes of infertility in women. The advantages of this technique is able to provide a fairly complete data on the condition of the uterine cavity and ovarian morphology in addition to the condition of tubal patency.[6] HyCoSy method has been widely studied in various scientific studies and meta-analyzes. The conclusion that can be drawn is this technique has proven to be a reliable method, efficient and can be an alternative HSG.[7] In addition, this method is considered simple, safe and can be performed as a clinic procedure that can add value to the benefit of the initial screening protocol to the cause of infertility in women.[5] Disadvantages of HyCoSy is on the imaging side of the tube, tubal patency detection is sometimes difficult because of the structure of the tube is not easily detected using ultrasound. It often happens that the patency of the tube is not known directly, but indirectly detected from the accumulation of free fluids in the Douglas cavity. This relates to the physical properties of the saline fluid flowing fast and can not survive long in the tube. In addition, saline give hypoechoic appearance on ultrasound, that sometimes not visible on ultrasound imaging.[2] This weakness led to the examiner sometimes can not determine with certainty tubes which are impaired.

Has recently developed a new technique to improve the diagnosis of HyCoSy using sterile gel as a contrast medium.[8] This technique utilizes air dispersion beaten in the gel as an alternative to saline. Air dispersion formed in the gel is used as a better natural contrast media because it gives a hyperechoic overview on ultrasound imaging that more easily observed. Another advantage of the gel as a contrast medium in the evaluation of tubal patency is able to produce distended uterine cavity and fallopian more stable, thus allowing the observation can be made longer. This technique is called by name by the innovator Hystero-Foam Sonosalpingography (HyFoSy).[9] The new technique was developed about two years. The first report on this new technique published in 2011 and received fairly extensive attention.

The problem is the use of this technique requires a sterile gel medium HyFoSy which still have to be imported from the country of origin so that the supply is still difficult and expensive. Based on data from the manufacturing of sterile gel medium used for HyFoSy containing 4% hydroxyethyl cellulose, 3% glycerol were added aquabidest up to 100%. This preparation has a

pH of 6-7 and a viscosity of 300-400 cP. Based on preliminary studies we have done, the availability of hydroxy ethyl cellulose in Indonesia is limited and the price is quite expensive. Hydroxy ethyl cellulose itself is a basic ingredient gel maker based hydrogels. Generally gel based hydrogels chosen because it is quite ideal for a variety of medical examination. Hydroxy ethyl cellulose is a group of modified cellulose is used as the base material of various products in the form of gel. In this group there are several basic types of materials that have identical physicochemical properties, namely ethyl selullosa, selullosa hydroxyethyl, hydroxypropyl cellulose, hydroxypropyl methyl cellulose, methyl cellulose and methyl ethyl cellulose. Hydroxy propyl cellulose more easily found in Indonesia and commonly used as an ingredient in the gel and sterile eye drops. Hydroxy propyl cellulose has physicochemical properties similar to hydroxymethyl cellulose.[10]

Based on this, it appears the idea to develop a sterile gel formulation based on hydroxy propyl cellulose instead of a sterile gel-based hydroxyethyl cellulose for HyFoSy examination that meets the requirements of Pharmaceutical. The pharmaceutical requirements include organoleptic parameters ie physical appearance gel (consistency, texture, color and smell) the suitability of pH, viscosity stability and sterility. Previous studies have been conducted to formulate preliminary composition hydroxy propyl cellulose-based gel that resembles the innovator product. Obtained three formulation composition. The first formula consisting of hydroxy propyl cellulose 3.5%, 5% glycerol, and aqua bidestilata, the second formula consisting of hydroxy propyl cellulose 4%, 3% glycerol and aqua bidestilata and third formulas consisting of hydroxy propyl cellulose 4%, 5% glycerol and aquabidestilata. All three of these formulations have organoleptic appearance, pH and initial viscosity that is almost like a product innovator. Therefore, further study is needed to determine which formulation has the best potential to be used as alternative media on HyFoSy. This development is very interesting because it brings many benefits, for example by formulating the gel itself is expected to be produced products at lower prices and with the local availability facilitate the clinician to perform HyFoSy examination, without having to wait for the import.

METHODS

The research was conducted in November 2012 and 2013, in the Laboratory and in the Laboratory of Pharmaceutical Formulation and sterile preparations Technology, Faculty of

Pharmacy, University of Padjadjaran and ASTER assisted Reproductive Technology Clinic, General Hospital dr. Hasan Sadikin.

Materials

Raw materials used in the preparation of gel is Hydroxy Propyl Cellulose (Lawsim®), glycerin (Merck®) and aquabidestilata sterile (Ikapharmindo®). Materials used for sterility test Trypticase Soy Broth (TSB) (Oxoid®) and Fluid Thioglycollate Medium (FTM) (Merck®).

Formulation design

Sterile gel formulation was conducted by two techniques namely aseptic formulation process and gamma ray irradiation. Three groups of formulations made with a composition based on the results of the preliminary study, the first formulation aseptically followed by filter sterilization of bacteria. Bacteria filter used 0.22 micron size. In the second group formulation aseptically followed sterilization by autoclave at a temperature of 121oC. In the third group all materials sterilized with gamma rays 25 kGy then formulated aseptically. Gel formula that is used in a sterile gel formulation based on hydroxypropyl cellulose sonohisterosalpingografi can be seen in Table 1.

Tabel 1. Formula of Sterile Preparations Gel based on preliminary studies

Ingredient(%)	F1	F2	F3	Innovator (F0)*
Hydroxy propyl cellulose	3,5	4	4	Contained
Glycerine	5	3	5	HEC + Glycerine
Aquabidestilata Steril add until	100	100	100	

*Innovator = based on manufacture data of ExEm gel

Information:

F0 : gel containing *Hidroxy etil cellulose* (HEC) and glyceril with viscosity of viskositas 300-400 cP and pH of 6-7

F1 : gel containing 3.5% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata

F2 : gel containing 4% hydroxy propyl cellulose and 3% glycerin with the addition of up to 100 ml of sterile aquabidestilata

F3 : gel containing 4% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata

Evaluation of physical properties of the formula (consistency, color, texture and smell) and the pH and viscosity of the formula compared to standard innovators every 7 days during the 42 days of observation. Results of observations compared to standard gel innovators.

Testing the physical properties of the gel with the organoleptic

Consistency : if the consistency resembles the consistency of innovators coded KM *, if consistency is thicker than KM * then coded KM ** and if consistency is thicker than KM ** then coded KM ***.

Color : If colored translucent gel then coded B, in case of turbidity then coded K, if the color of the gel becomes highly turbid then coded SK.

Texture : f testur slippery smooth soft coded LHL, in case of granulation or clumping coded G.

Odor : If it does not smell coded TB, if there is a change odor coded B

Gel sterilization

There are three types of sterilization are:

1. Sterilization by gamma ray irradiation on materials used then performed aseptically gel formulation.
2. Sterilization by using moist heat (autoclave).
3. Sterilization by filtration using a 0.22 micron filter bacteria.

Sterility testing performed on all formulas are sterilized with these three methods, followed by the growth of bacteria and fungi test using the test medium in the first 14 days of observation.

Sterility testing in Preparation of Sterile Gel

Sterility test includes an evaluation of the test medium (which include fertility testing and test of Fluid Thioglycollate medium effectiveness against bacteria Bacillus subtilis and media Tripticase Soy Broth against the fungus Candida albicans), examination of the number of microorganisms in the cabin Laminar Air Flow (LAF) using media Nutrient agar, and test sterility of sterile gel preparations.

Evaluation of Physical Properties of Gel Sterile Preparations

Evaluasi sifat fisik yang dilakukan terdiri dari pengamatan organoleptis, pengukuran pH dan pengukuran viskositas dari sediaan gel steril.

Data Analysis

To compare the differences between the mean organoleptic appearance, pH and viscosity levels used ANOVA, the difference was considered statistically significant if $p < 0.05$. Correlation test is used to find the correlation between changes in pH and viscosity of the observation time.

RESULTS

Results of Formulation Design

Determined from the results of the three variations of the design formula gel where F1 which is a gel formula containing HPC 3.5%, 5% glycerol and the addition of up to 100 ml of sterile aquabidestilata. F2 is a gel formula containing HPC 4%, 3% glycerol and the addition of up to 100 ml of sterile aquabidestilata. F3 is a gel formula containing HPC 4%, 5% and the addition of glycerin aquabidestilata up to 100 ml.

HPC after expands perfectly in sterile aquabidestilata colored translucent, soft and odorless. Besides as a surfactant (thickener) addition of glycerin is also intended as an anti-foaming to eliminate bubbles when developing HPC by using a digital stirrer.

Gel formulations according Sterilization Techniques

The results of the gel formulation made from cellulose hydroxy propyl made aseptically can be seen in the following table:

Tabel 2. Results of Gel Formulations prepared aseptically

Formula	Consistency	Color	Texture	Odor
F1	Km**	B	LHL	TB
F2	Km*	B	LHL	TB
F3	Km***	B	LHL	TB

Notes:
F1 : gel containing 3.5% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F2 : gel containing 4% hydroxy propyl cellulose and 3% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F3 : gel containing 4% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata
Km* : Viscous flow given one point (marked *)
Km** : Viscous flow given two points, with a thicker viscosity of one point (marked **)
Km*** : Viscous flow given three points, with a thicker viscosity than two points (marked ***)
B : Clear
LHL : Slick, Smooth and Soft
TB : Odorless

From Table 2 it can be seen that all three formulations sterile gel formulated aseptically has a different consistency, the F1 gel containing 3.5% HPC, 5% glycerin and added to 100 ml aquabidestilata, the gel has a thicker consistency than F2 HPC containing 4%, 3% glycerin and added to 100 ml of sterile aquabidestilata. While F3 containing 4% and 5% glycerin HPC and aquabidestilata added to 100 ml, the gel is more viscous than F2.

Color of the three formulas give the same color is clear. The clear color comes from the color HPC when inflate with sterile aquabidestilata not cause the color or clear. Similarly, the addition of colored translucent glycerin does not change the color of the gel.

The texture of the three gel formulations provide the same texture when applied to the back of the hand, which is smooth and soft. This texture is formed due to the physical properties of the HPC that make up the solution becomes soft, clear and colloidal.[11]

The smell of the three formulations gel is odorless. This was due to the absence of a substance that smelled on the formula used. Sterile gel formulations, which performed aseptically further attempted to do this by using the method of sterilization filtration using a 0.22 micron filter size. Its results can be seen in Table 3.

Tabel 3 Gel Formulations results are Sterilized by Filtration Method Using Filters

Formula	Consistency	Filtrate
F1	Km**	Tf
F2	Km*	Tf
F3	Km***	Tf

Notes:
F1 : gel containing 3.5% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F2 : gel containing 4% hydroxy propyl cellulose and 3% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F3 : gel containing 4% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata
Km* : Viscous flow given one point (marked *)
Km** : Viscous flow given two points, with a thicker viscosity of one point (marked **)
Km*** : Viscous flow given three points, with a thicker viscosity than two points (marked ***)
Tf : Unfiltrated

The conclusion from all three gel formula (F1, F2, F3) failed sterilized by filtration method using a 0.22 micron sized filter bacteria. Viscosity is too large so it is difficult to pass through the filter with a very small size.

Results of hydroxypropyl cellulose-based gel formulation sterilized by moist heat method (autoclaves) can be seen in table 4.

Tabel 4. Gel Formulations results sterile Sterilized by Wet Heat Method (autoclaves)

Formula	Cosistency	Color	Texture	Odor
F1	**Km****	**B**	**G**	**TB**
F2	**Km***	**B**	**G**	**TB**
F3	**Km*****	**B**	**G**	**TB**

Notes:
F1 : gel containing 3.5% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F2 : gel containing 4% hydroxy propyl cellulose and 3% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F3 : gel containing 4% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata; B: Clear; G: Clot; TB: Odorless.

To three gel formula (F1, F2, F3) were sterilized by moist heat method (autoclave) failed sterilized. This was due to the results of sterilization experience physical changes. Physical changes are visible on the gel formula that has undergone a sterilization process with moist heat methods (autoclaves) can be seen from the condition that no longer viscous (diluted). In addition, in the preparation formed white blob that settles HPC gelatinous mass shaped chewy. This is probably caused by the physical properties of the sediment and the formation of HPC large clumps at temperatures between 40 -50^{o}C.[11]

The results of gel formulations with materials previously sterilized with gamma rays then performed aseptically processing shown in Table 5.

Tabel 5 Sterile Gel Formulation results using materials that presterilized with Gamma Rays Then Formulated Aseptically

Formula	Consistency	Color	Texture	Odor
F1	Km**	B	LHL	TB
F2	Km*	B	LHL	TB
F3	Km***	B	LHL	TB

Notes:
F1 : gel containing 3.5% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F2 : gel containing 4% hydroxy propyl cellulose and 3% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F3 : gel containing 4% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata
Km : Viscous flow
B : Clear
LHL : Slippery, Smooth and Soft
TB : Odorless

The results showed that the gel formula with a base HPC can only be sterilized using all three methods, materials for gel formulation sterilized with gamma rays and then gel formulated aseptically. Bacteria filters and wet heat sterilization methods can not be used to sterilize these gel formulations.

Sterility Test Results of Sterile Gel Formulation

Sterility test is only performed in a sterile gel formulation with gamma ray sterilization method, because as has been noted previously that the gel formulations are sterilized by filtration method using bacteria filters and gels were sterilized by moist heat method (autoclave) the result is considered failed. Evaluation results of sterility in three sterile gel formulated aseptically performed for 14 days can be seen in Table 6

Table 6 Sterility Test Results of Sterile Gel Formulation

Observation day	Formula					
	F1		F2		F3	
	FTM	TSB	FTM	TSB	FTM	TSB
1	(-)	(-)	(-)	(-)	(-)	(-)
2	(-)	(-)	(-)	(-)	(-)	(-)
3	(-)	(-)	(-)	(-)	(-)	(-)
4	(-)	(-)	(-)	(-)	(-)	(-)
5	(-)	(-)	(-)	(-)	(-)	(-)
6	(-)	(-)	(-)	(-)	(-)	(-)
7	(-)	(-)	(-)	(-)	(-)	(-)
8	(-)	(-)	(-)	(-)	(-)	(-)
9	(-)	(-)	(-)	(-)	(-)	(-)
10	(-)	(-)	(-)	(-)	(-)	(-)
11	(-)	(-)	(-)	(-)	(-)	(-)
12	(-)	(-)	(-)	(-)	(-)	(-)
13	(-)	(-)	(-)	(-)	(-)	(-)
14	(-)	(-)	(-)	(-)	(-)	(-)

Notes:
F1 : gel containing 3.5% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F2 : gel containing 4% hydroxy propyl cellulose and 3% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F3 : gel containing 4% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata
(+) : Occur growth of bacteria / fungi
(-) : No growth of bacteria / fungi

From Table 6 it can be seen that the overall sterile gel formulation based hydroxy propyl cellulose does not occur growth of bacteria and fungi. This means that the whole formula meets the requirements of sterility, which is free of microbes. So sterile gel formulation made with pre-

sterilized materials using gamma rays followed by aseptic formulation can be used to create a sterile gel formula in accordance with the requirements of sterility or in other words the sterility requirements can be met.

Evaluation of Physical Properties, pH and viscosity stability Sterile Gel Formulation

Evaluation of sterile gel formulation continued to observations of physical properties include organoleptic observation, evaluation of the stability of pH and viscosity evaluation of the gel.

Observations organoleptic

Based on observations during 42 days of storage, look to three sterile gel formula that made no change in organoleptic. Preparations still translucent color, consistency viscous flow, smooth texture, smooth and soft portion and odorless. Based on observations concluded no organoleptic differences between the gel formulations.

Measurement of pH Stability Evaluation Results

PH measurement results into three formulas sterile gel formulated aseptically can be seen in Table 7.

Table 7 Sterile Gel pH Measurement Results During the 42 day storage

Formula	pH Gel at Day-						
	1	7	14	21	28	35	42
F1	6,52	6,50	6,48	6,45	6,47	6,47	6,44
F2	6,56	6,51	6,47	6,42	6,42	6,45	6,43
F3	6,68	6,65	6,61	6,61	6,64	6,69	6,67

Notes:
F1 : gel containing 3.5% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F2 : gel containing 4% hydroxy propyl cellulose and 3% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F3 : gel containing 4% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata

From Table 7 it can be seen that the results of measurements performed in the pH interval of 7 days to the day-42 ranged from 6.42 to 6.69. These results are still in the pH range that can be used as a sterile gel is between 6-7 according to the innovator formulation pH. Of the two samples innovators we tested, we get an innovator formulation is pH 7.40 and 7.03. Thus the pH generated by gel formula with hydroxy propyl cellulose base is lower than the pH of the gel innovator but still within the range of pH gel manufacturing. In addition, the pH value obtained is included in the pH range for the preparation of sterile gel-based hydroxy propyl cellulose normal, ranging from 5.00 to 8.50.[12]

Table 7 shows that there is a change of pH on the gel sterile, either the increase or decrease during the 42 days of storage. PH changes that occur range from 0.01 to 0.05 per week and start week 4, the pH of the gel began to stabilize. From Table 7 also note that F3 tends to be more alkaline than F1 and F2, it is probably related to differences in the concentration of each formulation, in addition to the effect of viscosity changes also affect the pH changes, based on the literature it is known that a decrease in viscosity can cause a chain of hydrogen hydrolyzed so pH of the preparation to decrease.[12]

In the ANOVA statistical analysis showed that the difference in pH between the fomula significantly different ($p < 0.05$), as well as the difference between the average pH of observation also significantly different ($p < 0.05$) except between day 28 and 42 are not meaningful ($p = 0.068$). correlation between pH with long observation and correlation between pH and formulas lead to the conclusion that the longer the time, the lower the pH; the higher the pH, the higher formulas.

Viscosity Stability Measurement Evaluation Results

The results of viscosity measurements of three formulas are formulated sterile gel can be seen in Table 8.

Table 8. Sterile Gel Viscosity Measurement Results During the 42 days of storage

Formula	Viscosity gel after storage day-						
	1	7	14	21	28	35	42
F1	489,40	482,74	472,21	468,39	470,87	467,56	453,13
F2	621,23	618,20	599,87	585,32	591,57	581,48	575,60
F3	630,70	628,43	603,56	615,72	600,81	589,04	577,31

Notes:
F1 : gel containing 3.5% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F2 : gel containing 4% hydroxy propyl cellulose and 3% glycerin with the addition of up to 100 ml of sterile aquabidestilata
F3 : gel containing 4% hydroxy propyl cellulose and 5% glycerin with the addition of up to 100 ml of sterile aquabidestilata

From Table 8 it can be seen that the results of viscosity measurements performed on every 7 days until day 42 ranged from 453.13 to 630.70. This result is within the viscosity that can be used as a sterile gel-based HPC is 300 to 600. However, when compared with the innovator who has a range between 300-400 cP viscosity of the whole gel formulation has a higher viscosity.

In the ANOVA statistical analysis showed that the difference in viscosity between the fomula significantly different ($p < 0.05$), as well as the difference between the average viscosity observation also significantly different ($p < 0.05$)

From the correlation between the average viscosity with a mean observation time and the viscosity of the formula it was concluded that the longer the time the lower the viscosity; the higher the higher viscosity formula.

Correlation between the pH of the viscosity (the control formula) is obtained: $r = -0.515$; $p < 0.001$. This means that the higher the pH the lower the viscosity

DISCUSSION

Tubal evaluation is an important component in the treatment of infertility.[1, 2] One technique that has been developed evaluation is Sonohysterosalpingography, which is a technique of examination of the reproductive tract, with the help of ultrasound.[6, 13] This method requires distension media in implementation.[14] Previously, saline solution is used as a contrast

medium and distension. But saline solution has its drawbacks because not last long in the reproductive tract so that the observation time is very limited. In addition to saline solution, the gel can also be used as a contrast medium. Gel is the best distension medium because it is durable and does not quickly disappear in the tube. Sonohysterosalpingography method using gel as a contrast medium is a new technique that is still under development. Sterile gel product for evaluation of tubal in this world there is only one with the trade name Ex-Em gel. This product uses a hydroxy ethyl cellulose as basic materials.[8, 9]

This study has the background to develop an alternative product that can be used for gel-based sonohysterosalphingography examination, but using gel own production cheaper and more secure availability. One alternative gel base that it can be used is hydroxy propyl cellulose. Hydroxy ethyl cellulose and hydroxy propyl cellulose has the physical and chemical properties are similar, so that hydroxy propyl cellulose is expected to be developed for sonohysterosalphingography examination.

In this study, done with sterile gel formulation hydroxy propyl cellulose base material with various concentrations to obtain sterile gel formulations in accordance with the requirements of pharmaceutical and approached the innovator product formulations that have been used to HyFoSy. Which became the focus of the development of this research is to develop a sterile gel formula hydroxy propyl cellulose-based alternatives are eligible sterility, stability physical appearance, pH stability and viscosity stability. Stages of the study include the preparation of tools and materials, design formula, gel formulation, sterilization preparation, sterility testing and evaluation of physical properties include organoleptic observations and observations pH and viscosity stability during the 42 days of observation.

At the beginning of the study is to design a sterile gel formulation. With reference to the manufacturing data ExEm Gel then made basic formulation containing 4% hydroxy propyl cellulose and 3% glycerol, and then designed two other preparations were estimated would produce a gel with the physical properties and chemistry are almost the same, namely 3.4% hydroxy propyl cellulose and 5% glycerol and 4% hydroxy propyl cellulose and 5% glycerol. The design of these three formulations intended to obtain alternatives and comparison of various formulations are expected to design similar to the innovator product. This design results obtained from the product which has good physical properties, such as a clear gel, soft and odorless with a

pH of 6-7 and a viscosity of 400-600 cP. The resulting preparation meets pharmaceutical standards are good for hydroxy propyl cellulose-based gel. However, there are inherent limitations that although the viscosity is still within the range of normal viscosity but the results are still above the innovator product with a viscosity of 300-400 cP. Because the ultimate goal of this formulation is to make a gel that will be used to evaluate tubal then a higher viscosity than the innovator product is expected to be an obstacle because the resulting gel viscosity higher so difficult to run through the oviduct whose size is limited. However, the literature indicates that the product innovators also be diluted first with 20 cc saline solution before use, then the range of 400-600 cP will produce a gel that is still able to pass through the oviduct.

To determine the sterilization techniques that can be used for sterilizing gel then conducted experiments with three methods of sterilization, the method of filtration with a filter size of 0.02 microns, wet heat sterilization by autoclave method and sterilization of materials to be used with gamma rays then formulated aseptically. The first two methods fail because the gel can not be filtered by using a bacterial filtrate possibility because the gel molecules is greater than the diameter of the filter, while the wet heat sterilization, the result gel clot after sterilization by autoclave, this can be explained as hydroxy propyl cellulose experience coagulation temperature of 40-45oC. So that the most suitable sterilization technique to sterilize this gel is the third method is by gamma ray irradiation on materials to be used and carried out in aseptic formulation. To prove sterility then observed for 14 days by using the media TSB and FTM to detect the growth of bacteria and fungi / mold. The results of sterility tests showed that the formulations are sterilized in this way remain sterile in the 14-day observation period that qualifies sterile pharmaceutical gel formulation.

Furthermore, sterile gel formulation was tested organoleptic for 42 days, from the observation that the gel remains stable, clear with a soft consistency, colorless and odorless during the observation period therefore concluded that this gel formulation pharmaceutical qualified in organoleptic.

Besides organoleptic observations, carried out also monitoring the pH and viscosity stability on the third formulation was observed. The results showed the difference in pH between the three formulations were statistically significantly different ($p < 0.05$), all three formulations undergo variation of pH change, and there is a correlation between long observation with a decrease in pH. Obtained pH range is between between 6.42 to 6.69. These results are still in the

pH range that can be used as a sterile gel, which is 6-7. At week 4, the pH of the gel is beginning to stabilize. Observations pH stability is very important because if a preparation has a pH that is not stable, then deemed ineligible pharmaceutical good. From the statistical test shows that the formulation affects the pH of the gel, but because the entire formula is still in the expected pH range, it can be concluded that all three formulations have pH values as expected. The weakness of this study is the possibility of confounding variables that influence changes in humidity in the period of observation long enough for the pH of the formulation. This can occur because of lack of impermeable packaging may cause the gel to absorb moisture from the outside, thereby increasing the volume of water in the gel.

On the issue of the stability of the viscosity, the three formulations have a higher viscosity than expected. Statistically, there is a correlation between the decrease in viscosity with longer observation time in the period, but the value remains above 400 cP. Benchmark 400 cP is derived from product innovators, ideally resulting gel viscosity approaching the viscosity of the gel innovators. From the statistical test showed that the pH effect on the viscosity, the higher the pH, the lower the viscosity. This is probably related to the existing HPC concentration in the formulation.

CONCLUSION

The conclusion is the observation organoleptic and sterilization no difference to the entire formula. pH formulas vary according to the composition of the gel and storage time affects the pH changes, but the levels are still within the appropriate range. There are differences in viscosity were also influenced by the formulation and a decrease in viscosity in accordance with the length of time of observation. Therefore, the best formula is a formula that is closer to the nature of the innovator product, then the formula I is a more appropriate formula that further research is needed to analyze the feasibility of the use of formula one. This gel medium sterilization techniques can only be sterilized by gamma ray irradiation technique in the materials before use and continued with aseptic formulation.

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