

Effect of Continuous Mating on Maternal and Mating Behaviour, Conception Rate, Gestational Length, Litter Size and Kit's Growth Alive in New Zealand White Rabbits

By

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A thesis submitted in fulfilment of the requirement for the degree of Bachelor of Applied Science (Husbandry Science) with Honours

Faculty of Agro-Based Industry

UNIVERSITI MALAYSIA KELANTAN

2022

DECLARATION

I declare that this thesis entitled "Effect of Continuous Mating on Maternal and Mating Behaviour, Conception Rate, Gestational Length, Litter Size and Kit's Growth Alive in New Zealand White Rabbits" is the results of my own research except as cited in the references.

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ACKNOWLEDGEMENT

First of all, I am grateful to The Almighty God for ease my journey in finishing my Final Year Project successfully.

Besides, I sincerely express my deep sense of gratitude to my supervisor, Dr. Raja Ili Airina Binti Raja Khalif for her extraordinary cooperation, invaluable knowledge, guidance and supervision. This thesis is the result of her continuous support and patience throughout Final Year Project thesis writing.

In addition, I am hugely appreciative to Nur Imanina Binti Termizi for allowing me to use Re:Live Sdn. Bhd. rabbit house University Malaysia Kelantan Jeli Campus during my research. Without helps from Re:Live Sdn. Bhd. staff Mohammad Rohaimi Bin Abdul Aziz, I cannot complete my research work. In addition, I am also grateful to Dr. Raja Ili Airina Binti Raja Khalif for allowing me to use her ultrasound machine during ultrasonography test on the pregnant does.

Last but not least, it is impossible to finish this task without any supports and help from my family, FYP mate, Nur Shahida Nafisah Binti Buhaira and my dearest soulmate Nur Syakirah Syahrain Binti Mustakim for supporting and encouraging me physically and spiritually. I humbly extend my thanks to these people for helping me to complete this project. Effect of Continuous Mating on Maternal and Mating Behaviour, Conception Rate, Gestational Length, Litter Size and Kit's Growth Alive in New Zealand White Rabbits

ABSTRACT

Rabbit industry have a lot of potential due to high demand in rabbit meat and serve as an alternative to beef and mutton. In 2017, the global rabbit meat market sales were \$6.4 billion, up 12% from the previous year, and is likely to continue growing in the near future. However, standard operating procedures (SOP) for breeding in rabbits to optimize productivity are still scarce. Mating continuously for three days was a common practice and believed to boost the doe's pregnancy rate among farmers. However, the frequency of ejaculations and repetitive service from buck may result in sexual weariness and affect conception rate. Therefore, the aims of the study were 1) to investigate the effect of different mating frequencies on mating behaviour, maternal behaviour and conception rate and 2) to determine its effect on gestational length, litter size and kit's growth alive in New Zealand White (NZW) rabbits. A total of 8 does and 4 bucks of NZW were divided into 2 groups. Does were mated for 1 day (Group 1) and 3 days continuously (Group 2), respectively. Growth and maternal behaviours of does were analysed in 7 weeks. Abdominal palpation was carried out on Day 11 until Day 15 and while ultrasound on Day 21 respectively. Prior to analysis of kit's growth for 10 days, conception rate, gestational length and litter size were observed. Results showed that no significant differences of difference mating frequencies on the mating behaviour, maternal behaviour and conception rate in NZW does. In addition, different frequencies of mating also showed no significant differences on gestational length, litter size and kit's growth in NZW. Further work on the effect of difference frequency of mating on sperm quality is recommended to optimize the productivity of rabbit meat industry in Malaysia.

Keywords: New Zealand White rabbits, mating behaviour, maternal behaviour, conception rate, gestational length, litter size, sperm quality.

Kesan Mengawan Berterusan terhadap Tingkah Laku Keibuan dan Tingkah laku

Mengawan, Kadar Konsepsi, Tempoh Masa Kehamilan, Jumlah Anak yang

Dilahirkan dan Pertumbuhan Anak Arnab Hidup di Arnab Putih New Zealand

ABSTRAK

Industri arnab mempunyai banyak potensi kerana permintaan yang tinggi terhadap daging arnab dan berfungsi sebagai alternatif kepada daging lembu dan kambing. Pada 2017, jualan pasaran daging arnab global ialah \$6.4 bilion, meningkat 12% daripada tahun sebelumnya, dan berkemungkinan akan terus berkembang dalam masa terdekat. Bagaimanapun, prosedur operasi standard (SOP) untuk pembiakan arnab bagi mengoptimumkan produktiviti masih terhad. Mengawan secara berterusan selama tiga hari adalah amalan biasa dan dipercayai dapat meningkatkan kadar kehamilan betina di kalangan penternak. Walau bagaimanapun, kekerapan ejakulasi dan servis berulang dari pejantan boleh mengakibatkan keletihan seksual dan menjejaskan kadar kehamilan. Oleh itu, matlamat kajian adalah 1) untuk menyiasat kesan frekuensi mengawan yang berbeza ke atas tingkah laku mengawan, tingkah laku keibuan dan kadar kehamilan dan 2) untuk menentukan kesannya terhadap panjang kehamilan, jumlah anak yang lahir dan pertumbuhan anak-anak yang hidup pada arnab New Zealand White. (NZW). Sebanyak 8 betina dan 4 pejantan NZW dibahagikan kepada 2 kumpulan. Betina-betina telah dikawinkan selama 1 hari (Kumpulan 1) dan 3 hari secara berterusan (Kumpulan 2), masing-masing. Pertumbuhan dan tingkah laku keibuan dan jumlah anak yang lahir dianalisis dalam masa 7 minggu. Palpasi abdomen dilakukan pada Hari ke-11 hingga Hari ke-15, manakala ultrasound dilakukan pada Hari ke-21. Sebelum analisis pertumbuhan anak-anak selama 10 hari, kadar kehamilan, panjang kehamilan dan jumlah anak yang dilahirkan diperhatikan. Keputusan menunjukkan bahawa tiada perbezaan ketara untuk perbezaan frekuensi mengawan pada tingkah laku mengawan, tingkah laku keibuan dan kadar kehamilan di NZW. Selain itu, frekuensi mengawan yang berbeza juga tidak menunjukkan perbezaan yang ketara pada panjang kehamilan, jumlah anak yang dilahirkan dan pertumbuhan anak-anak di NZW. Kajian lanjut mengenai kesan perbezaan kekerapan mengawan terhadap kualiti sperma adalah disyorkan untuk mengoptimumkan produktiviti industri daging arnab di Malaysia.

Kata kunci: Arnab New Zealand White, tingkah laku mengawan, tingkah laku keibuan, kadar kehamilan, tempoh masa kehamilan, jumlah anak yang dilahirkan

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LIST OF ABBREVIATIONS

DM	Dry Matter
g	Gram
kg	Kilogram
mm	Millimeter
min	Minutes
NZW	New Zealand White
SEM	Standard Error Mean
SOP	Standard Operating Procedure

FYP FIAT

MALAYSIA

KELANTAN

LIST OF SYMBOLS



CHAPTER 1

INTRODUCTION

1.1 RESEARCH BACKGROUND

Malaysia's livestock industry is one of the most important contributors to the country's agricultural production. In comparison to other ruminant or non-ruminant industries, the rabbit industry is considered underdeveloped. The rabbit industry is still growing in a few areas, such as meat production, wool production, and research, based on the Department of Agriculture and Food.

In the livestock industry, rabbits have a higher reproductive rate than other livestock. They reach sexual maturity after a few months of birth and have short pregnancies. They have huge litters and are unique in that they can be rebred shortly after being kindled (giving birth). A doe can be estimated to have 60 weaned young per year with an intensive breeding program. However, such intense breeding is not suggested for beginners and is rarely employed in commercial production (Michigan State University, 2017).

In female rabbits, the release of eggs is initiated by sexual intercourse rather than a hormonal cycle as in humans (Diane McClure, 2020). Naturally, the buck will breed with

the doe right away. After a few sniffs to alert him to the situation, the buck quickly circles around to the hind end of the doe, mounts the doe, completes the rabbit mating, and then fall off the doe with a grunt. The effectiveness of mating can be increases by allowing the breeding pair to be together for half an hour. Gives the rabbits plenty of time to mate, preferably 2-3 times. Having them mate numerous times can improve with litter size and breeding success. Instead of allowing them to mate three times in a single session, some breeders breed them again after an hour or later the same day to ensure a successful mating. Before removing the doe, a second rabbit mating appears to boost the success rate and litter size. Simply leave the doe in the buck's cage. The buck will regain his breath, lose interest in thumping the floor, and rediscover his interest for the doe. He'll remount her, she'll lift her hind end, and another mating will take place (Karen Patry, 2009).

Mother rabbits' activity prior to parturition was geared toward nest construction, as it is in nature, whereas her postpartum actions were directed toward the pups and humans (intruders). Maternal behaviour in rabbits begins before parturition; during the latter third of the 30-day pregnancy, females begin excavating an underground burrow where the maternal nest will be built. When female rabbits are pregnant, they immediately make a nest out of hay or straw in rabbit industries. Aside from that, soon-to-be mothers utilise their own fur as a blanket to keep the kits warm. The doe may growl defensively or refuse to be handled or stroked on rare occasions (Gonzalez-Mariscal and Rosenblatt, 1996). In this research, we are going to investigate the effect of different mating frequencies on the effectiveness of pregnancy, mating behaviour, gestation length, litter size and maternal behaviour of the doe. Other than that, we also going to investigate whether continuous mating frequencies on the buck affects the performance of the bucks. The aim of this research was to compare the effectiveness of pregnancy, mating behaviour, gestation length, litter size and maternal behaviour of the doe between group A (mating 1 times for 1 days) and B (mating 1 times for 3 days).

1.2 PROBLEM STATEMENT

Currently, rabbit industries have a lot of potential because rabbit meat is in high demand and might be a good alternative to beef and mutton. However, many local breeders are still unaware of the proper approach for breeding rabbits. Farmers are aware of the rabbit industry's opportunity, but many are hesitant to participate because there is no guarantee that the products would find a market. Novices breeders tend to only reared rabbits in a small group. They tend to breed their rabbits instinctively and traditionally. To reduce production costs, the male to female sex ratio is 1: 3. The issue for breeders is that the buck with the best performance will be employed in mating frequently in order to generate the highest quality rabbit progeny. According to many breeders, mating continually for three days will effectively boost the doe's chances of becoming pregnant. However, the frequency of ejaculation and the repetitive service of rabbit buck may result in sexual weariness.

According to Lloyd-Jones and Hays (1918) research, the rabbit conception rate gradually decreased from 72% at the first service to 35% at the 20th service. They just run out of energy after constant mating and are unable to complete the entire mating process. A young buck may be permitted to mate with one doe every 3 to 4 days. After 12 months of age, it may mate 4-6 times in 7 days. Bucks above the age of six should be culled since their sperm quality deteriorates. The frequency of ejaculation affects sperm output, sperm qualities, and fertility. Repetitive ejaculation reduces semen volume, which is linked to

protein, fructose, and citric acid deficiency (Kirton, 1966). In this experiment, we will observe if the difference in mating frequency between the doe and the buck affects mating behaviour, maternal behaviour, conception rate, gestational length, litter size, and kit growth in New Zealand White (NZW) does.

1.3 HYPOTHESIS

This study comprises two hypothesis:

 \mathbf{H}° = There is no significance difference of the different frequencies of mating on mating behaviour, maternal behaviour and conception rate in New Zealand White (NZW) does.

 H^1 = There is a significant difference of the different frequencies of mating on mating behaviour, maternal behaviour and conception rate in New Zealand White (NZW) does.

 \mathbf{H}° = There is no significant difference of the different frequencies of mating on gestational length, litter size and kit's growth.

 \mathbf{H}^{1} = There is a significant difference of the different frequencies of mating on gestational length, litter size and kit's growth.



1.4 OBJECTIVE

- 1. To investigate the effect of different frequencies of mating on mating, maternal behaviour and conception rate in New Zealand White (NZW) does.
- 2. To determine the effect of different frequencies of mating on gestational length, litter size and kit's growth.

1.5 SCOPE OF STUDY

The study will begin with the selection of four bucks and eight does for natural mating. The mating of the does took place at the same time of day, from 8:00 to 9:00 a.m., for each group. The first group consisted of a doe who had only mated with the bucks for one day. Meanwhile, males mated with the second group of doe for three days. The colour of the vulva approach was used to assess the receptiveness of does prior to mating. During mating, the frequency of occurrence of some courtship features such as ear sniffing, genital sniffing, chin rubbing, leg striking, backward falling, or sideways falling was observed. The pregnancy of a doe was then confirmed 11 days after mating by performing manual abdominal palpation. Building a nest is one of the maternal behaviours observed before parturition at about 26 days of pregnancy. Maternal effects include changes in body weight and gestation length. The conception rate, parturition rate, litter size at birth, and weight of kits for 10 days were used to estimate the fertility parameters of does.



1.6 SIGNIFICANT OF STUDY

The significance of this study is to determine the effect of repetitive mating on conception rate and does performance. Short-term frequent ejaculation resulted in a consistent decline in sperm density, sperm volume, and fructose content. Male rabbits are used selectively in breeding practise. Typically, twice a day, no more than two or three times per week. The current findings, however, indicate that significantly more intensive use is associated with good fertility, at least for short periods of time. However, excessive use of males for sperm production is not advised (Adams and Singh, 1981).

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CHAPTER 2

LITERATURE REVIEW

2.1 RABBIT INDUSTRY

Rabbit farming has been classified as a new industry in Malaysia since 1988 by the government for export and domestic use (Raharjo and Bahar, 2016). For Malaysians, rabbit is still seen as a pet rather than a meat source, and it is still frowned upon by culture for a variety of reasons. This may be one of the reasons why the majority of farmers operate on a micro and small scale (Raharjo, 2008). All of this scepticism and tentative adoption prompted the government to launch a strategy and incentive schemes to make business growth more appealing to farmers while simultaneously adapting them to new modern breeding concepts. Rabbit meat began to receive positive reviews and reactions from consumers after a few decades through a variety of items such as satay and marinated meat. In 2018, the number of farmers rose considerably, indicating a rise in market demand (Gunalan *et al.*, 2019). External factors such as ecological, fiscal, and sociological factors affect the sustainability of rabbit meat production in Malaysia. Internal considerations may include genetic improvement, services, diet, fitness, and some other associated problem.

In order to increase rabbit meat yield, some farmers incorporate technologies from the ruminant industry, such as artificial insemination programs and others, into their farming systems. According to Gunalan (2019), the rabbit industry in Malaysia has the potential to become an integral part of the diverse breeding operations, providing a rare opportunity for the breeder. Depending on the method and size of the property, rabbit breeding can be lucrative Rabbits have the potential to become one of Malaysia's main industries, similar to the poultry sector, which is now regarded as a stable sector with a large customer base.

2.2 **REPRODUCTION**

Rabbits can get pregnant at a young age, with some smaller breeds beginning their reproductive life as early as 4 months of age. A female rabbit is fertile for all but around three days per month, but will only produce an egg for insemination when sexually interacted by a male rabbit. According to McClure (2020), once impregnated, a mother rabbit is pregnant for approximately 31 to 33 days and will have a litter of approximately six (or so) young, though a litter of fourteen offspring is possible. The mother rabbit can become pregnant again almost immediately after giving birth (Ali, 1994). In theory, this means that a female might have a litter every month of the year. It is no wonder that rabbits have quite the reputation for the reproduction rate among livestock animals (Michigan State University, 2017).

2.2.1 Spermatogenesis

Seminal plasma, which contains a wide range of substances, is formed by the testicle, the seminiferous ducts, and the glands. The secretion, cell debris, sperm, and immature germ cells from the accessory glands are all present in the ejaculated. Seminal fluid is produced inside the testicle by the tubule recti, rete testis, and epididymis. As indicators, some seminal plasma proteins produced by various tissues are used. The rise or fall in the levels of these markers could indicate a pathological process in a specific tissue. Ricardo (2018) stated that fructose is the most common sugar associated with metabolism and sperm motility, and it is a significant predictor of seminal vesicle performance. Diabetes, oligozoospermia, and azoospermia patients have been found to have abnormally high fructose levels (Trang, 2018). Fructose concentrations are lower in ejaculates containing a high sperm density and motile spermatozoa. It demonstrates that spermatic fructolysis lowers fructose concentration (Mauss *et al.*, 1974).

2.2.2 Ovulation

The rabbit is one of the few mammals that do not ovulate on their own. There is no such thing as an oestrus cycle. Ovulation requires a mating trigger and is hence initiated in nature. Ovulation occurs as a result of sexual stimulation during copulation or in reaction to exogenous gonadotropins (Marques *et al*, June 19, 2018). Females can sometimes stimulate one another to the point of stimulation. This type of ovulation is expected to result in becoming sterile or pseudo-pregnant for a few days. Ovulation is most likely to occur between 9 and 13 hours. However, it often occurs 10 hours after mating. Does are supposed to be in continual heat throughout the year or during breeding season. It is known, that follicles develop and regress in 15–16 day cycles. There is a time when the doe loses interest in the buck. Mechanical stimulation on the vagina can also be used to promote ovulation. (TNAU Agritech Portal, 2009).

2.2.3 Mating Behaviour

A buck develops breeding abilities at the age of eight months. A healthy buck should have the ability to reproduce for at least two to three years. Every three to four days, a young buck may be allowed to mate with one doe. However, once it reaches the age of 12 months, it may mate 4-6 times per week. Bucks over the age of six should be culled because the quality of their sperm decreases with age. Breed, nutritional status, and seasons all have an impact on a doe's reproductivity. Smaller breeds develop sexuality more quickly than larger breeds. A small breed may accept breeding at 3-4 months, whereas a larger breed may accept breeding at 8-9 months. A doe can be used for reproduction until she is three years old. Following that, the doe should be culled.

A doe, whether in heat or not, is difficult to identify from the outside. However, does may exhibit symptoms such as restlessness, anxiousness, and rubbing of the head and chin on the edge of the cage or other things. The vulva swells and turns purple in colour. However, the acceptability of the does to the bucks or the reaction of the does to the bucks should be used as a factor for heat. As a result, heat detection through buck should be performed before to allowing copulation. The average age of the first mating is around 5-6 months. To avoid fighting, doe should always be taken to buck's cage but never vice versa. (Washington State University Cooperative Extension and the U.S. Department of Agriculture, 1991). The best times for mating are early morning and early evening. A receptive doe will lift her tail and allow mating to take place. Male sexual drive varies tremendously. A buck may be slow in providing service to an unfamiliar cage (Nelson, 2015). Mating will occur quite instantly if a buck is virile and a doe is in ideal heat. Following a successful mating, the buck normally gives out a typical cry and falls to one side of the doe (Ali, 1994). Usually, one mating is enough. If a female refuses to allow mating, the keeper should wait 3 to 4 days or aid in mating while holding the female. The doe should be returned to her enclosure after mating (Mitruka *et al*, 1976). Farmers that raise rabbits for a living would prefer to have five or six litters per doe every year. This is only conceivable if the litter is weaned at five weeks of age and the doe is mated shortly after weaning. Each breeding cycle will last between 65 and 75 days. It is also possible to achieve this by mating the doe 21 days after kindling (TNAU Agritech Portal, 2009).

The ideal or suitable time for natural mating of New Zealand White (NZW) rabbit does will be in the morning around 8:00 a.m. to optimise and maximise rabbit productivity and performance. Early morning activity may also result in increased blood glucose levels, which are primarily controlled by body hormones, particularly thyroxin and adrenal cortical hormones, which prepare females for reproductive efficiency (Kishk *et al.*, 2006).

2.2.4 Pregnancy

In rabbits, the gestation (pregnancy) phase lasts 31-32 days or approximately 30 days (Mitruka *et al*, 1976). The nest box should be kept within the cage to make it easier for the doe to provide bedding for the new born. The nest should be available at least 5-6 days before parturition. Nesting materials such as straw, grass, and wood shavings should be included in the nest box. Sawdust should never be used as bedding. A doe may pluck some of her own hairs to construct a nest for her litters (Emery, 2012). During pregnancy, adequate feeding and management strategies should be implemented. During the first 10 to 15 days of pregnancy, the amount of feed should be increased. There should be plenty of fresh water

available. Environmental pressures should be avoided to the greatest extent practicable (Martin, 2014).

2.2.5 Kindling (Parturition)

Kindling refers to the process of giving birth to a new rabbit baby. It is a normal physiological occurrence. Parturition usually occurs late at night or early in the morning (Patry, 2014). It is possible that the keeper will not need to intervene. The process normally takes between 7 and 30 minutes to finish. It is possible that not all litters will be born in order. Some may be born after a few hours or a day. The pregnancy may need to be terminated with an oxytocin shot. Following parturition, the does lick the young and may consume the placenta. The mother rabbit will try to nurse the newborn. If the litter size is eight, all of the babies may be able to suckle because the doe has eight teats. Baby rabbits who are unable to nurse may become weak and susceptible to sickness (Absolon, 1987). Many of them may even die prematurely. During this time, the does should not be disturbed and should be fed *ad lilbitum*. Adequate food and water should be provided so that the baby rabbits have access to the maximum amount of milk. Rabbits used to nurse their young only once, generally at night or early in the morning. A single kindling can produce 6-12 offspring (TNAU Agritech Portal, 2009).

2.2.6 Weaning

Baby rabbits are completely reliant on their mothers immediately after birth. They are born without fur. However, after around 7 days, their hair and vitality start to grow. After 10 days, they open their eyes (Cheeke, 1979). At around 21 days of age, newborn rabbits

can survive without their mothers' milk. The kits should be separated from their mother no later than the fourth week. The doe should be taken out of the cage. Foods such as concentrates and grasses should be available. After three weeks, the newborn rabbits can chew and consume (Romney, 1977). The does can be rebred if their physical conditions are good after one week of kindling (Absolon, 1987).

2.3 PREGNANCY DIAGNOSIS

In general, pregnancy diagnosis necessitates a multifaceted approach involving three main diagnostic tools. These include a medical history and physical examination, laboratory testing, and ultrasonography. At the moment, physicians can use all of these tools to diagnose pregnancy at an early stage and to rule out other pathologies. However, there are several methods of detecting pregnancy in the rabbit industry.

2.3.1 Weight Gain Method

Changes in body weight-There are significant change in body weight from mating up to 30 days. Average gain of around (300-400)g has been suggested from mating to 30 days in large sized rabbit (TNAU Agritech Portal, 2009).

2.3.2 Palpation Technique

Palpation is a technique of feeling with the fingers or hands during an animal's physical examination. Palpation of the abdomen of the doe, which allows embryos to be felt by hand. This should be done about two weeks after mating. This method can only be

perfected with training. Another distinguishing trait is uterine swelling—the uterus can enlarge up to 12 mm 9 days after mating. At 13 days, it may reach 20 mm. Only an experienced keeper may be able to precisely anticipate the changes (Suckow, 2012). Abdominal palpation gave an accurate diagnosis of 10.9 ± 0.3 by day. Manual palpation requires the least amount of effort and does not require any supplies. However, objective findings are only necessary after the 12th day of pregnancy. However, according to Morell (1995), if used excessively, it can put a lot of pressure on the doe and cause foetal loss. If the female is not pregnant, mating should be repeated after palpation. Re-breeding was done a maximum of three times before they were diagnosed as pregnant.

2.3.3 Remating

Aside from that, position the buck near the doe for mating. A buck may not mate with a pregnant female (Absolon, 1987).

2.3.4 Ultrasonography Method

Ultrasonography has been commonly used to detect early pregnancy in domestic animals. This beneficial technological advancement enables for the early, accurate, and practical diagnosis of pregnancy (Mannion, 2007). However, interpreting sonograms of the reproductive system requires an understanding of the image composition as well as an awareness of the potential aberrations that can occur and lead to misdiagnosis. Acoustic augmentation, for example, will show as a hyper echoic patch deep within the fluid (anechoic) area. Gas and bone will completely reflect the sound waves and provide the highest echo signals, resulting in a near-white image on the screen. In rare situations, the return of echoes is so thorough that sound waves do not penetrate deep into these locations, resulting in a lack of imaging that presents as a black zone and is known as acoustic shadowing.

This should not be mistaken for an anechoic image of fluid-filled structures (Mannion, 2007). In most animals, the development of anechoic fluid within the uterine lumen is the first sonographic indication of pregnancy. This fluid continues to rise until the embryo proper is apparent as a hypoechoic structure floating within it, and the fetus structures, as well as the foetal membranes, become more evidently clear. When the foetal heart pulse becomes observable as a hypoechoic flickering structure, the viability of a growing fetus is determined. Goddard (1995) describe the methods of evaluation of pregnancies using ultrasonography for various species in sufficient detail. The rabbit uterus is ultrasonographically examined transcutaneously. The probe is placed externally against the abnormal wall, while the doe is confined in a restraining device in dorsal recumbency (Goddard, 1995). The doe's ventral abdomen is cut, and an ultrasonography scanning gel is applied to the examination location. The overall time required for the sonographic examination (restraint, clipping, and examination) is 5 to 10 minutes (TNAU Agritech Portal, 2009).

2.3.5 Pseudo Pregnancy

False pregnancy, also known as pseudocyesis in the medical term, is the assumption that a parent is expecting an offspring when they are not. With the exception of an actual fetus, parents with pseudocyesis experience many, if not all, of the symptoms of pregnancy (Waters, 1936). Pseudo pregnancy in rabbits can occur as a result of sterile

copulation and lasts 16 to 17 days. She may pluck hair from her body and try to create a nest at the end of this phase, as well as exhibit growth of the uterus and mammary gland (Thomsen, 1968).

2.4 MATERNAL BEHAVIOUR

The idea of a constantly present mother providing continuous care to her offspring is widely accepted as the pattern of maternal care in mammals (Gubernick and Klopfer, 1981). Indeed, mothers in rodents and ungulates (the most studied mammalian orders) exhibit intense interaction with their young throughout most of lactation: they may lick their anogenital region, drink their urine, nurse them frequently, retrieve pups that have strayed away from the nest, emit distress vocalisations if separated from the litter, and show aggression towards intruders approaching the nest. (Numan, 1994). Lagomorphs (rabbits, hares, and pikas) on the other hand, have developed an "absentee" mothering system in which the mother-young relationship is kept to a minimum. Nonetheless, as seen by their global spread, this mothering mechanism has allowed lagomorphs to thrive and inhabit a wide range of habitats (Rosenblatt and Snowdon, 1996).

2.4.1 Nest Building

Lagomorph parents, like other mammals, prepare a nest in which their offspring will be born. Before parturition, snowshoe, *Lepus americanus* (Rongstad and Tester, 1971) and European, *Lepus europaeus* (Broekhuizen and Maaskamp, 1980) Hares which give birth to premature young gather grass from the field, arrange it on the ground in a quiet area chosen for parturition, and form the grass into a nest. The young remain in the nest for the first few days of life, following which they disperse and begin to consume solid food. Nonetheless, the young will return to the nest for the daily feeding period throughout lactation. Pikas, Ochotona princeps (Whitworth, 1984), also build a nest (above ground) out of the surrounding plants, where they nurse. The maternal nest of swamp, Sylvilagus aquaticus (Sorensen et al., 1972) and European, Oryctolugus cuniculus (Gonzalez-Mariscal *et al.*, 1996) is a significantly more complex structure than that of hares and pikas. For this species, nest construction begins a few days before parturition, when females dig multiple shallow basins (swamp rabbits) or real underground burrows (European rabbits). Straw or grass is then removed from the field, collected, brought into basins or burrows, and formed into "straw nests." (Denenberg et al., 1963). As parturition approaches, the mother chooses one of the straw nests to nurse the young. Preparturient females complete the development of the "maternal nest" within this structure. Doe rabbits pluck their densely rooted hair with their mouths and incorporate it into the straw nest to produce a compact "hairy" structure—the maternal nest (Sawin and Crary, 1953). To the best of our knowledge, rabbits are the only mammals that use their body hair to build a maternal nest.

2.4.2 Nursing and Lactating

For European rabbits, the mother-young connection occurs exclusively during nursing, which occurs once a day for a brief period of 3-5 minutes (Gonzilez-Mariscal *et al.*, 1996), 5-10 min in snowshoe hares (Rongstad and Tester, 1971), 10-20 min in swamp rabbits (Sorensen *et al.*, 1972). The duration of the nursing bout in European hares diminishes from roughly 4 to 6 minutes at the start of lactation to 1 to 2 minutes as weaning

approaches (Broekhuizen and Maaskamp, 1980). Pikas are in the middle, nursing the litter for 10 minutes every 2 hours during the first week of lactation, then reducing both the duration and the number of nursing bouts per day (Whitworth, 1984). Despite the brief duration of the nursing experience, altricial rabbit pups (hairless, with closed eyes, and unable to regulate their body temperature) may locate the mother's nipples and sip enough milk to nourish them for the next 24 hours.

This incredibly efficient method of breastfeeding is made possible by the activity of effective mechanisms in both the mother and the child. Hudson and Distel (1983, 1984) have shown that Lactating rabbits generate an olfactory signal (called nipple pheromone) through their ventrum, which is detected by the young's primary olfactory system. The perception of this chemical signal causes the newborns to engage in a stereotyped searching behaviour on the mother's ventrum, allowing them to identify the maternal nipples in seconds, grab them, and suckle. It has to be seen whether such a remarkable mechanism exists in other for lagomorphs to maximise breastfeeding. The processes that enable this system's highly efficient operation are unclear. Behavioural observations in hares and European rabbits, on the other hand, show that both mother and young engage in activities that aid their interaction once every 24 hours.

Lagomorph mothers must make their way back to the nest every day since they leave the nest after each nursing round. Similarly, young hares leave their mother's nest a few days after birth, only meeting her for nursing around the birth site (Broekhuizen and Maaskamp, 1980; Rongstad and Tester, 1971). Though young rabbits do not have to worry about finding their mother's nest, pups in the nest have behaviour that is tuned to the mother's circadian nursing bouts. Pups are quite active shortly before the mother's arrival, exposing themselves from the nest material and appearing very frantic. As a result, when the mother reaches the nest, the pups are fully awake and can immediately begin suckling, guided only by the release of the nipple pheromone from her ventrum (Hudson and Distel, 1982). Aside from the obvious role of providing nutrients for the pups, nursing in rabbits also serves as a way of chemical information transfer. from mother to kits (Bilko *et al.*, 1994; Hudson and Altbacker, 1992).

Milk appears to retain elements of the mother's food, and as pups drink it, they gain information about food preference that is critical for their survival after weaning. Aside from milk, the mother rabbit leaves an average of four fecal pellets before leaving the nest during each nursing period (Hudson and Altbacker, 1992). These pellets appear to contain bacteria from her gut flora as well as residues of the mother's diet. The young nibble on these pellets as they grow older and eat less milk, and may also continue to receive chemical information from their mother. It is uncertain whether this method of passing chemical information from mother to litter works in other lagomorphs.

2.4.3 Body Weight Changes

According to Agaviezor and Ologbose (2020), the physiological changes that occur in the doe's body during pregnancy may explain the positive changes in body weight before mating and during pregnancy.

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2.4.4 Gestation Length

According to Biggers *et al.* (1963), the number of conceptuses determines the gestation period. Because there are roughly the same number of corpora lutea in the ovaries as there are foetuses in the uterus in a normal pregnancy, it is possible that the ovaries control gestation rather than the uterus. In contrast, Manresa (1933) found a significant negative correlation between gestation length and litter size in rabbits, and while Hammond (1925) claims there is no inverse relationship between gestation length and litter size in rabbits, his data show a highly significant negative regression of gestation length on litter size.

2.5 DOES PERFORMANCE

2.5.1 Conception rate

Kishk *et al.* (2006) found that mating in the morning results in a high percentage of pink and a dark red vulva colour, as well as the highest sexual responsiveness. These findings indicate that genital characteristics such as colour and turgidity are closely related to sexual behaviour and are influenced by hormones. White vaginal vulva, on the other hand, is associated with a higher rate of male rejection (Marai and Rashwan, 2003).

2.5.2 Number of Kits at Birth

Litter size is influenced by genetics and farm management, which includes housing, feeding, and breeding. Female rabbits fed a controlled diet had high reproductive efficiency, a large number of litters, and excellent milk production (Eiben *et al.* 2001). Furthermore, rabbits that mated while in standing heat (receptive) and had a red and swollen vulva had a

higher litter size and conception rate. (Szendrő *et al.*, 2006) According to Fayeye and Ayorinde (2010), the number of litter size in Nigerian rabbits was lower during the rainy season than during the summer season. According to Mousa (2004), the age of the buck influenced the litter size (number of kits). They discovered that does mate with a buck of an average age of 8 months had the largest litter size (7 kits), while does mate with a buck of an average age of 6 months had 6.2 kits, and does mate with a buck of an average age of 4 months had 3.4 kits.

2.5.3 Number of Kits Born Alive

Pre-weaning mortality can be influenced by genotype, pathological cause, doe characteristics such as litter size, birth weight, frequency of parturition, order, season of birth, management, nest quality, and weather conditions. Genetics, weaning weight, kit age, and external environmental conditions after weaning may all have an impact on young rabbit mortality. Pre-weaning mortality rates differ by breed and between purebred and crossbred rabbits. Another study found that the genetic variability of rabbit pre-weaning mortality was 12 %, with enteritis and pneumonia being the leading causes. The advantage of the maternal component of heritability over the paternal component demonstrates the mother's significant influence on the viability of the young (Asghar *et al*, 2015). Although undesirable characteristics can be corrected, genetic advancement can help to reduce pre-weaning mortality. Scott *et al.* (1998) discovered that crossbred animals were more efficient than purebred animals at showing maternal behaviour, producing young per litter and nurturing them. Habeeb *et al.* (1997) discovered that New Zealand White (NZW) rabbits had a higher

survival rate than California rabbits from 5 to 14 weeks of age (88% vs 81%). Pre-weaning mortality increased insignificantly as doe weight at conception and kindling increased.

2.5.4 Litter Size

According to Poigner *et al.* (2010), the number of rabbits in the uterus determines the foetal and birth weight. The maternal influence had a significant influence on birth weight and litter size. According to Agaviezor and Ologbose (2020), litter size has a significant impact on rabbit litter weight. The weight of the doe before and after mating also had an impact on the weight of the rabbit litter. Increasing a doe's body weight before and after kindling could result in an increase in litter weight. In their study on the effect of birth weight and litter size on rabbit growth and mortality, Poigner *et al.* (2010) reported that kits from larger litter sizes had lower weight at weaning than kits from smaller litter sizes.

As the litter size at birth increased, so did the percentage of newborns who died during the suckling phase (Caro *et al* 1984). Mortality rises in both small and large litters. According to, pre-weaning mortality in New Zealand White (NZW) rabbits was reduced in litters of 5 or 7 young (Askar, 1989). According to some research, there is no link between litter size and mortality. The increase in preweaning mortality with increased litter size at birth appears to be due to a decrease in average individual weight per litter at birth. Furthermore, as litter sizes grow, so does competition for teats, resulting in smaller rabbits receiving less milk (Afifi *et al.*, 1973).
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CHAPTER 3

MATERIALS AND METHODS

3.1 EXPERIMENTAL ANIMAL

About 8 does and 4 bucks of heterogenous New Zealand White (NZW) breed, *Oryctolagus cuniculus* (Figure 1) housed in individual cages at the University Malaysia Kelantan Campus Jeli Rabbit Farm are used for this study. The does are not lactating but varied in parity from 1-4 and body weight from 2,400-3,250g at the commencement of the experiment. The bucks weighing between 2200–3,400g are all intact and of proven fertility. The animals are exposed to the prevailing 12:12 light:dark hour cycle and offered 60g/animal/day (about 3% of body weight) of compounded diet containing 2400 kcal digestible energy/kg dry matter (DM) and 18% of crude protein as well as *ad libitum* mixture of *Pennisetum purpureum* (Napier grass) and *Leucaena leucocephala* (River tamarind) foliages. Water is also supplied ad libitum.





Figure 1 shows New Zealand White (NZW) rabbit breed, Oryctolagus cuniculus

3.1.1 Treatment

Group 1 which is doe 1, doe 2, doe 3 and doe 4 will be mate with the male rabbits for 1 day only. Meanwhile, for Group 2 which consist of doe 5, doe 6, doe 7 and doe 8 will be mate with the male rabbits for 3 days continuously.

3.2 DATA COLLECTION

3.2.1 Mating Behaviour

Behavioural attitude of the does monitored includes: aggressiveness toward the male; flattening to a corner of the cage; circling within the cage; allowing mounting but no lordosis posture (Figure 2); and allowing mounting with a lordosis posture (Figure 3). The time interval between the introduction of the female into the male cage and the actual mating is noted with a stop clock. In determining this parameter, the pair is first allowed full 5

minutes for spontaneous copulation in the absence of which mating is assisted or forced as described above within additional 5 minutes. Mating readiness or Acceptance rate is scored as a variable of Bernoulli: Refusal to mate and assisted or forced mating=0; spontaneous mating with or without lordosis posture=1.



Figure 2 shows the allowing mounting behaviour



Figure 3 shows lordosis

3.2.2 Early Pregnancy Sign

The does are thereafter monitored throughout pregnancy. Abdominal palpation (Figure 4 is done on the 15th day post mating and non-pregnant does are remated. At delivery litter size and weight are recorded. Fertility index is calculated as the product of litter size and kindling rate.



Figure 4 shows abdominal palpation technique

The ultrasonographic examination (Figure 5) of the rabbit uterus is performed transcutaneously on the 10th until 15th day post mating. The probe is positioned externally against the abdominal wall, while the doe is restrained at dorsal recumbency in a restraining device. It is imperative, for optimal image quality, that the space between the probe and the animal's skin is free of air. For this reason, prior to scanning, the ventral abdomen of the doe is clipped and ultrasound scanning gel is applied at the examination site The sonographic

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examination proceeds from caudal to cranial. The probe is positioned directly in front of the pubic bone and held in a sagittal plane. At this position, the image of the long vagina can be seen in a longitudinal section, as an echogenic linear tubular structure. The probe is then moved cranially, following the course of the uterus, while its beam is directed either longitudinally or transversely to it. Due to its small diameter, it is almost impossible to sonographically distinguish a non-gravid uterus, at a cross section, among the intestinal gyri. The total time needed for the sonographic examination (restraint, clipping, examination does not exceed 5 to 10 min. Clipping takes a few minutes longer when non-lactating does are to be examined, since more hair is present on their ventral abdomen.



Figure 5 shows ultrasound of rabbit

3.2.3 Post-Pregnancy Behaviour (Maternal Behaviour)

Maternal behaviour before parturition day of the does monitored includes: the does collect material for constructing the straw nest (Figure 6); the does shredded excelsior ward

and shaped as a nest (Figure 7). In determining this parameter, the does is prepared with the straw, excelsior ward and other material in order to build and shaped the nest. The does will be observed until kindling. Maternal behaviour is scored as a variable of Bernoulli: Refusal to build a nest=0; build a nest=1.

Maternal behaviour after parturition monitored includes: the does lick, feed and sniff their kits (Figure 8); the does refuse to milk their kits. In determining this parameter, the does will be observed until for 2 days. Maternal behaviour is scored as a variable of Bernoulli: Refusal to nurture the kits = 0; nurture the Kits = 1.



Figure 6 shows rabbit nest building





Figure 7 shows rabbit nest



Figure 8 shows doe feed her kits milk

3.2.4 Fertility parameter of does

Fertility parameters of does were estimated as

1. Conception rate = $\frac{No.of \ pregnant \ does}{Total \ no.of \ mating \ does} \times 100\%$

2. Parturition rate $=\frac{No.of \ delivering \ does}{Total \ no.of \ mated \ does} \times 100\%$

3. Litter size at birth: Number of kits at birth

3.3 STATISTICAL ANALYSIS

Compare the effect of different mating frequencies on the maternal and mating behaviour, conception rate, gestational length, litter size and kit's growth alive in New Zealand White rabbits. Data recorded as percentage of occurrence are statistically compared by independent t-test.



CHAPTER 4

TAP FIAT

RESULTS AND DISCUSSION

Rabbits reproduce at a higher pace than other livestock. They are sexually mature within a few months of birth and have brief pregnancies. They have large litters and are unique in that they can be rebred soon after they are kindled (giving birth). With an aggressive breeding programme, a doe can have 60 weaned young each year. Such intensive breeding, however, is not recommended for novices and is rarely used in commercial production (Michigan State University, 2017). According to many breeders, mating continually for three days will effectively boost the doe's chances of becoming pregnant. A young buck, on the other hand, may be permitted to mate with one doe every 3 to 4 days. However, after 12 months of age, it may mate 4-6 times in 7 days. Bucks above the age of six should be culled since their sperm quality deteriorates. Aside from that, some bucks get tired too quickly. They just run out of energy after constant mating and are unable to complete the entire mating process. As a result, this study has implemented alternative mating frequencies in order to enhance mating behaviour, maternal behaviour, and conception rate in New Zealand White (NZW) does.

4.1 THE EFFECT OF DIFFERENT FREQUENCIES OF MATING ON MATING BEHAVIOUR, MATERNAL BEHAVIOUR AND CONCEPTION RATE IN NEW ZEALAND WHITE DOES.

Mating Behaviour (%)		*Group	
Denaviour (%)	G1 (n=4)	G2 (n=4)	P-value
Day 1	100 (4)	100 (4)	-
Day 2	-	25 (1)	0.5370
Day 3	-	50 (2)	0.1340

Table 1 showed analysis of mating behaviour observation

*G1 - mating once, G2 mating continuously for 3 days.

In this study, mating continuously for 3 days was concluded to have no significant effect on the mating behaviour, maternal behaviour and conception rate in New Zealand White (NZW) does. Harriman (1995) describes courtship behaviours during mating as ear sniffing, genital sniffing, chin stroking, leg striking, and backward or sideways falling after mating. Honking or oinking: Sounds similar to "honk" or "oink" are regularly used to attract attention. Even with neutered rabbits, honking is frequently associated with desire mating. Table 1 showed the mating behaviour for Group 1 (mating once) and Group 2 (mating continually for 3 days). The results showed that on the first day, both Group 1 and Group 2 exhibited 100% mating behaviour. On the second day, 25% increase in mating behaviour was seen in Group 2. Mating behaviour in Group 2 revealed better outcomes than in Group 1, although there is no significant difference between the groups. Mating behaviour may be influenced by seasons, time of mating, mating order, receptive doe, body weight and age of rabbits at mating. In this study, natural mating was done on the rabbits between the 8 a.m. and 10 a.m., and receptive does were chosen.

When a male is exposed to a series of susceptible females, he may copulate *ad libitum* and exhibit an enormous number of mounts, intromissions, and ejaculations until sexual activity terminates. Bucks may perform six ejaculations in 30 minutes, according to Melin and Kihlström (1963), with the first one occurring 19 seconds after the doe was introduced. No distinction was noted between mounts that resulted in ejaculation and those that did not. Overall, the trials imply that male rabbits can achieve sexual fulfilment if they are permitted to copulate freely with a sequence of receptive does for one day. Larsson (1979) found that the number of mounts that did not terminate in copulation did not alter significantly across test days in rabbits, while the number of mounts that culminated in ejaculation decreased dramatically between the first and last day of testing.

Post-ejaculatory latency in rats, on the other hand, increased with each copulatory series. The number of mounts without intromission is largest in the first ejaculation series, decreases in the second and third series, and increases in the fourth series in the rat. According to Jiménez *et al.* (2012), the buck is frequently driven to participate in sexual activity at the start of testing since they are unable to complete ejaculation after many days of *ad libitum* courtship. Furthermore, no significant changes in the animals' day activity were seen following sexual fatigue tests, showing that the reduction in sexual activity and chinning is due to the stimulation of inhibitory systems rather than a general physical depletion. As a result, the percentage of mating behaviour for Group 2 decreases after three days of mating.



Figure 9 showed percentages of pregnant does in Group 1 and 2 using abdominal palpation.

*G1 - mating once,	G2 mating	continuously	for 3 days.
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Palpation is a technique using fingers or hands during a physical examination of an animal for medical check-up or pregnancy diagnosis. This technique was carried out on doe's abdomen, allowing the shape of foetuses to be examine by hand. This should be done two weeks after mating. Repetition and continuous training are necessary for skilled technicians. Uterine swelling is another distinctive feature whereas the uterus can grow up to 12 mm after 9 days of mating. It may reach 20 mm after 13 days. Changes may be predicted by skilled technician (Suckow, 2012).

Figure 9 showed no significant differences on the percentages of positive pregnancies between Group 1 and Group 2. Even though, higher positive pregnancy rate was observed on Day 13 onwards for Group 2, there was no significant differences between groups. This finding is consistent with Harcourt-Brown (2002) assertion that the preferred time for pregnancy detection is 10-14 days after mating, when the foetal units can be felt as olivesized masses. The handler's experience also may have a significant impact on the accuracy of abdominal palpation diagnosis. Furthermore, the diagnosis with abdominal palpation were 100% accurate because the positive pregnancies were similar with parturition in both groups.



Figure 10 showed ultrasound imaging in G1 (A) and G2 (B) on the 21st day of pregnancy.

Ultrasonography has long been used in domestic animals to detect early pregnancy. This advantageous technological innovation allows for the early, accurate, and practical detection of pregnancy (Mannion, 2007). Interpreting sonograms of the reproductive system, on the other hand, necessitates a comprehension of image composition as well as awareness of various aberrations that can arise and lead to misdiagnosis. Acoustic augmentation, for example, will manifest itself as a hyper echoic patch deep within the fluid (anechoic) region. Gas and bone totally reflect sound waves and generate the strongest echo signals, resulting in a near-white image on the screen.

Based on the Figure 10 showed ultrasound imaging Group 1 (A) and Group 2 (B) on Day 21 of gestation. A total of 75% and 100% pregnant does were detected in Group 1 and Group 2, respectively. Positive signs of pregnancy indicate white structure that would appear to be the femur or ribs on the abdomen of doe. Movement of the foetus also

indicates pregnancy in does.

In this study, in comparison between ultrasound and abdominal palpation, the suitability for pregnancy diagnosis in rabbit is manual abdominal palpation. Morrell (1995) also suggested that this method requires the least amount of effort and requires dexterity and skill from Day 12 of pregnancy onwards. However, if used excessively, it can put a lot of strain to the doe and cause foetal loss (Morrell, 1995). The use of ultrasound scanning of the abdomen has proven to be a safe and practical method of detecting pregnancy at an early stage. The total amount of time required, including restraint, clipping, and examination, is never more than seven minutes. In rabbits, ultrasound scanning of the abdomen is also useful in diagnosing extrauterine pregnancy, ovarian tumours, abscesses, cysts, pyometra, and hydrometra (Varga, 2014). Plus, pregnancy can be detected in does between Day five and nine, which is as early as or earlier than other similar studies have reported (Ypsilantis and Saratsis, 1999).

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Figure 11 showed analysis of pregnancy diagnosis through mating with pregnant doe test. *G1 - mating once, G2 mating continuously for 3 days.

Figure 11 showed no significant differences on the percentages of positive pregnancies between Group 1 and Group 2 through mating with pregnant doe. Even though, higher positive pregnancy rate was observed on Day 13 onwards for Group 2, there was no significant differences between groups. Absolon (1987) stated in his study that a buck may not mate with a pregnant female. This is similar with the result obtained from abdominal palpation diagnosis in Figure 9. Thus, we would suggest that mating with pregnant does test showed the same efficacy as abdominal palpation in rabbit.

4.1.1 Post-Pregnancy Behaviour (Maternal Behaviour).

Rabbits have developed an "absentee" mothering system in which the mother-young relationship is minimised. Nonetheless, as evidenced by their global distribution, this mothering mechanism has allowed rabbits to thrive and inhabit a diverse range of habitats

(Rosenblatt and Snowdon, 1996). As the time for parturition approaches, the mother selects one of the straw nests to nurse the young. Within this structure, preparturient females complete the development of the "maternal nest." Doe rabbits use their mouths to pluck their densely rooted hair and incorporate it into the straw nest, resulting in a compact "hairy" structure—the maternal nest (Sawin and Crary, 1953). To our knowledge, rabbits are the only mammals that use their body hair to construct a maternal nest.

Nest Building (%)		*Group	
	G1 (n=4)	G2 (n=4)	
Day 29	75 (3)**	0**	
Day 30	75 (3)	50 (2)	
Day 31	75 (3)	100 (4)	
Day 32	75 (3)	100 (4)	
Day 33	75 (3)	100 (4)	

Table 2 showed analysis of maternal behaviour observation through nest building.

*G1 - mating once, G2 mating continuously for 3 days.

**P<0.05

As shown in Table 2, there were significant differences (P<0.05) in nest building behaviour between Group 1 (75%) and Group 2 (0%) on Day 29, respectively. However, from the 30th to the 33rd day, there were no significant differences (P>0.05) between the two groups. In our study, the result showed that nest building behaviour is highly positive on one day before or on the day of parturition for both groups.

This nest-building behaviour is common in rabbits. In the case of the rabbit, the nestbuilding behaviour appears to begin with a straw nest, followed by a fur-lined nest. According to Zarrow *et al.* (1961), the rate of maternal nest building among normal pregnant does was 98%. A total of 86.7% built the nest on the day of parturition, 82% built nests a day or more before parturition, and 5.1% built nests immediately after the kits were born.

The significant result on the 29th day could be attributed to the buck's routine of mating repeatedly. On the first day, four bucks are permitted to mate with eight does, beginning with doe 1 in Group 1 and ending with doe 8 in Group 2. On second and third day, the bucks only mating with the does from Group 2. The buck may have exhausted from the mating of Group 1. Hence, result in the lack of efficiency in the mating of Group 2 on the first day. This is not the case for the second and third day. As a result, it will cause slight delay in the maternal behaviour for doe in Group 2.

It was worth mentioning, that all pregnant does demonstrated maternal behaviour through nest building in this study. This finding was related to the endocrine control of nest building behaviour via prolactin hormone secretion. The rabbit maternal behaviour study (Zarrow *et al.*, 1961) provides evidence for the endocrine system's potential influence in one component of maternal behaviour (nest building). Nest building behaviour is especially useful because it has been demonstrated in other animals with an endocrine component, particularly birds, but appears to be unique to a few species of lagomorphs (Cahalane, 1947).

Normally, the hair used for nest building becomes insufficient for heat resistance when the straw nest or other material becomes inadequate for heat resistance, which may increase mortality due to chilling (Schlolaut *et al.*,1984). The increase in litter temperature has resulted in a decrease in hair nest construction. Minor changes in the external environment have little effect on the climate, but they can influence the nest's construction. Temperature appears to be more important because it influences the amount of fur used for the nest layer (Szendro' *et al.*, 1998). Rather than the doe's experience, various factors that play a role in the formation of the hair nest are related to specific stimuli such as hormone changes and environmental temperature (Zarrow *et al.*, 1961).

Nursing and		*Group	
Lactating (%)	G1 (n=4)	G2 (n=4)	P-value
Day 33	0	75 (3)	0.0240
Day 34	25 (1)	75 (3)	0.1340
Day 35	50 (2)	75 (3)	0.5370
Day 36	50 (2)	75 (3)	0.5370
Day 37	50 (2)	75 (3)	0.5370

Table 3 showed analysis of maternal behaviour observation through nursing and lactating.

*G1 - mating once, G2 mating continuously for 3 days.

According to Table 3, there were significant differences (P<0.05) in nursing and lactating behaviour between Group 1 and Group 2 on the 33^{rd} . Only 0% of Group 1 and 75% of Group 2 express nursing and lactating behaviour after parturition on the 33^{rd} . In this study, three does (G1= 2 does, G2- 1) experienced natural mating for the first time. As a result, the lack of maternal experience may influence the does' nursing and lactating behaviour.





Figure 12 Nursing, Lactating and Mortality rate in Group 1 and Group 2.

Nursing, lactating and mortality rate (%)

According to El-Kholya (2011), a good mother can suckle a large number of kits while maintaining their vitality, thereby reducing pre-weaning mortality. As a result, as shown in Figures 12, the lower the percentage of nursing and lactating kits, the higher the mortality rate of the kits. Pre-weaning mortality caused by diseases, climatic conditions, and maternal behaviour (nest building and lactation). Yamani (1991) reported that rabbit kits are completely dependent on does milk for feeding during the first 20 days of lactation. The failure of a doe to nurse her young is most likely due to nipple tenderness, and the young rabbit will die as a result of the doe's inability to provide milk. According to Schlolaut (1980), mortality rate is also influenced by insufficient milk supply and the ability of young animals to successfully discover teat during one a day for 2-3 minutes. When there is a lack of

postnatal maternal ability due to limited milking and suckling abilities, the mortality rate rises.

4.2 THE EFFECT OF DIFFERENT FREQUENCIES OF MATING ON GESTATIONAL LENGTH, LITTER SIZE AND KIT'S GROWTH IN NEW ZEALAND WHITE DOES.

Another goal of implementing different mating frequencies was to optimise gestational length, litter size, and kit growth in New Zealand White (NZW) does. In this study, however, continuous mating for three days was found to have no significant effect on the gestational length, litter size, or kit growth in New Zealand White (NZW) does.

Types of d <mark>oe</mark>	Litter size	Gestation length (day)	Mortality rate (%)
G1	2.75 ± 1.89	24 days	47.6
G2	3.50 ± 1.32	32 days	36.4
P-value	0.7559	0.3856	ſ

Table 4 showed analysis of litter size, gestation length of the doe and mortality rate of kits.

*G1 - mating once, G2 mating continuously for 3 days.

Table 4 depicts the findings of this study, which showed that there is no significant difference in gestation length and number of kits after kindling between Groups 1 and 2. This can be explained by saying that the length of gestation is determined by the total number of foetuses in a female. According to Biggers *et al.*, (1963), the number of conceptuses determines the gestation period. Because there are roughly the same number of corpora lutea in the ovaries as there are foetuses in the uterus in a normal pregnancy, it is possible that the

ovaries control gestation rather than the uterus. In contrast, Manresa (1933) found a significant negative correlation between gestation length and litter size in rabbits, and while Hammond (1925) claims there is no inverse correlation between gestation length and litter size in rabbits, his data showed a highly significant negative regression of gestation length on litter size.

According to Table 4, there is no significant difference (p>0.05) in the gestation length of the doe between Group 1 and Group 2. Kadry and Afifi (1983) discovered that buck performance and number of services had no effect on litter size at birth. In contrast, Nelson *et al.* (1979) and Toson *et al.* (1995) found a significant difference in litter size between does mate with different numbers of services. Does that mated by two services from different bucks had larger litter size than does mated by two services from the same buck or one service.

Litter size could be affected by different factors such as controlled diet (Eiben *et al* 2001), doe showed indication of red and swollen vulva (Szendro *et al.*, 2006), season (Fayeye and Ayorinde, 2010) and age of buck (Mousa, 2004). Female rabbits fed a controlled diet had high reproductive efficiency, a large number of litters, and excellent milk production (Eiben *et al.* 2001).

Furthermore, rabbits that mated while in standing heat (receptive) and had a red and swollen vulva had a higher litter size and conception rate. (Szendro *et al.*, 2006) According to Fayeye and Ayorinde (2010), the number of litter size in Nigerian rabbits was lower during the rainy season than during the summer season.

According to Mousa (2004), the age of the buck influenced the litter size (number of kits). They discovered that does mate with a buck of an average age of 8 months had the

largest litter size (7 kits), while does mate with a buck of an average age of 6 months had 6.2 kits, and does mate with a buck of an average age of 4 months had 3.4 kits.

Statistical analyses of the doe's litter size in Table 4 also revealed that Group 2 had a lower mortality rate (36.4%) than Group 1 (47.6%). Pre-weaning mortality can be influenced by genotype, pathology, doe characteristics such as litter size, birth weight, frequency of parturition, order, season of birth, management, and nest quality, disease, and weather conditions (Rashwan *et al.*, 2010).

Pre-weaning mortality rates differ by breed and between purebred and crossbred rabbits. Another study found that the genetic variability of rabbit pre-weaning mortality was 12%, with enteritis and pneumonia being the leading causes. The advantage of the maternal component of heritability over the paternal component demonstrates the mother's significant influence on the viability of the young (Asghar *et al.*, 2015).

Does mated to younger bucks with an average age of 4 months had a higher preweaning mortality rate than does mated to older bucks with an average age of 7 months. Furthermore, the number of services mated had an effect on pre-weaning mortality percentages, with does mate two services, either from different or same buck, having a lower percentage than does mate one service only. Does remated at 1 and 5 days after parturition had a significantly higher pre-weaning mortality rate than does remated at 10 and 15 days postpartum. This discovery explained why the remating intervals were so important in this feature. Does that remated postpartum cause a decrease in milk production (Mendez *et al.*, 1986).



Figure 13 showed analysis of weekly average weight of doe. *G1 - mating once, G2 mating continuously for 3 days.

Figure 12 revealed that there is no significant difference (P>0.05) in the body weight changes of does in Groups 1 and 2. However, doe in Group 1 gained 280 g of body weight and doe in Group 2 gained 160 g of body weight during pregnancy (15^{th} days). This finding agreed with Agaviezor and Ologbose (2020), who stated that the physiological changes that occur in the doe's body during pregnancy could explain the positive differences in body weight before mating and during pregnancy. The findings agreed with those of Oguike *et al.* (2008), who discovered no significant difference in the body weight of pregnant doe rabbits at mating and parturition. Meanwhile, De Blas *et al.* (1985) discovered that the body weight of pregnant doe body weight change during pregnancy, the literature does not take litter size or doe body weight range into account (Russell *et al.*, 2013).



Figure 14 showed analysis of weekly average weight of kits for 10 days. *G1 - mating once, G2 mating continuously for 3 days.

The analysis of weekly average weight of kits for 10 days in Groups 1 and 2 can be seen in Figure 13. There is no statistically significant difference between these two groups (P>0.05). The weight of a kit is affected by genetics and farm management, which includes housing, feeding, and breeding (Eiben). According to Zerrouki *et al.* (2012), the average weight of kits at birth increased with the amount of milk produced until the 21st day, regardless of litter size. This increase in milk production with kit weight was most likely due to the females giving birth to heavier kits being in better physiological condition than those producing weak young rabbits at kindling. According to Lukefahr *et al.* (1983), the viability of the young rabbit between birth and weaning is dependent on live weight at birth.

CHAPTER 5

CONCLUSION AND RECOMMENDATION

5.1 CONCLUSION

In conclusion, different frequencies of mating between Group 1 (mating once) and Group 2 (mating continuously for 3 days) showed no significant differences on the effect on mating behaviour, maternal behaviour and conception rate in New Zealand White (NZW) does. In addition, different frequencies of mating in Group 1 and Group 2 also showed no significant differences on gestational length, litter size and kit's growth in NZW.

This study suggested and contributed additional data and information on the effect of different mating condition to the conception rate and litter size in NZW does. This is crucial due to the preference of NZW breed among farmers in Malaysia. Nonetheless, future work needs to be carried out for accurate and reliable result.

5.2 **RECOMMENDATION**

A few recommendations would be suggested for future work. First, facilities in rabbit housing in University Malaysia Kelantan (UMK) can be improved to ensure execution of mating trial is unhindered. For example, an indoor or closed system that would regulate the temperature of rabbit house. An additional of fan would be preferred especially from May until mid-September where the surrounding temperature is too hot and not favourable for rabbits. This is due to myriad research that suggest temperatures above 32.2°C and excessive humidity do not suit rabbits. In the summer, cooling system are set up at industrial level ensuring proper breeding and growth.

Furthermore, the study suggest that a specific diet would be given to does for good reproductive efficiency, high number of litter size, and great milk production. Pregnant does' special dietary such as *Leucaena leucocephala* (River tamarind) foliages was a common practice and believed to boost the doe's milk production rate. Thus, further understanding and determination on the level of the rabbit's reproduction can be achieved by execution of analysis on special dietary throughout gestation period.

Last but not least, the frequency of ejaculations and repetitive service from buck may result in sexual weariness and affect conception rate. Added parameters such as weekly rest for the bucks and sperm quality analysis is highly recommended. The information can optimized the rate of reproduction in rabbit industry especially in Malaysia.



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APPENDIX A



Figure 15 showed weighing of does



Figure 16 showed cutting the grass to make hays



Figure 17 showed collecting hays



Figure 18 showed rabbit mating





Figure 19 showed abdominal palpation



Figure 20 showed hair clipping of the does


Figure 21 showed ultrasonography test



Figure 22 showed doe 7 nest building



Figure 23 showed doe 8 nest building



Figure 24 showed doe 8 giving birth





Figure 25 showed disposing the dead kits



Figure 26 showed assist milking of the rabbit





Figure 27 showed weighing of the kits



Figure 28 showed the kits of doe 7 all grown up



APPENDIX B

Week	Types	of doe	Weight (kg)	Total weight (kg)	Average total weight (kg)
		Doe 1	3.05		
	Group	Doe 2	2.6	10.85	2.7125
	1	Doe 3	2.4		
Week		Doe 4	2.8		
1		Doe 5	3.1		
	Group	Doe 6	2.7	12.15	3.0375
	2	Doe 7	3.25		
		Doe 8	3.1		
		Doe 1	3.3		
	Group	Doe 2	2.65	11.65	2.9125
	1	Doe 3	2.65		
Week		Doe 4	3.05		
2		Doe 5	3.2		
	Group	Doe 6	2.4	12.55	3.1375
	2	Doe 7	3.55	E.K.	
		Doe 8	3.4		
		Doe 1	3.45		
	Group	Doe 2	2.75	11.95	2.9875
	1	Doe 3	2.75	λV	$C \perp \lambda$
Week	11	Doe 4	3		O I F
3		Doe 5	3.25		
	Group	Doe 6	2.45	12.8	3.2
	2	Doe 7	3.65	N T P	1.1. 7.
	K	Doe 8	3.45		AN
		Doe 1	3.5		

Table 5 showed weight of does for 7 weeks.

	~	Doe 2	2.75	12.2	3.05
	Group	Doe 3	2.8		
	1	Doe 4	3.15		
Week		Doe 5	3.35		
4	Group	Doe 6	2.6	13.15	3. <mark>2875</mark>
	2	Doe 7	3.6		
		Doe 8	3.6		
		Doe 1	3.55		
	Group	Doe 2	2.7	12.35	3.0875
	1	Doe 3	2.9		
Week		Doe 4	3.2		
5		Doe 5	3.3		
	Group	Doe 6	2.75	13.25	3.3125
	2	Doe 7	3.75		
		Doe 8	3.45		
		Doe 1	3.42		
	Group	Doe 2	2.73	12.04	3.01
	1	Doe 3	2.62		
Week		Doe 4	3.27		
6		Doe 5	3.06		
	Group	Doe 6	2.88	12.96	3.24
	2	Doe 7	3.49		
		Doe 8	3.53		
		Doe 1	3.48		
	Group	Doe 2	2.73	12.5	3.125
	1	Doe 3	2.95	LIN	
Week		Doe 4	3.34		
7		Doe 5	3.14		
	Group	Doe 6	2.86	12.7	3.175
	2	Doe 7	3.29	AY	
	-	Doe 8	3.41		~ * * *

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	Average total weight, kg		
Week	(x)	x-xbar	(x-xbar)^2
1	2.7125	-0.271071 <mark>429</mark>	0.07347972
2	2.9125	-0.071071 <mark>429</mark>	0.005051148
3	2.9875	0.003928 <mark>57</mark> 1	1.54337E-05
4	3.05	0.066428 <mark>571</mark>	0.004412755
5	3.0875	0.10392 <mark>857</mark> 1	0.010801148
6	3.01	0.026428571	0.000698469
7	3.125	0.141428571	0.020002041
Sums	20.885	-3E-09	0.114460714

Table 6 showed average total weight of doe (kg), x-xbar and (x-xbar)2 of Group 1.

Table 7 showed calculation square Group 1.

	Ungroup	Command
n	7	
xbar (mean)	2.983571429	
Sum(x-		
xbar)^2	<mark>0</mark> .114460714	
s^2	<mark>0</mark> .019076786	0.019076 <mark>786</mark>
S	<mark>0</mark> .138118738	0.138118 <mark>738</mark>
SEM	0.052203976	

Table 8 showed average total weight of doe (kg), x-xbar and (x-xbar)2 of Group 2.

Week	Average total weight, kg (x)	x-xbar	(x-xbar)^2
1	3.0375	-0.161071429	0.025944005
2	3.1375	-0.061071429	0.003729719
3	3.2	0.001428571	2.04082E-06
4	3.2875	0.088928571	0.007908291
5	3.3125	0.113928571	0.012979719
6	3.24	0.041428571	0.001716327
7	3.175	-0.023571429	0.000555612
Sums	22.39	-2.22045E-15	0.052835714

	Ungroup	Command
n	7	
xbar (mean)	<mark>3</mark> .198571429	
Sum(x-		
xbar)^2	0.052835714	
s^2	<mark>0.</mark> 008805952	0.008805 <mark>952</mark>
S	0.093840036	0.093840 <mark>036</mark>

0.0354682

Table 9 showed calculation square Group 2.

Table 10 showed weight of kits for10 days.

SEM

Day	Types	of doe	Weight of kits (g)	Total weight (g)	Average total weight (g)	
		Doe 1	35			
	Group	Doe 2	0	108.4	27.1	
	1	Doe 3	33.8			
		Doe 4	39.6			
Day 1		Doe 5	30.5			
	Group	Doe 6	43.3	118.7	47.175	
	2	Doe 7	63.2			
		Doe 8	51.7]		
		Doe 1	0		DC	T T
	Group	Doe 2	0	96.8	24.2	
	1	Doe 3	37.5			
		Doe 4	59.3			
Day 5		Doe 5	0		~ ~	· ·
	Group	Doe 6	66.3	280.2	70.05	1 /
	2	Doe 7	108.6	11		1 1
		Doe 8	105.3			
		Doe 1	0			
Dav	Group	Doe 2	0	171.1	42.775	AN
10	1	Doe 3	63.8			
	-	Doe 4	107.3			

	Doe 5	0		
Group	Doe 6	118.2	499.1	124.775
2	Doe 7	163.6		
	Doe 8	217.3		

Table 11 showed average total weight of kits (g), x-xbar and (x-xbar)2 of Group 1.

Day	Average total weight, g (x)	x-xbar	(x-xbar)^2
1	27.1	-4.258333333	18.133403
5	24.2	-7.158333333	51.241736
10	42.775	11.41666667	130.34028
Sums	94.075	0	199.71542

Table 12 showed calculation square Group 1.

	Ungroup	Command
n	3	
xbar (mean)	<mark>3</mark> 1.35833333	
Sum(x-xbar)^2	<mark>. 1</mark> 99.7154167	
s^2	99.85770833	99.85770 <mark>833</mark>
S	9.992882884	9.992882 <mark>884</mark>
SEM	5.769393623	

 Table 13 showed average total weight of kits (g), x-xbar and (x-xbar)2 of Group 2.

Day	Average total weight, g (x)	x-xbar	(x-xbar)^2
1	47.175	-33.49166667	1121.691736
5	70.05	-10.61666667	112.7136111
10	124.775	44.10833333	1945.545069
Sums	242	0	3179.950417



	Ungroup	Command
n	3	
xbar (mean)	80.66666667	
Sum(x-xbar)^2	<mark>3</mark> 179.950417	
s^2	1589.975208	1589.975 <mark>208</mark>
s	<mark>39.8744932</mark>	39.8744 <mark>932</mark>
SEM	<mark>23.</mark> 02154939	

Table 14 showed calculation square Group 2.

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