



Antifertility Test of Maja Fruit Infusa (*Aegle marmelos* Corr) on Female White Rats (*Rattus norvegicus*)

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Abstract. Contraceptives developed from natural materials are still very minimal, while the use of synthetic contraceptives in the community causes many complaints. Phytosterols in Maja fruit are classified as steroids having a cyclopentane perhydro phenanthrene ring shape, are estrogenic, and can affect follicular development and have anti-gonadotropic and anti-implantation effects, causing inhibition of ovulation and are anti-fertility. This study aims to determine the infusion of maja fruit has the potential as an anti-infertility against female white rats. This research method by grouping into 5 groups of female rats that have passed the fertility test then consisting of 2 control groups and 3 treatment groups... The antifertility effect on female white rats using Maja fruit infuse with a concentration of 2%, 5% and 10% given orally for 7 consecutive days. In all three treatments, the rats were infertile.

Keywords: Infusion, Infertile, Maja, Contraception

INTRODUCTION

For sexually active people, reproductive services, such as health provision of contraception and treatment for sexually transmitted infections, are either unavailable or provided in a way that makes people feel unwelcome and the community embarrassed. As a result, it tends to rely on resources outside the formal health service delivery system, such as home remedies, traditional contraceptive methods, clandestine abortions or medicines from shops or traditional health practitioners. To address this issue, a number of women's initiatives in Indonesia. As evidence, more than half (57.4%) of all women of childbearing age use family planning. The most popular is the injectable method, but the IUD (Intra Uterial Device) method is also gaining traction. Interestingly, the province with the highest percentage of women using modern family planning in Indonesia is Yogyakarta. In fact, the province, which is one of the centers of Javanese culture, is famous for the saying many children many fortunes. (LDFKUI, 2013). have been developed and implemented that have made it easier for the general public to get the quality health services they need. (Budhiarta and Muttaqin, 2019).

Family planning (KB) seems to have become part of life The use of traditional family planning in women of childbearing age includes diligently drinking turmeric and betel juice. That is one way of natural birth control. Although there are many modern family planning tools and methods, traditional family planning is still the choice of 3.8% of women of childbearing age in Indonesia. According to the survey, this method is widely used by women aged 35-39 years. But you need to be careful too, if the dose is not right, traditional birth control can actually be dangerous. (LDFKUI, 2013). Family planning methods can be divided into modern and traditional method.

Modern family planning methods are sterilization, pills, IUDs, injections, fertilizers, condoms, intravaginal / diaphragm, emergency contraception and Lactational Amenorrhea Method (MAL). Traditional methods include periodic abstinence and interrupted intercourse. (Ministry of Health, 2013). There are still few contraceptives developed from natural materials, especially Indonesian natural materials. In Indonesia there are 18 types of medicinal plants that have the potential as anti-fertility men. Some of these plants include papaya seeds (*Carica papaya*), Gandarusa (*Justicia Gandarusa*) and bitter melon (*Momordica charantia*) (Ministry of Health, 2006).

Some studies that have been conducted on medicinal plants include the secang plant (*Caesalpiniasappan* L), The results of research by Astuti, et al in 1995 showed that secang wood *in vivo* can reduce the motility of mice spermatozoa. On the other hand, the use of synthetic contraceptives in the community causes many complaints, while in nature there are many contraceptives available from plants that are safe, cheap and without side effects, including maja plants. The maja plant (*Aegle marmelos* Corr), which grows in many regions in Indonesia, is very effective as a natural contraceptive that is easily available, and safe for women to use. The plant contains maja β - the sitosterol, stigmasterol. These phytosterols are classified as steroids having a cyclo pentane-perhydro-phenanthrene ring shape, estrogenic in nature.

These phytosterols can affect follicular development and have anti-gonadotropic and anti-implantation effects. These substances, if produced in excess, can cause inhibition of ovulation and are anti- fertility. The activity of anti-fertility agents can be contraceptive, interceptive, and abortifacient. Anti-fertility agents may act through one or more mechanisms and sites. In female mammals, anti- fertility agents can affect the activity of hypothalamic, pituitary, ovarian, uterine, and vaginal functions by disrupting pre-ovulation, pre-implantation, and post-implantation mechanisms. According to the tradition of the people of Pinrang (Makassar), Aceh, Palembang and Java, the maja plant is believed to be effective in regulating menstruation in women and treating sexual diseases, analgesic, antiviral, anti-fungal, anti-cancer, anti-diarrhea. In addition, maja leaf extract is very potential to be used as anti- testicular and anti-spermatogenic in male rats. The purpose of this study is to determine the infusion of Maja fruit (*aeglemarmeloscorr*) has the potential as anti-infertility against female white rats (*rattusnorvegicus*).

METHODS

The research instruments used in this study were test animal cages, food and drink containers, gloves, measuring cups, drop pipettes, stirring rods, beakers, test tubes, blenders, aluminum foil, analytical scales, filter paper, glass funnels, scissors, oral sonde, sudip, water bath, and 5 ml disposable syringe. The materials used in this study were Maja fruit infusa concentrations of 2%, 5% and 10%, distilled water, mycroginin, female and male white rats.

The research procedure consists of 3 processes including:

a. *Simplisia* Preparation

Part of the *simplisia* used is fruit. Maja fruit that will be used in this study has gone through a wet sorting process and washing to separate impurities using clean running water, then a 1 kg sample of Maja fruit is made infusa using distilled water at 60° C for 15 minutes.

b. Antifertility Test

Pre-test Control The test animal fertility test is a preliminary test or pre-test before treatment or administration of this preparation aims to ensure that the rats used are fertile or infertile rats. Fertility tests are carried out by mating female rats with male rats naturally by uniting female rats and male rats in one cage with a ratio of 1 male: 3 females into 5 treatment groups. After 1 week

of combining male and female rats, then the female rats were observed for their physical characteristics whether they were pregnant or not by looking at the growth of mammae in females, if there were mammae then the female rats were indicated to be pregnant. Of the 15 female rats that have been observed mammary growth, all rats have indicated pregnant growth because the mammae in each group are enlarged. have grown Monitoring and characteristics of pregnancy based on mammary growth refers to Prawiroharjo (2008). According to Prawiroharjo, signs and symptoms of pregnancy can be divided into several parts, one of which is the mammae becoming tense and enlarged. This situation is due to the influence of estrogen and progesterone hormones that stimulate the ducts and alveoli of the breast. So it can be concluded that the female rats used are fertile rats or not sterile.

c. Administration of Preparations

Female rats that have passed the fertility test are then grouped into 5 groups consisting of 2 control groups and 3 treatment groups. Each group consists of 3 female rats. Group 1 consists of a negative control group (K (-)) given aquadest, group 2 consists of a positive control group (K (+)) given microgynon suspension, group 3 consists of treatment group 1 (KP1) given maja fruit infusa with a concentration of 2%, group 4 consists of treatment group 2 (KP2) given maja fruit infusa with a concentration of 5%, and group 5 consists of treatment group 3 (KP3) given maja fruit infusa with a concentration of 10%. Oral treatment of test animals was carried out by giving a predetermined dose with a volume of 4 ml per test animal. Giving test samples was done in the morning (08.00-09.00), female white rats were given infusa extract of maja fruit each with a concentration of 2%, 5%, and 10% for 7 consecutive days, after giving infusa the last day female rats were combined with male rats so that mating occurred until day 8. After that the female rats were separated from the male rats. Then the female rats were kept until the 21st day after mating.

RESULTS AND DISCUSSION

After conducting research on the activity of Maja fruit infusa (*Aegle marmelos*) as a contraceptive in white rats (*Rattus norvegicus*), the results were seen in table 1 below:

Table 1 Fertility rate of white rats (*Rattus norvegicus*) after maja fruit infusion compared with negative control and positive control.

In this study, the test animals used were female white rats that were given infusa of maja fruit for 7 consecutive days, after the third day of extract administration, female rats were combined with male rats so that mating occurred until day 7.

After treatment, female rats were separated from male rats and maintained for 21 days. The purpose of being treated for 7 consecutive days and then combined between female rats and male rats is so that female rats can reach their estrus cycle so they can copulate (Namal apriyanti, 2011). Estrus/birahi is a condition where female rats are willing to accept male rats. The estrus cycle is a period from the start of estrus to the next estrus. The length of the estrus cycle in female rats ranges between 4-6 days, so this is the reason male rats are combined on day 4 after giving the extract, so that female rats can reach the estrus cycle so that they can copulate (Namal apriyanti, 2011).

The observed result was infertility of female white rats. This study used 15 female white rats and 5 male white rats in a ratio of 3 : 1 which was divided into 5 groups, each group consisted of 3 female white rats and 1 male white rat. Maja fruit infusa was varied with 3 concentrations (doses) of 2%, 5%, and 10% which were compared with Na- CMC as negative control and Microgynon suspension as positive control. The contraceptive activity of maja fruit infusa on female white rats is known by looking at the percentage of infertility of test animals from each concentration. The percentage of infertility of female white rats after giving negative control is 0% and after giving

Treatment (Dose) Number of Fertile Animals
Table 1.

Treatment (Dosage)	Number of Animals	Fertil	Infertile	Infertile Percentage (%)
Aquadest	3	3	0	0%
Microginon Suspension	3	0	3	100%
2% Maja Fruit Infusa Solution	3	0	3	100%
Maja Fruit Infusa Solution 5%	3	0	3	100%
10% Maja Fruit Infusa Solution	3	0	3	100%

Aquadest 3 3 Infertile 0 Percentage Infertile (%)
Microginon Suspension 0% 3 0 Maja Fruit Infusa
Solution 2% 3 3 100% 0 Maja Fruit Infusa
Solution 5% 3 3 100% 0 3 Maja Fruit Infusa

Solution 10% 0 3 3 100% 100% microginon suspension (13.7 μ g/kgBB) is 100%. At the time of administration of maja fruit infusion with variations at a concentration of 2% is 100%, at a concentration of 5% is 100% and at a concentration of 10% is 100%. The percentage of unbunting of the positive control microginon suspension (13.7 μ g / kgBB) is 100% which is also the same result, which is shown at 2%, 5% and 10% concentrations of maja fruit infusa. This indicates that at concentrations of 2%, 5% and 10% have antifertility activity characterized by no fertility in female white rats given maja fruit infusa.

Disruption of the production process may occur at various stages of the reproductive process such as disruption of ovum development which ends with ovulation, disruption of ovum transportation and fertility of female white rats and nidation. Inflation itself can be viewed from two angles, namely the state of the ovum or zygote and the readiness of the uterus in receiving the zygote, the main disturbance in the reproductive process can change the disturbance of hormone levels that control reproduction. This is the basis of the mechanism of action of antifertility containing estrogen and progesterone (Hafez. 1980 in Namal Apriyanti. 2011). The effect of antifertility in female mammals can provide action through the balance of the hormone system a number of workplaces. The workplace in female mammals is the hypothalamus, ovaries, fallopian tubes, uterus, uterine mouth and vagina. Substances may exert antifertility effects at more than one site of action and not necessarily by the same mechanism. Conversely, various antifertility substances have the same site of action to inhibit fertility but with different mechanisms (Partodiharjo, 1987).

White rats (*Rattus norvegicus*) are classified as spontaneous ovulators, in this group the phylculotropin hormones namely (FSH and LH) stimulate follicular development and estrogen secretion gives feedback to the hypothalamus in the form of messages. Conversely horny ovulator, for example in rabbits, estrogen conveys its own hypothalamus message in addition to that the follicle has matured and ready to ovulate. Centrally can cause lust in experimental animals and stimulate the hypophysis to be sensitive to stimuli caused by the peak wave of LH which then causes ovulation and stimulates the secretion of progesterone (Partodiharjo, 1987).

The results also showed that the potency of the test sample was relatively the same as the contraceptive pill preparation used in the positive control group. Sitasiwi and Mardiaty (2016) stated

that contraceptive pills contain synthetic estrogen and progesterone. The synthetic estrogens in the test material are ethynil estradiol and levonorgestrel Ethynil estradiol has the same structure as estrogen. Antifertility compounds in principle work in 2 ways, namely through cytotoxic or cytostatic effects and through hormonal effects that inhibit the metabolic rate of sex cells by interfering (Herdiningrat, 2002).

The mechanism of action of bioactive compounds contained in maja fruit is thought to work as antifertility through these 2 ways. Meanwhile, flavonoid compounds are estrogenic active ingredients, flavonoid compounds are also known to stimulate the formation of estrogen in mammals, and from their structure there is a similarity with estrogenic hormones (Nurliani, 2007). According to Robinson (1995) in Kristan (2010), this active ingredient is estrogenic or estrogen-like. Isoflavones which are a class of flavonoids are substances similar to estrogen, but different from OH bonds. In the body isoflavones are similar to estrogen. It has been shown that isoflavones inhibit tyrosine kinase which can inhibit cell growth and development.

According to Setyowati (2015), flavonoid compounds, especially isoflavone derivatives, namely genistein, are estrogenic or estrogen-like active ingredients. Substances whose structure is analogous to the hormone will associate themselves with hormone receptors, but do not stimulate the receptor. If genistein occupies estrogen hormone receptors (ER α or ER β) as a result, the action of hormones on target cells will be reduced. This can occur due to the similarity of space between genistein and estrogen hormone.

CONCLUSIONS AND SUGGESTIONS

From this study it can be concluded that maja fruit infusa has the potential as an anti-infertility against female white rats.

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