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Development of animal health and production  
improving the sustainability of livestock farming  
in integrated agriculture systems"

Bambang P. Priosoeryan  
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## Immunoglobulin Titer of Suckling Kids as a Result of Colostrum Intake

Daisy D.S.J. Tambajong<sup>11</sup>

### Abstract

*This study is concerned with the supply of newborn kids with antibodies via colostrums. Twenty kids were assigned to three groups, the first of which received Colostrum within the first hour after birth, the second and third after 4h and 8h, respectively. The first four feedings took place at 4h intervals, the amount fed being 57.5 g/kg body weight. IgG<sub>1</sub>, IgG<sub>2</sub>, IgA and IgM concentrations were assessed in the blood of the mother, in milk and in the blood of the kids. In each treatment group the serum concentration of all immunoglobulin rose soon after the first uptake of colostrums. It peaked after 24h, waning gradually until the end of the third week. The highest mean IgG<sub>1</sub> concentration was reached in the group receiving Colostrum immediately after birth (Group 1), the lowest in Group 3 (53.8, 45.3 and 36.6 mg/ml for Groups 1, 2 and 3, respectively). IgG<sub>2</sub> and IgA concentrations also reached a maximum by 24h in all groups. After that they dropped to a low at 21 days. Thereafter, IgG<sub>2</sub> titers started rising again, surpassing the initial maximum by day 56 p.p.. IgA titers remained below the assay base line. Highest IgG<sub>2</sub> and IgA concentrations were arrived in the kids of Group 2 (0.4 mg/ml for IgG<sub>2</sub> and 0.9 mg/ml for IgA, whereas the kids of Groups 1 and 2 reached maximum values of 0.3 and 0.7 mg/ml for IgG<sub>2</sub> and IgA, respectively. Maximum IgM concentrations were reached 20h p.p. (3.9 mg/ml) in Group 1 and 24h p.p. (3.7 mg/ml) in Group 2. They were maintained at that level until 40h p.p.. Kids of Group 3 reached maximum concentrations of 4.2 mg/ml at 32h p.p., subsiding soon thereafter. The concentrations in maternal blood serum of IgG<sub>1</sub>, IgG<sub>2</sub> and IgM showed a slight, statistically non-significant decrease until 12 hours after parturition. The IgA concentration remained unchanged in the range of 0.2 to 0.3 mg/ml. The concentrations of the four immunoglobulins in maternal milk showed a distinct decrease immediately after parturition. Within the first 48h they dropped by 90 to 96% and remained at a very low level.*

**Keywords:** colostrum, Serum, IgG<sub>1</sub>, IgG<sub>2</sub>, IgA, IgM concentration, Newborn kids

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Morphologically, Enami *et al.* (1985) investigated that there was a correlation between collagen gel concentration and the collagen gel strength by comparing the gel concentration of 0,1 dan 0,63%; increasing the gel concentration is also increased the gel strength. In contrast to the cultured cell on petri dish using solution medium, growth of MCM-B2 cell on 10.1% collagen gel resulted the pattern of cell colonies as duct-like structure (Priosoeryanto *et al.*, 1995b). Our study indicated that cell growth on collagen gel more suitable for observation of cell characteristics which mimicking the in vivo condition, therefore collagen gel system is an appropriate assay for study proliferation, differentiation and invasion of tumor cell.

The present study showed that tumor cell proliferation and invasion were inhibited by the *Luffa cylindrica* extracts combined with rCaIFN, this phenomenon indicated that there is a synergism effect of both substances. We suggest that these extracts plants in combination with rCaIFN could be developed and use for treatment of tumors disorders. Further study on the isolation and identification of the bioactive compound of these two plants as well as toxicity and safety on animal model should be conducted before field application.

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## Materials and Methods

### Extraction of the Plants

The extracts of *Luffa cylindrica* seed were prepared using methanol and chloroform according to the method of Anonymous, (1985). Briefly, 50 grams each of *Luffa cylindrica* seed powder were macerated using 500 ml of chloroform or methanol and kept for 5 days, and were then filtered. The wastes were dissolved into a sufficient amount of chloroform or methanol and were filtered until the total volume of extracts was 100 ml. The extracts were evaporated to get the desired concentrated filtrates and were kept until use. Working concentrations of each extracts were made by dilution the extracts until the tested concentration was achieved.

### Brine Shrimp Lethality Test

Ten larvae of *Artemia salina* on 12 vials each were used (3 concentrations of extracts and one control with 3 replicates). After 24 hours of extracts treatment, the dead *Artemia salina* was counted (Meyer *et al.* 1982). The data were processed statistically using Probit Test.

### Anti-proliferation Activity Assay

The MCM-B2 cell lines (Priosoeryanto *et al.*, 1995a) were cultured with the density of  $10^3$  cell/ml on the 24-well dish using a growth medium comprises from DMEM and 10% FCS (Priosoeryanto *et al.*, 1995a; 2000). The tested dose of each extracts was determined after the  $LC_{50}$  of each extracts were recognized. The extracts were added to the culture dish (3 holes for each dose). For the control positive, anti-tumor commercially drugs Vinblastine was used. After the confluence of cell growth was achieved on the control negative dishes, the cells were harvested and the average of the total number of cells on each dishes were counted using a hemacytometer with Trypan Blue dye. The data were then analyzed to determine the anti-proliferation activity level.

### Collagen Gel and Anti-invasion Assay

Collagen type 1 derived from porcine tendon (Cell matrix IA) was used. The gel was prepared according to the manufacturer recommendation and was then added with DME/F-12 medium, FCS and antibiotic. Collagen gel were stored in petri dishes with 0.3 cm in thick and kept in 37°C, with 5% CO<sub>2</sub> for polymerization. After the gel was polymerized, single tumor-cell solution were added to the surface of the gel. Growth and invasion activities of the tumor cells were observe daily using phase-contrast microscope.

### Data Analysis

All quantitative data were statistically analyze, while qualitative data were describe naratively according to Priosoeryanto *et al.*, (2000).

## Result and Discussion

### Brine Shrimp Lethality Test

The  $LC_{50}$  for each plant extracts were 66.8287 ppm for chloroform extract and 141,22 ppm for methanol chloroform extract of *Luffa cylindrica*. Based on the  $LC_{50}$  we decided to use the dose for anti-proliferation and anti-invasion assays of the extract was 100 ppm for the chloroform extract and 350 ppm for the methanol extract.

### Anti-proliferation Activity

The anti-proliferation activity was detected in all extracts combination with rCaIFN. In general, this anti-proliferation activity on the combination form was more higher compared to the extracts or rCaIFN alone. The degree of this activity on both extracts combination was varied. The highest anti-proliferation activity of the methanol extract-rCaIFN combination was 84%, while the chloroform extract-rCaIFN combination was 60% (Figure 1 & 2).

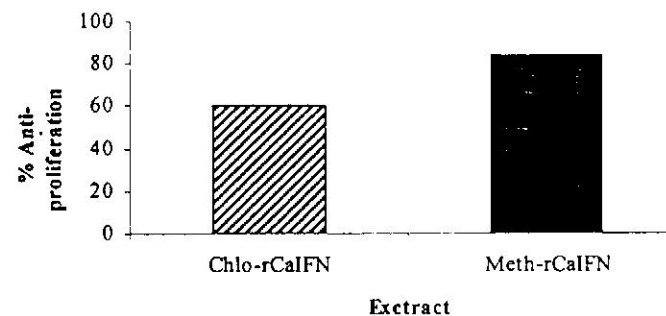


Figure 1. Anti-proliferation activity of combination between rCaIFN-Chloroform and rCaIFN-Methanol extracts of *Luffa cylindrica* on MCM B2 cell lines.

There is no report on the activity of combination of IFN with plant extracts in order to treat tumor disorder, even activity of the combination between IFN with commercially anti-tumor drugs has been reported before. Several investigator indicated that there was an increasing of anti-tumor activity on the combination between IFN with some anti-tumor substances such as Decarbazine, Vincristin, Bleomycin dan Lomustine (Pyrhoenen *et al.*, 1992), Fluorouracil (Raderer and

## In Vitro Anti-Proliferation and Anti-Invasion Activities of the Combination Between Recombinant Canine Interferon (rCaIFN) with *Luffa cylindrica* Seed Methanol and Chloroform Extracts on MCM-B2 Derived Tumor Cell Line In Collagen Gel Medium

Bambang Pontjo Priosoeryanto<sup>8</sup>, Gunanti<sup>9</sup>, Hernomoadi Huminto<sup>1</sup>  
and Ros Sumarny<sup>10</sup>

### Abstract

*In vitro* antiproliferation and anti-invasion activity of the combination between recombinant canine interferon (rCaIFN) and *Luffa cylindrica* seed methanol and chloroform extracts on MCM-B2 tumor cell lines was studied. The dose of the extract was 100 ppm for the chloroform extract and 350 ppm for the methanol extract, while the dose of rCaIFN was 10<sup>4</sup> IU/ml. The highest anti-proliferation activity of the methanol extract-rCaIFN combination was 84% while the chloroform extract-rCaIFN combination was 60%. The anti-invasion activity was detected on the semi solid medium of collagen gel system. There was an inhibition on the invasion activity of tumor cell to pass the collagen gel on both extract combinations even there was a variation on the inhibition capacity among them. The methanol extract gave the highest inhibition of the cell invasion activity. The ability of the inhibition of cell invasion activity was similar to that of anti-proliferation activity. The result of the present study indicated that the combination of *Luffa cylindrica* and rCaIFN have a synergetic effect on the inhibition of cell invasion and we concluded that this combination give a promising hope for the tumor disorders treatment. The mechanisms of this combination activity is still unclear and under investigation.

**Key words:** Anti-invasion, anti-proliferation, canine interferon, *in vitro*, tumor cells line, *Luffa cylindrica*

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### Introduction

A tumors or neoplasm can be defined as a disturbance of growth characterized by excessive, abnormal and uncontrolled proliferation of transformed or altered tissue at one or more primary points within the host, and frequently at one or more metastatic sites. In the course of spontaneous development of tumor in human and animals, groups of neoplastic cells may be present for years before a tumor. Even after the neoplastic growth becomes detectable, it may remain at relatively stable size and degree of invasiveness for prolonged periods of time before its full malignant potential is manifest.

Interferon (IFNs) has a broad range antiviral immunomodulatory and anti-proliferation effects. In human case, the anti-tumor effects have led to the clinical use of IFN in a variety of diseases (Johnson *et al*, 1994). Recombinant canine interferon (rCaIFN) was produced in a recombinant Baculovirus system by using silkworm (*Bombyx mori*) (Priosoeryanto *et al*, 2000). Anti-proliferation and anti-invasion activities of rCaIFN has been clarified in our previous study (Gunanti *et al*, 2004)

Organics substances isolated from plants are known as metabolite substances. These natural metabolites are widely use in the medical, pharmacy, agro-chemistry and chemical industries (Harborne, 1996). In some Asian countries, metabolites derived from several plants are use for the alternative treatment or traditional medicine for some disorders in human and animals. Our previous studies (Harran *et al*, 2001; Priosoeryanto *et al*, 2001; Tumilisar *et al*, 2001) showed that some plants extracts had an *in vitro* anti-tumor activity by inhibited the tumor cell proliferation.

Indonesia is a tropical country which rich of medicinal plants. Indonesia Drug and Food Control Agency indicated that medicinal herbs were produced in Indonesia by 326 manufacturers and were used not less than 180 medicinal and aromatic plants. The total of raw materials consumption annually reach about 6.223 tons. The Agency was also counted that 45 important drugs in the USA are originated from tropical medicinal and aromatic plants, in fact, 14 plants species are coming from Indonesia. The big number of medicinal and aromatic plants species grow in Indonesia is an indicator that the land and climate conditions of Indonesia very potential for the cultivation development of medicinal and aromatic plants.

The aim of the present study is to elaborate the anti-invasion activity from *Luffa cylindrica* seed methanol and chloroform extracts combined with rCaIFN on the MCM-B2 tumor cell line *in vitro*, in order to find the potential anti-tumor drugs for medical purposes.

Evaluation of Nutrition and Organoleptic Physical Quality for Pellet Using Some Levels of Seaweed ( <i>Sargassum</i> sp) ( <i>R. I. Pujaningsih, S. Sumarsih, BIM. Tampoebolon</i> ).....	71
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## Physiological Status of Indoor Sheep in the Tropical Rain Forest (HPGW) Environment

Agik Suprayogi<sup>1</sup> and D.A. Astuti<sup>1</sup>

### Abstract

*As an attempt to improve the animal health and production, agrosilvopastoral (ASP) system has been introduced in the tropical countries. ASP system usually was established as a pasture of animals in the agriculture land or forestry environment, otherwise development ASP system with indoor animal in the tropical rain forest environment are still scare. Establishing of the animal production system in the tropical forest environment has a certain consequence related to the influence of physiological status and animal health. The objective of the study is to evaluate the physiological status of indoor sheep in the Gunung Walat Education Forest (HPGW)-IPB, Sukabumi-west Java-Indonesia which has tropical rain forest climatic type. Ten Javanese thin-tailed ewes, average body weight of 25 kg, in the indoor stable system were feed and water ad libitum under 24 hours continues monitoring of stable humidity and temperature. Measurement of hearth rate, respiration rate, and body temperature were carrying out to the each ewe in the morning and afternoon. This study reveals that the average humidity in the stable a day in the HPGW-IPB is  $(97.52 \pm 4.87)$  % rel. and average temperature a day is  $(22.26 \pm 1.62)$  °C. The consequence of bioclimatic condition is directly to the physiological status of the ewes, such as hearth rate by  $(71.00 \pm 10.51)$ , respiration rate by  $(29.25 \pm 5.39)$  and body temperature by  $(38.73 \pm 0.56)$  °C. The average humidity a day in the stable shows uncomfortable for the animal health and production system, although the stable temperature is still comfortable enough. The condition is dominantly influenced by density and diversity of vegetation in the tropical rain forest. The high humidity in the stable affects enhancing of respiration rate of the ewes, although hearth rate and body temperature tend to the normal physiological value.*

**Keywords:** *Animal physiology, bioclimatic, sheep, forest*

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## Preface

In the development of integrated animal health and production for improving the sustainability of livestock farming in the integrated agriculture systems several factor has been involved. For the sustainability in the animal health sector, three main issues has been recognized, there were 1) strategic animal diseases (13 animal diseases), 2) animal and environment health and 3) improvement of reproductive performance in order to increase the livestock population. To solving the problem of animal health, the main factor that should be prioritises is an animal health technology which including development and implementation of indigenous knowledge; phytomedicine, vaccine technology, bio-security and others related factors.

In the livestock production sector, the main focus is on the sustainability of integrated livestock production. The other problem is the condition of low consumption on the animal protein; animal protein is the essential food substance that cannot be substitute by other non-animal protein. The important component for the development of livestock production are good reproduction and breeding system, development and implementation of local resource of animal feed, restructuring of livestock industry, post harvest technology, veterinary public health (food safety) including market regulation.

In the integrated agriculture, agro-sylvo-pastoral (ASP) system is to believe as one activity that could reduce poverty, increasing farmer income as well as increasing the condition of environment. The strategy that will be implement is the establishing of the ASP institution, implementation of good management and local knowledge as well as improving the productivity. The problem that still inhibit on the implementation of ASP is that there are some differences in the perception on ASP between the sector of agriculture, animal husbandry and forestry, therefore the same perception between the three sectors in the integrated using of forest for agriculture and livestock activities, including the reclamation of former mining land, plantation and fresh water fisheries should be taken as a priority.

There are 4 important points resulted from the Miniworkshop :

1. Recommendations resulted from the Miniworkshop will be deliver to the policy maker, farmers and others related institutions.
2. The points resulted from the Miniworkshop is hoping could be used as a recommendation in the Livestock and Animal Health Regulation that now is on the way of preparation and debate in the Parliament.
3. Action plan from this Miniworkshop will be taken on the kind of proposal and will be submit to the central and local government as well as international institution especially in the Germany or others countries and will be coordinated by the SEAG Coordinator as one of SEAG activity.
4. The next propose activity will be a miniworkshop on the "Development and Implementation of Indigenous Knowledge" in Manado or Kupang on the middle of 2006 or 2007.

Organizing Committee  
January, 2006

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**Foreword**

The proceeding is a produced from papers collected during the the Mini Workshop of Southeast Asia Germany Alumni Network (SEAG) on the topic of : "Development of Animal Health and Production for Improving the Sustainability of Livestock Farming in the Integrated Agriculture Systems" held in Bogor-Indonesia on April 25-26th, 2005

Nineteen selected papers were presented in this proceeding from 33 participants which coming from 13 universities in Indonesia and 1 from Thailand.

We would like to highly appreciate and deeply thanks to DAAD for the financial support as a main sponsorship in this Mini Workshop that it made the program very successfully conducted. The same thing is also going to SEAG-Indonesia who fully supported this event as one of their scientific program.

Finally, we would like to thanks to all Steering and Organizing Committee who work very hard for the symposium including the preparation and finalization of this proceeding.



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