

# Performance, Follicle Diameter, Response to Estrous Synchronization, and Productivity of Lao-Native Goats That Received Addition of Paper Mulberry (*Broussonetia papyrifera*) Leaf-Based Diets with Concentrate

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## Abstract

The objective of this research was to investigate the effects of supplementation of the paper mulberry leaf-based diets with concentrate on growth performance, preovulatory follicle diameter, response to estrous synchronization treatment, and productivity in Lao-native goats. Group 1, female goats were given the paper mulberry leaves (PML) as the basal diet (PML diet; n=12). Group 2, female goats were given the PML as the basal diet and 400 g/head/day of the concentrate (PML+C diet; n=12). The periods of estrus were synchronized with controlled intravaginal drug release (CIDR) devices inserted for 14 days and injections of 300 IU of human chorionic gonadotropin upon the removal of CIDR devices. The ovaries were scanned on the day of CIDR removal and every 12 h after the withdrawal of CIDR to evaluate the preovulatory follicle diameter. The positive changes in the live weight of the female goats in the PML+C group were greater than those of the PML group. The proportion of synchronized goats displaying estrus at 48 h after the withdrawal of CIDR was significantly higher in the group that received the PML+C diet than in the group that received the PML diet. The productivity tended to be greater in the goats that received the PML+C diet than in goats that received the PML diet. Thus, these data highlight that the positive changes in the live weight, the proportion of animals exhibiting estrus, and the preovulatory follicle size of synchronized Lao-native goats were increased by the addition of the concentrate in diets based on PML.

**Keywords:** Concentrate; estrus; estrous synchronization; Lao-native goats; paper mulberry leaves (PML)

## 1. Introduction

In Lao PDR, goat production forms the main part of generating cash income because the demand for goat meat undergoes a slight increase every year. However, most Lao-native goats are produced in extensive production systems, and they normally consume diets having very low nutrient value [1,2]; as a result, the productivity and the reproductive performance obtained have been low [3]. Lately, researchers have been showing interest in focusing on tropical plants in improving the ruminant's diet [4]. Paper mulberry (*Broussonetia papyrifera*), a member of the Moraceae family [5], is among other potential plant sources in the region. The leaves contain approximately 29.1% of dry matter and 11.9% of ash, and they have been considered to be a potential feed resource for goat production [6].

Many researchers have reported that good nutritional condition has a strong influence on the activity of the hypothalamic-pituitary-gonadal axis (HPG axis) in small ruminants [7-9]. Nutrition is considered to be an important factor that affects the reproductive capacity and influences the onset of ovarian activity in female goats [10,11]. The mechanism of nutritional effects on follicular growth and development (folliculogenesis) is probably not affected by the quantity of nutrient supply; it is much more likely that there are specific nutrient signaling effects that link reproduction with favorable environmental conditions for reproduction [12]. Moreover, management of feeding with concentrate supplementation during the estrous cycle has been indicated as a tool to increase the ovarian function and the reproductive capability in small ruminants [9,13].

The increased popularity of goat meat and, hence, goat production leads to increased interest in reliable methods to manage reproduction in goats [14]. Estrous synchronization is a key element in all of the assisted reproductive technology protocols and has a major influence on enhancing the

overall capabilities of reproductive function in female goats [15]. It has been reported that estrous synchronization has been successfully used for management of reproduction in goats [16]. Feed flushing (concentrate addition) has been already applied as a feeding strategy prior to breeding; however, concentrate supplementation of paper mulberry leaf-based diets during the estrous synchronization protocol, particularly, their effect on estrous response, ovarian follicular performance, and productivity in female Lao-native goats, has not yet been determined. We hypothesized that management of nutrition with the addition of the concentrate during the estrous cycle and estrous synchronization treatment could be made to influence the estrous response, ovarian function, and the subsequent productivity in female goats. Therefore, the objective of the present study was to investigate the effects of supplementation of a paper mulberry leaf-based diet with concentrate on the growth performance, preovulatory follicle diameter, response to estrous synchronization treatment, and productivity in female Lao-native goats.

## 2. Materials and Methods

### 2.1 Location of study

The experiment was conducted at the goat production farm of Northern Agriculture and Forestry College, Luangprabang, Lao PDR. The geographical coordinates of the location are latitude 19° 58-992' N and longitude 102° 14-668' E, and its altitude is 395 m above the sea level. The climate is tropical with distinct differences between dry (October to April) and wet (May to September).

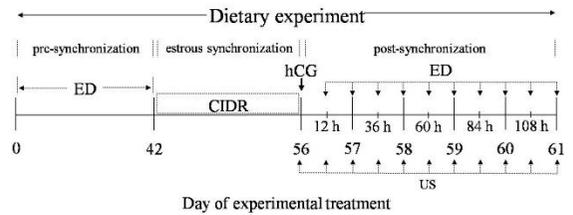
### 2.2 Animals

Before experiment, nonlactating female Lao-native goats (13 mo of age and 25.4±0.7 kg of body weight) that were observed for one complete cycle (spontaneous estrous cycle) were selected for study. All goats were determined to ensure and absence of reproductive problems and all goats remained healthy throughout the study.

Female Lao-native goats were vaccinated against foot and mouth disease (FMD) and administrated against parasites. Prior to the experiment, the estrous cycles of female Lao-native goats were synchronized by treating with controlled intravaginal drug release (CIDR) devices (Eazi-Breed CIDR, 0.3 g progesterone; Pfizer Animal Health, New Zealand) for 14 days. The estrous behavior was monitored in the presence of two bucks at 06:00 h and 18:00 h for 5 days, following the removal of CIDR devices. Commencement of estrus was defined as the time when the female goats first stood to be mounted by the bucks [17]. The first day of estrus was designated as day 0 of the nutritional period. Consequently, 24 female Lao-native goats that exhibited estrus were selected for the dietary treatments.

**2.3 Experimental design, feeding, and housing**

Nutritional treatment was conducted for a period of 61 days, with 42 days for pre-synchronization (covering of two estrous cycles), 14 days for estrous synchronization, and 5 days for post-synchronization periods (Figure 1). Twenty-four Lao-native goats were randomly assigned to either of the two dietary treatments as follows. Group 1 (PML; n=12): female goats which received paper mulberry leaves as the basal diet. Group 2 (PML+C; n=12): female goats which received paper mulberry leaves as the basal diet and 400 g/head/day of the concentrate (30% rice bran; 20% corn; 15% soy bean meal; 32% broken rice; 2% premix; 1% salt). The experimental diets were fed daily as two meals at 08:30 h and 16:00 h. The feed refusals were removed and weighed each time. The chemical compositions of the feedstuffs are demonstrated in Table 1. The female goats in the two groups were given *ad libitum* access to water. The animals were kept in individual pens made of wood, with roofs of tiles, with the pens having the following dimension: 1.0 m in width, 1.5 m in length, and 1.5 m in height.



**Figure 1.** A schematic representation of the experimental design. CIDR = controlled intravaginal drug release devices; hCG = human chorionic gonadotropin; ED = estrous detection; US = ultrasonography.

**2.4 Estrous synchronization protocol**

On day 42 of the nutritional treatments, all the female goats in the two groups received CIDR insert for estrus of 14 days, and were treated with an intramuscular injection (i.m.) of 300 international unit (IU) of human chorionic gonadotropin (hCG; Chorulon®, Intervet International B.V., New Zealand) at the removal of CIDR (day 56) [15]. After the withdrawal of CIDR devices, the overt signs of estrus were detected twice daily for a 5-day period (at 12 h, 24 h, 36 h, 48 h, 60 h, 72 h, 84 h, 96 h, 108 h, and 120 h after CIDR removal) with the aid of two teaser bucks (Figure 1).

**Table 1.** Chemical composition of feedstuff.

Item	PML	C
	----- % -----	
DM	26.5	88.8
OM	78.1	83.7
CP	19.0	15.4
CF	14.5	7.3
Ash	15.2	5.8
NFE	47.7	59.9
NDF	41.4	23.5
ADF	22.5	9.6

PML = paper mulberry leaves, C = concentrate, DM = dry matter, OM = organic matter, CP = crude protein, CF = crude fiber, NFE = nitrogen free extract, NDF = neutral detergent fiber, ADF = acid detergent fiber.

## 2.5 Fecal collection trial and chemical analysis

Twenty-four female goats were adjusted from day 0 to day 42 of pre-synchronization period. Feces of goats were collected from day 42 to day 61, which was in the periods of estrous synchronization and post-synchronization. Feces voided by each goat was weighed and recorded on the collection day. The feces samples were pooled and thoroughly mixed together. After that, 10% of the sample was taken and stored at  $-20^{\circ}\text{C}$ . During the feces collection, all feed offer and feed refusal were also collected for evaluation in the case of each individual goat. The samples of feed, refusals, and feces were calculated through proximate chemical composition for the determination of dry matter (DM), organic matter (OM), crude protein (CP), crude fiber (CF), ether extract (EE), and nitrogen free extract (NFE) by following the methods of AOAC [18]. Neutral detergent fiber (NDF) and acid detergent fiber (ADF) were determined using the procedures of Van Soest *et al.* [19]. Every animal was examined for the metabolizable energy (ME) intake of the feed that the goats had consumed. The ME was calculated from the digestion coefficient, which was derived from the *in vivo* digestion trial using the equation of Shiemann *et al.* [20] and Lee *et al.* [21], as follows:  $\text{ME (MJ/kg)} = 15.2\text{DCP} + 34.2\text{DEE} + 12.8\text{DCF} + 15.9\text{DNFE}$ . The voluntary feed intake and nutrient intake of the PML and PML+C diets are demonstrated in Table 2.

**Table 2.** Nutrient intake according to dietary treatment diets.

Item	Dietary treatment	
	PML	PML+C
DM, g/head/day	768.8	1090.6
OM, g/head/day	600.4	870.4
CP, g/head/day	146.4	195.4
CF, g/head/day	111.6	133.9
Ash, g/head/day	116.6	133.8
NFE, g/head/day	369.0	563.9
NDF, g/head/day	318.1	391.2
ADF, g/head/day	173.2	202.2
ME, MJ/kg DM	8.9	13.8

PML = paper mulberry leaves, PML+C = paper mulberry leaves + concentrate, DM = dry matter, OM = organic matter, CP = crude protein, CF = crude fiber, NFE = nitrogen free extract, NDF = neutral detergent fiber, ADF = acid detergent fiber, ME = metabolizable energy.

## 2.6 Measurement of growth performance

During the nutritional treatments, all the animals were weighed individually each week before feeding and giving water in the morning. To calculate average daily gain (ADG), female goats were weighed at day 0 and day 61. The feed offer and the feed residual were collected individually every day in the morning, and the individual feed intake was used to calculate the individual dry matter intake (DMI).

## 2.7 Estrous response

All the female goats in the two groups were monitored for estrus twice daily for a minimum of 30 min per detection with two teaser bucks within 12 h to 120 h after the removal of CIDR (Figure 1). The onset of estrus was defined as the time when the female goat first stood to be mounted by the teaser bucks. The duration of estrus was defined as the interval between the onset of estrus and the end of estrus. The end of estrus was considered to be the time when the female goats did not accept the teaser bucks. On the basis of response measurements, the estrous

rate (number of female goats exhibiting estrus/total number of synchronized goats  $\times$  100; %), the interval to the onset of estrus (h), and the duration of estrus (h) were calculated for each of the treatment groups.

### 2.8 Ovarian ultrasonography

Ovarian follicular determination was carried out according to the procedures described by Moonmanee and Yammuen-art [22] using transrectal ultrasonography (HS-1600V, Honda Electronics, Japan). Briefly, the ovaries were scanned using transrectal ultrasonography with a 7.5 MHz rectal transducer on the day of CIDR withdrawal (day 56) to evaluate the diameter of the largest preovulatory follicle. Then, the size of the largest preovulatory follicle was measured and recorded on follicular maps, which allowed the identification for subsequent analyses. The ovaries were scanned by the same operator on the day of CIDR removal and every 12 h after CIDR withdrawal (up to 120 h if incidence of ovulation had not occurred) to evaluate the ovulation time (Figure 1). Ovulation of the largest preovulatory follicle was considered to occur when only one follicle, greater than 5.0 mm in diameter and observed in the previous scanning, had disappeared [23]. On the basis of ovulatory measurements, the ovulation rate (number of female goats exhibiting ovulation/total number of synchronized goats  $\times$  100; %), the diameter of the preovulatory follicle (mm), the interval from the CIDR removal to ovulation (h), and the interval from estrus to ovulation (h) were calculated for each of the treatment groups.

### 2.9 Measurement of reproductive performance

After the end of the synchronization period, female goats exhibiting estrous behavior were taken to a separate pen where they were mated by natural service using fertile bucks. On the basis of reproductive measurements, the fecundity (number of pregnant female goats/number of female goats mating  $\times$  100; %), the infertility (number of non-pregnant female goats/number of female

goats mating  $\times$  100; %), the fertility (number of female goats kidding/number of female goats mating  $\times$  100; %), the single kidding rate (number of female goats with single kid/number of female goats kidding  $\times$  100; %), the twin kidding rate (number of female goats with twin kids/number of female goats kidding  $\times$  100; %), the productivity (number of kids born alive/total number of female goats in each treatment group; no.), the kid yield (number of kids born alive/total number of female goats mating; no.), and the prolificacy (number of kids born alive/total number of female goats kidding; no.) were also calculated for each treatment groups [24]. Additionally, the average number of kids varies according to birth type and gender was also determined for each treatment groups.

### 2.10 Statistical analyses

The data are presented as mean  $\pm$  SEM. The body weight (BW), the body weight change, the average daily gain (ADG), the feed conversion ratio (FCR), the diameter of the largest preovulatory follicle, the time of ovulation, the interval to the onset of estrus, the duration of estrus, the productivity, the kid yield as well as the prolificacy were analyzed with the ANOVA procedure of SAS (SAS Institute, Cary, NC, USA). Differences between the means were used for the student's *t*-test. The estrous rate, the ovulation rates, the fecundity, the infertility, the fertility, the kidding rate, and the proportion of kids varies according to gender were analyzed by chi-square analysis [25]. Differences with  $P \leq 0.05$  were considered significant, and those with  $0.05 < P < 0.10$  were considered a tendency [26].

## 3. Results and Discussion

### 3.1 Live weight change and growth performance

The body weight, body weight change, and growth performance of the goats during the experimental period are presented in Table 3. There was no difference in the initial body weights between the PML and the PML+C groups ( $P > 0.05$ ). Similarly, there was

no difference ( $P>0.05$ ) between the PML and the PML+C groups as regards final body weights after the end of the feeding period. However, the positive changes in the live weight of the female goats in the PML+C group were greater than those in the PML group ( $P<0.05$ ). The body weight of female goats did not differ significantly between the PML and the PML+C groups ( $P>0.05$ ) on day 0, day 7, day 21, day 28, day 35, day 49, day 56, and day 61 of the nutritional period (Figure 2). On the other hand, the body weight tended to be higher ( $P=0.08$ ) in the PML+C group than that in the PML group on day 42 of the nutritional period (Figure 2). Moreover, the ADG of the female goats receiving the PML+C treatment had a greater ( $P<0.05$ ) value compared with the goats receiving the PML treatment (Table 3). The FCR of the female goats in the PML+C group was lesser ( $P<0.05$ ) than that in the PML group (Table 3).

**Table 3.** Effects of supplementation of paper mulberry leaf-based diet with concentrate on live weight, average daily gain, and feed conversion ratio in synchronized Lao-native goats.

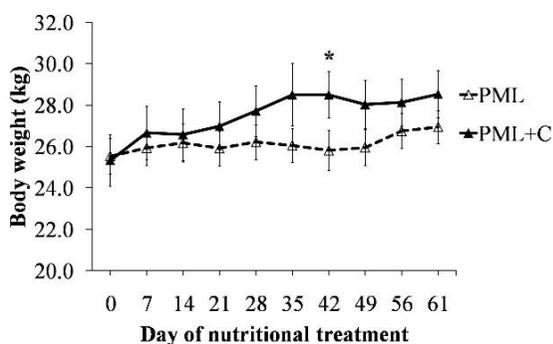
	Dietary treatment	
	PML	PML + C
Initial body weight, kg	25.5±3.1	25.3±4.3
Final body weight, kg	26.9±2.7	28.5±3.9
Body weight change, kg	1.4±0.6 <sup>a</sup>	3.2±0.9 <sup>b</sup>
ADG, g/day	24.9 <sup>a</sup>	53.2 <sup>b</sup>
FCR	34.8 <sup>a</sup>	22.2 <sup>b</sup>

ADG = average daily gain, FCR = feed conversion ratio, PML = paper mulberry leaves, PML+C = paper mulberry leaves + concentrate.

<sup>a,b</sup> Treatments denoted with different superscript letters are different at  $P<0.05$ .

Until now, little research has focused on explaining the effects of supplementation of paper mulberry leaf-based diets with concentrate on growth performance, estrous and ovulatory

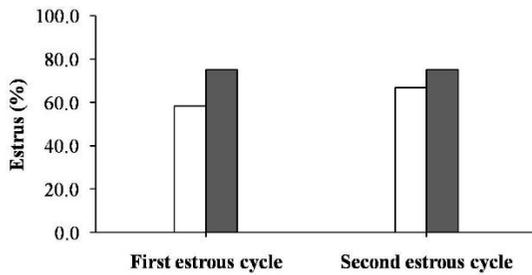
responses, and productivity of synchronized Lao-native goats. The present study provides the first description of the positive effects of addition of paper mulberry leaves (PML) with concentrate (C) on growth performance and subsequent response to estrous synchronization treatment in female Lao-native goats. The present research showed that female goats in the PML+C group (well-fed group) had significantly greater positive changes in live weight than those in the PML group. The high level of metabolizable energy (ME) intake described in this dietary treatment increased the positive changes in the body weight relative to the PML+C diet. Similarly, in Batina and Dhofari goats which received high ME diet compared to low ME diet, an increase in the daily weight was achieved to enhance the live weight [27]. The greater final live weight of the concentrate-supplemented goats may probably be due to the greater ME intake of the goats in the PML-C group. Moreover, it has been found that feeding female Mashona goats  $0.27 \text{ MJ ME kg}^{-1} \text{ W}^{0.75}$  decreased the expression of estrus and reproductive performances compared to feeding them  $0.53 \text{ MJ ME kg}^{-1} \text{ W}^{0.75}$  and  $1.06 \text{ MJ ME kg}^{-1} \text{ W}^{0.75}$  [28].



**Figure 2.** The body weight of female goats received the paper mulberry leaf-based diet (PML; dotted line) or the paper mulberry leaves and 400 g/head/day of the concentrate (PML+C; solid line). \*Treatment denoted with asterisk tends to be different at  $P=0.08$ .

### 3.2 Response to synchronization of estrus

In the pre-synchronization period (covering two estrous cycles), there were no significant differences ( $P>0.05$ ) between the PML and the PML+C groups as regards the proportions of female goats displaying estrus in the first (58.3% vs 75.0%) and the second (66.7% vs 75.0%) estrous cycles (Figure 3).



**Figure 3.** Proportion of female goats exhibiting spontaneous estrus during the pre-synchronization period (covering two estrous cycles) in the group of goats receiving the paper mulberry leaf-based diet (PML; open bars) or the paper mulberry leaves and 400 g/head/day of the concentrate (PML+C; solid bars).

In the synchronization period, the proportion of synchronized goats displaying estrous behavior was not significantly different ( $P>0.05$ ) between the PML and the PML+C groups (Table 4). Nevertheless, synchronized goats that were fed only the PML diet tended to have a delay ( $P=0.09$ ) in the interval to the onset of estrus in comparison with goats that were fed the paper mulberry leaves and 400 g/head/day of the concentrate (PML+C; Table 4). The durations of estrus of the synchronized goats did not differ ( $P>0.05$ ) between the PML and the PML+C groups (Table 4).

Interestingly, the proportion of synchronized goats displaying estrous behavior at 48 h after CIDR withdrawal was significantly higher ( $P<0.05$ ) in group that received the PML+C diet than in the group that received the PML diet (75.0% vs 33.3%; Figure 4). At 60 h and 72 h after the CIDR

removal, the proportions of female goats that exhibited estrus did not differ statistically ( $P>0.05$ ) between the PML and the PML+C groups (75.0% vs 91.7% and 25.0% vs 25.0% for 60 h and 72 h, respectively, after the CIDR removal; Figure 4).

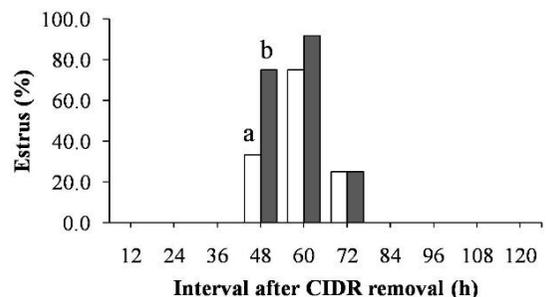
**Table 4.** Effects of supplementation of paper mulberry leaf-based diet with concentrate on response to estrous synchronization treatment, diameter of preovulatory follicle, and time of ovulation in synchronized Lao-native goats.

Item	Dietary treatment	
	PML	PML + C
Female goats, no.	12	12
Estrus, %	75.0 (9/12)	91.7 (11/12)
Interval from CIDR removal to estrus, h	54.7±2.1 <sup>x</sup>	50.2±1.6 <sup>y</sup>
Duration of estrus, h	22.7±3.7	25.1±1.2
Ovulation, %	75.0 (9/12)	91.7 (11/12)
Diameter of preovulatory follicle, mm	5.7±0.1 <sup>a</sup>	6.1±0.1 <sup>b</sup>
Interval from CIDR removal to ovulation, h	88.0±4.5 <sup>x</sup>	77.5±4.1 <sup>y</sup>
Interval from estrus to ovulation, h	32.0±2.8	30.5±3.3

PML = paper mulberry leaves; PML+C = paper mulberry leaves + concentrate; CIDR = controlled intravaginal drug release devices.

<sup>a, b</sup> Treatments denoted with different superscript letters are different at  $P<0.05$ .

<sup>x, y</sup> Treatments denoted with different superscript letters tend to be different at  $P=0.09$ .



**Figure 4.** The cumulative proportion of female goats displaying estrous behavior in 12 h to 120 h after CIDR withdrawal in the group of goats receiving the paper mulberry leaf-based diet (PML; open bars) or the paper

mulberry leaves and 400 g/head/day of the concentrate (PML+C; solid bars). CIDR = controlled intravaginal drug release devices.

Among the extrinsic factors inducing reproduction in female goats, the level of nutrient values is one of the most important factors. In small ruminants, the mechanisms controlling reproduction should comprise both the present nutritional status, particularly energy availability [29,30], and the level of body reserves, as stability for the whole reproductive process [31]. On the basis of these mechanisms, the effects of nutrition on the reproductive process are separated as follows: dynamic (nutritional balance in terms of energy and protein) and static (live weight, and body condition or body lipid stores) effects [31,32]. During the reproductive cycle, energy status is a major factor playing an important role in the reproductive process of adult female goats. For instance, in multiparous goats, resumption of estrous and ovarian activities are not correlated to the live weight of the dam at parturition but to the live weight changes (energy balance) [31]. Taken together, the present study demonstrated that although the percentage of female goats displaying estrous behavior did not differ between the PML and the PML+C diets during the pre-synchronization period (covering two estrous cycles), the percentage of female goats exhibiting estrous behavior at 48 h after the end of the hormonal treatment was significantly greater in the group that was offered the PML+C diet than in the group that was offered only the PML diet. Moreover, it was observed that goats receiving the PML+C diet had a tendency to be in estrus much earlier after the end of the synchronization protocol in comparison with goats receiving only the PML diet. These results strongly imply that at 48 h after hormonal synchronization with synthetic progesterone and hCG, fewer underfed goats go into estrus, as well as the interval to the onset of estrus tends to be longer, compared to well-fed goats. On the one hand, the interval to the onset of estrus

after the end of estrous synchronization tended to be longer in the feed-restricted goats [33]. Thus, the supplemented goats had increased expression of estrus at 48 h after CIDR removal, by which these data support the hypothesis of feed flushing during the synchronization period playing a crucial role in estrous response to hormonal treatment. Although the concentration of blood estradiol after CIDR withdrawal was not determined in this study, a possible reason for the early time in the estrus of the supplemented goats (PML+C group; high ME diet) after the end of the hormonal treatment could be the higher level of blood estradiol. These findings are supported by a previous study [34], which indicated that estradiol concentration at 48 h after CIDR removal was higher in high energy supplemented (barley grain diet) goats compared with control-treated (basal diet) goats. On the basis of previous observations, extraovarian factors such as dietary mediation in metabolic hormones (e.g., insulin, insulin-like growth factor-I, and growth hormone) also directly affect ovarian folliculogenesis (follicular growth and development) and subsequent oocyte quality [31,35,36]. On the cellular level, the enhanced production of estradiol which is synthesized by follicular cells is basically related to the development of waves of follicular growth and development in female goats [37]. During the period of ovarian folliculogenesis, growth is completed by follicular cell (theca and granulosa) proliferation [38] and an increase in the number of layers of follicular cells, as indicated by the increase in the diameter of the ovarian follicle [39]. Moreover, it has been previously suggested that the level of estradiol increases with the diameter of the ovarian follicles in the follicular phase of female goats [40]. The present study highlighted that the diameter of the largest preovulatory follicle is significantly greater in synchronized goats receiving high-nutrition diets (the PML+C diet) than in induced goats receiving low-nutrition diets (the only PML diet). The sufficient nutrition, particularly energy, of the

well-fed goats observed in this study may be the reason for the increased growth rate and the subsequent increase in the diameter of the preovulatory follicle. These findings are supported by a previous study, which reported that nutritional stimulus provided by high-energy diets stimulates the growth of dominant follicles in addition to enhancing the concentrations of glucose and insulin of female goats [9]. In fact, glucose consumed by ovarian follicular cells (theca and granulosa) can be used for major energy production of ovarian folliculogenesis to maintain follicular growth and prevent follicular atresia during the development of a growing follicle [9,12,41] in which glucose is largely produced in the liver via gluconeogenesis from propionate which is absorbed from the rumen [42].

### **3.3 Ovulation, diameter of largest preovulatory follicle, and time of ovulation**

Incidences of induced ovulation in the goats were not significantly different ( $P>0.05$ ) between the PML and the PML+C groups (Table 4). However, the female goats that received the PML+C diet had greater ( $P<0.05$ ) diameters of the largest preovulatory follicle than the female goats that received the PML diet (Table 4). Moreover, the female goats that received only the PML diet tended to have a delay ( $P=0.09$ ) in the interval from the CIDR removal to the ovulation in comparison with the goats that received the PML+C diet (Table 4). However, the intervals from estrus to ovulation of the synchronized goats did not differ ( $P>0.05$ ) between the PML and the PML+C groups (Table 4).

Taking into consideration the response to the estrous synchronization treatment, the diameter of the preovulatory follicle, and the time of ovulation, we also found that there are two phenomena of well-fed Lao-native goats: one is that the estrus response and the time of ovulation occurred earlier, and the other is that the largest preovulatory follicle exhibited a greater size after the end of the synchronization protocol. This is consistent with the results obtained in

the synchronized sheep model carried out by Moonmanee and Yammuen-art [22], who found that follicular diameter was negatively related with time to onset of estrus but positively related with blood glucose levels in ewes exhibiting estrus. Additionally, in the cattle model, it was observed that follicular size has a positive relationship with peak concentrations of estradiol, but only among synchronized cows displaying estrus [43]. The results of this study support the hypothesis that supplementing paper mulberry leaf-based diets with 400 g/head/day of concentrate can increase the response to estrous synchronization treatment in terms of the proportion of synchronized goats exhibiting estrus at 48 h after CIDR withdrawal (a short interval) and enhanced preovulatory follicle size. On the other hand, restricted feeding is related with a decrease in the proportion of female goats displaying the preovulatory surge of gonadotrophins, decreased magnitude of the surge, and subsequent low incidence of ovulation of the preovulatory follicle [44].

### **3.4 Reproductive performance**

Overall, the fecundity, the infertility, the fertility, and the kidding rate, the proportion of kids varies according to gender, the kid yield, and the prolificacy were not significantly different ( $P>0.05$ ) between the PML and the PML+C groups (Table 5). However, the average number of kids born alive per total number of female goats (productivity) tended to be greater ( $P=0.08$ ) in group that received the PML+C diet than in group that received the PML diet (Table 5). In goats that received the PML+C, incidences of births were observed, with the majority of the goats having single and some of them having twin births. However, none of the kidding goats that received only the PML exhibited twin birth (Table 5). Nutritional supplementation (high-energy diet) improved number of ovulatory follicles and ovulation rate in cycling goats [9]. Correlatively, mean number of kids born was influenced by ovulation rate in dam under better nutritional

management [45]. This is possibly correlated to the availability and sufficient supply of nutrient from well-fed management, which may have influenced ovulation rate and the subsequent productivity in female goats.

#### 4. Conclusions

The consequent increase in the response toward ME intake of the PML+C-treated goats (well-fed group) may be expected to result in increased response to synchronization treatment, and rate of preovulatory follicle growth, as indicated by the increase in the diameter of the largest preovulatory follicle. Upon taking into consideration the positive changes in the live weight, the shorter time interval as regards the withdrawal of the hormone, estrus, and ovulation as well as the subsequent productivity, it can be concluded that the PLM+C diet can be applied in feeding practices during the reproductive management of female Lao-native goats.

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**Table 5.** Effect of supplementation of paper mulberry leaf-based diet with concentrate on reproductive performances in synchronized Lao-native goats.

Variable	Dietary treatment	
	PML	PML + C
Female goats, no.	12	12
Female goats mating, no.	9	11
Pregnant female goats, no.	6	10
Female goats kidding, no.	5	9
Fecundity, %	66.7 (6/9)	90.9 (10/11)
Infertility, %	33.3 (3/9)	9.1 (1/11)
Fertility, %	55.6 (5/9)	81.8 (9/11)
Single kidding rate, %	100.0 (5/5)	88.9 (8/9)
Twin kidding rate, %	—	11.1 (1/9)
Kids born alive, no.	5	10
Single kid, no.	5	8
Female, %	80.0 (4/5)	62.5 (5/8)
Male, %	20.0 (1/5)	37.5 (3/8)
Twin kids, no.	—	2
Female, %	—	100.0 (2/2)
Male, %	—	—
Productivity, no.	0.4±0.15 <sup>x</sup>	0.8±0.17 <sup>y</sup>
Kid yield, no.	0.6±0.17	0.9±0.17
Prolificacy, no.	1.0±0.23	1.1±0.19

PML = paper mulberry leaves; PML+C = paper mulberry leaves + concentrate.

<sup>x, y</sup> Treatments denoted with different superscript letters tend to be different at P=0.08.

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