Indones J Anim Vet Sci. 20(4): 250-256
Indonesian Journal of Animal and Veterinary Sciences
e-ISSN: 2252-696X p-ISSN: 0853-7380

Role of Sweet Orange (*Citrus sinensis*) Waste in Lowering the Meat Cholesterol and Fat of Padjadjaran Sheep

Adriani L, Hernawan E, Hidayat U

Faculty of Animal Husbandry, Universitas Padjajaran
Jl. Raya Bandung-Sumedang KM 21 Jatinangor Bandung 45363 Indonesia
E-mail: lovita_yoghurt@yahoo.co.id

(received 21-09-2015; revised 12-11-2015; accepted 26-11-2015)

ABSTRAK

Adriani L, Hernawan E, Hidayat U. 2015. Peran limbah jeruk manis (*Citrus sinensis*) dalam menurunkan kadar kolesterol dan lemak daging pada domba Padjadjaran. Indones J Anim Vet Sci. 20(4): 250-256. DOI: http://dx.doi.org/10.14334/jitv.v21i1.1250

Penelitian bertujuan untuk mengevaluasi peran limbah jeruk manis dalam menurunkan kadar kolesterol dan lemak daging pada domba Padjadjaran. Dua puluh ekor domba jantan dengan bobot badan 29,66±2,74 kg dan koefisien variasi <10% dilibatkan dalam penelitian dengan menggunakan Rancangan Acak Lengkap (RAL). Perlakuan terdiri atas 4 taraf limbah jeruk manis dalam ransum masing-masing T1 (0%), T2 (7%), T3 (12%) dan T4 (17%), perlakuan diulang 5 kali. Penelitian dilakukan selama 5 minggu, dengan peubah yang diamati meliputi konsumsi ransum diukur setiap hari, pertambahan bobot badan harian dan konsumsi air minum diberikan secara *ad libitum*, kolesterol dan trigliserida darah diukur pada akhir penelitian. Sampel daging diambil secara acak dari hasil pemotongan tiga (3) domba per ulangan. Hasil pengamatan menunjukkan penurunan kadar kolesterol daging tertinggi diperoleh pada T2, yaitu bagian paha 9,43±0,04 mg/mg dan dada 9,71±0,04 mg/mg, sementara penurunan lemak daging diperoleh pada perlakuan T4, yaitu bagian paha dan dada secara berurutan 9,70±3,98 ug/mg dan 10,48±1,85 ug/mg. pH rumen pada tidak menunjukkan perbedaan yang nyata (P>0,05) pada semua pelakuan, dengan kisaran nilai dari 6.80±0,01 - 6,50±0,02 µg/mg. Kesimpulan, limbah jeruk manis (*Citrus sinensis*) mampu berperan menurunkan kadar kolesterol dan lemak dan mempertahankan ekologi rumen dalam hal tingkat pH rumen Domba Padjadjaran.

Kata Kunci: Limbah Jeruk Manis, Kolesterol Daging, Lemak Daging, Domba Padjadjaran

ABSTRACT

Adriani L, Hernawan E, Hidayat U. 2015. Role of sweet orange (*Citrus sinensis*) waste in lowering the meat cholesterol and fat of Padjadjaran sheep. Indones J Anim Vet Sci. 20(4): 250-256. DOI: http://dx.doi.org/10.14334/jitv.v21i1.1250

This research is aimed to evaluate sweet orange's role in lowering meat cholesterol and fat in Padjadjaran sheep. Twenty sheeps with body weigth 29.66±2.74 kg and variance coefficient <10% were used in this research using Completely Randomize Design (CRD). The treatments were consists of four level sweet orange addition in ration. The treatments were T1 (0%), T2 (7%), T3 (12%) and T4 (17%) that were repeated 5 times. The research was conducted for five weeks with observed variables are ration consumption that was measured everyday, body gain that was measured every week, cholesterol and triglycerides measured at the end of the research. During the research water consumption was given ad libitum. Meat samples were selected randomly from three for each repeated treatments. Observation result showed the highest decrease on meat cholesterol level was T2 treatment with 9.43±0.04 mg/mg on thigh and 9.71±0.04 mg/mg on breast. Meanwhile for fat, the highest decrease was from T4 treatment with 9.70±3.98 ug/mg dan 10.48±1.85 ug/mg. Rumen's pH did not show significant difference (P>0.05) on all treatments with value range between 6.80±0.01 - 6.5±0.02. In conclusion, sweet orange (*Citrus sinensis*) waste is capable to lowering cholesterol level and fat in meat of Padjadjaran sheep.

Key Words: Sweet Orange Waste, Meat Cholesterol, Meat Fat, Padjadjaran Sheep

INTRODUCTION

Animal product usually contains high lipid; therefore nowadays people are more selective to choose their diet. This condition surely affects people preference on animal product. Now, is the right time for an innovation to produce high quality meats that have low content of cholesterol and fat. Herbal materials use as a mean to decreasing cholesterol and fat on blood and meat, have been published so many times, one of the herbal material is sweet orange (*Citrus sinensis*) waste. It has so much potential to decrease cholesterol and fat level on animal meat (Hernawan E & Adriani L 2014).

One option of fitofarmaka is sweet orange (*Citrus sinensis*) waste , because Indonesia produces 2,355,550 tons *Citrus sinensis* each year, (Ministry of Agriculture 2010), while only about 35-40 % which can be used by consumers, for food and beverage processing industry, whereas the remaining 65% is wasted that can not be

utilized. This is a good opportunity in livestock feed because it has a superior active compound (Mirzae & Naser 2008).

Efforts to reduce cholesterol and triglycerides using fitofarmaka for example adding bay leaf flour (*Syzygium polyanthum* Wight) have done by group of Animal Husbandry student, Universitas Padjadjaran. The result showed that blood cholesterol levels decreased from 297.75±51.73 to 139±12.03 mg/dl, with use 4% bay leaf in ration of Broiler. Other Research using *Citrus sinensis* waste in quail, showed a good result and adding up to 6% can reduce the cholesterol content of the meat from 60.11 to 29.82 mg/dL, (Fadilah 2011) and the optimum dose for decreasing fat and cholesterol in poultry not more than 6 % in ration.

The content of the active compound in citrus waste including volatile oil 0.91%, tannins 0.95%, flavonoids 0.46%, and 0.84% saponin (LKO Unpad 2013).

The active compound in sweet oranges such as tannins, saponins, flavonoids and essential oil is able to inhibit the absorption of cholesterol in the intestine. The presence of tannins will stick or lining the intestine membrane thus inhibiting the absorption of cholesterol (Oluremi et al. 2007).

Essential oils are believed have the function of lowering the activity of Glycerol-3-Phosphate Dehydrogenase (GPHD), an enzyme that plays role in triglycerides synthesis. Essensial oil could inhibit triglyceride synthesis in the liver and small intestine may result to decrease the triglycerides (He et al. 2009). The dominant component compound of *Citrus sinensis* is hesperidin and narirutin classified as flavonoids that act as antioxidant (Peterson et al. 2006)

Hesperidin worked through the mechanism of inhibition the activities of HMG-CoA reductase, so the enzyme activity involved in the biosynthesis of cholesterol is inhibited.

Padjadjaran sheep, is a local breed that has related with Garut sheep from Wanaraja area still in purification for meat products, has white feathers and width ear characteristics (Bandiati et al. 2012). They have high potential as meat source and are highly adaptive.

Attempt to decrease cholesterol and fat levels on Padjadjaran sheep's blood by using waste citrus sinensis was feared to harm the sheep's biological function especially rumen's ecology and the value of hematology would improved.There is a trend to modify the animal cholesterol and fat content in order to produce high quality products. *Citrus sinensis* waste has been reported to reduce blood cholesterol and triglyceride level, therefore this study was conducted with the objectives to study the effect of dietary inclusion of *Citrus sinensis* waste on meat cholesterol and fat of Padjadjaran sheep.

MATERIALS AND METHODS

Animal Experimental

The sheep were procured from Animal Breeding Station, Animal Husbandry Faculty Universitas Padjadjaran. The research trial was conducted at Faculty of Animal Husbandry, Universitas Padjadjaran, Indonesia from April 2015 to August 2015. Twenty sheep were used for the study. The sheep were reared in cages having age group between 24-30 months with average body weight 29.66±2.74 kg and coefficient of variation <10 %. The experimental period lasted after five (5) weeks. The concentrate ration was used andgrass was used asfed. The consentrate are rice bran, cassava flour, tofu waste, coconut cake, sweet orange (*Citrus sinensis*) waste, molasses, pollard and cassava peal.The sheep were randomly allocated to four treatment groups as T1, T2, T3 and T4, in a Completely Randomized Design (CRD). The sheep in the first group (T1) were givenrasion without the addition of *Citrus sinensis* while in other groups *Citrus sinensis* was supplemented at the rate of 7, 12, and 17% in T2, T3, T4 respectively.

Proceesing of *Citrus sinensis*

The *Citrus sinensis* was obtained from the sweet orange processing units located around Bandung, was sun dried (<50ºC) until the moisture content reached 10% and followed with milled with hammer mill at Faculty of Animal Husbandry, Universitas Padjadjaran.

Samples Collection

Meat samples were collected at the end of experimental period, randomly selected from three sheeps from each treatment group, so the total is 12 healthy individuals (60% of total sheep) was slaughtered with a cutting technique streak (Butterfield 1988).

Halal slaughter method is done after fasting for 22 hours to reduce the slaughter weight variation. Meat samples analyzed were taken from the right side of carcass longisimus dorsi – (LD) muscle and the biceps femoris (BF) muscle, were taken at the thigh. The meat was taken randomly as much as 10 grams each, at multiple locations on longisimus dorsi and biceps femoris.

Purbowati & Suryanto (2000) research showed that LD muscle meat fat content (3.10%) higher than the BF (2.16%) because the thigh muscles more used for moving. There is no significantly different (P>0.05) because the sheep were kept in cages so the motion

Table 1. Ration composition, nutrient content and energy

Feed Ingredients	Nutrient Content(100% Dry matter)			
	R0	R1	R2	R3
	---------- % ----------			
Rice Bran	27.92	27.59	27.82	28.09
Cassava flour	5.36	3.22	5.4	5.43
Tofu Waste	0.99	1.22	1.34	1.24
Cococnut Meal	13.00	10.14	8.1	6.59
LJM (*Citrus sinensis)*	0.00	7.00	12.00	17.00
Molasses	3.10	3.11	3.13	3.14
Pollard	21.65	22.76	23.25	23.65
Cassava peal	27.98	25.32	19.53	15.58
Total	100.00	100.00	100.00	100.00
Nutrient Composition				
Dry Matter	85.15	85.419	85.848	86.305
Ash	7.56	7.721	7.937	8.134
Crude Protein	12.00	12.00	12.00	12.00
Crude Fat	5.004	4.929	4.903	4.903
Crude Fiber	10.955	11.16	11.205	11.312
Nitrogen Free Extract	51.753	51.692	52.309	52.435
Total Digestible Nutrient	73.00	73.00	73.00	73.00

Source: Caltucated aplication from Winfeed

activities in the thigh muscle is limited, as a result fat tends to stockpiled (Lambuth et al. 1970).

Meat cholestrol was estimated using Cholesterol kit (Biolab), using CHOD-PAP (Cholesterol Oxidase Phenylperoxidase Amino Phenozonphenol) method (Richmond 1973 in Kasturi & Singhania 2014). Meat fat was estimated using Soxhlet method (AOAC 2006).

Estimation of rumen pH (ruminal fluid from slaughtered sheep) was collected and pH was measured using pH-meter.

Statistical analysis

Data collected were subjected to analysis of variance (ANOVA) (Gaspersz 2006) was used to test the significance of difference between means considered significant at $P<0.05$.

RESULT AND DISCUSSION

It can be seen from Table 2, that the feed intake of all treatments are relatively equal, concentrate between 375.19±27.89 to 390.32±8.72 and grass consumption 2438.86±154.74 to 2538.21±210.65. Average daily gain ranged from 8.73-64.28 g. The feed intake and body weigh for all treatments was not significantly different ($P>0.05$).

Forage dry matter intake can be seen in Table 2. Consumption of dry matter forage and concentrates are relatively the same in all treatment, 400-440 g/head/day and exhausted. Average consumption of feed dry matter is 3.3% of body weight that is in the normal range.

The highest daily weight gain achieved by treatment T3 with body weight gain 64.28 g for 5 weeks (Table 2), with adaptations for 1 week. This illustrates that the provision of *Citrus sinensis* up to 12% shows the highest body weight gain compared to other treatments, is closely related to the active compounds including flavonoids and essential oils to improve metebolisme, and its impact on body weight gain (Hernawan & Adriani 2014).

Triglycerides were decreased in all treatment, its serves an energy reserve used (Malinow et al. 1987; Oluremi et al. 2007), so the blood levels to be reduced. The presence of flavonoids also believed to play a role in suppressing the concentration of triglycerides. Flavonoids are thought to activate cAMP synthesis resulting in increased protein kinase resulting in increased triglyceride hydrolysis so the triglycerides in the blood and liver will reduce (Olivera et al. 2007;

Table 2. Feed consumption and body weight

Parameter	T1	T2	T3	T4
Concentrate consumption (g)	375.19±27.89[a]	389.95±18.82[a]	390.32±8.72[a]	388.04±15.72[a]
Grass consumption (g)	2438.86±154.74[a]	2524.64±234.94[a]	2538.21±210.65[a]	2560.62±236.15[a]
Body Weight (Kg)	28.34±3.46[a]	30.72±2.96[a]	30.37±3.03[a]	33.35±1.31[a]

Table 3. Blood Cholestrol and Triglyceride Contain

Parameter	R0	R1	R2	R3
Triglyceride (mg/dl)	30.0±11.0	30.4±7.6	19.0±2.0	20.5±7.5
Cholesterol (mg/dl)	126.61±10.96	129.65±16.44	106.52±12.17	120.98±15.98

Rusell 2009). Protein kinase activates triglyceride lipase through forforilasi here in after described triglycerides into free fatty acids and glycerol by triglyceride lipase (Rusell 2009).

Cholesterol comes from food into the digestive tract and in the small intestine to be absorbed by enterocytes of the small intestine mucosa, the next will be esterification into cholesterol esters. After that, lipoprotein cholesterol esters will formed chylomicrons, then get into the flow of lymph and end up in the bloodstream (Shepherd 2001; Linder 2006).

The active compounds contained in sweet oranges such as tannins, saponins, flavonoids and essential oil is able to inhibit the absorption of cholesterol in the intestine (Malinow et al. 1987; Francis et al. 2002; Oluremi et al. 2007). The presence of tannin can stick or coat the intestinal membrane thereby inhibiting the absorption of cholesterol (Oluremi et al. 2007). In addition, the saponin in the gastrointestinal tract to form a bond with cholesterol complexes that are difficult absorbed by the intestine so that most cholesterol will come out with feces (Malinow et al. 1987). The presence of flavonoids are also believed to inhibit the absorption of cholesterol by inhibiting the formation of micelles ration so that the absorption of cholesterol settles and can be pressed (Olivera et al. 2007; Gropper et al. 2009).

Table 4 is the major research,the results revealed that dietary inclusion sweet orange waste in all levels decreasing the meat cholesterol and fat content of sheep meat, when compared to the control group (T1). Highest reduction of thigh and breast meat cholesterol level 9.43±0.04 and 9.71±0.04 µg/mg respectively, was significantly different ($P<0.05$) recorded in all treatments, fed 7% citrus waste (T2) in the diet when compared to control groups. Further, the highest decrease in fat content of thigh and breast were 9.70±3.98 and 10.48±1.85 µg/mg respectively, was observed in the group fed 17% citrus waste (T4) in the diet compared to other treatments including control.

Flavonoids also produce multi-enzyme systems such as cytochrome P-450; affecting on lipid metabolism and bile acids. The enzyme cytochrome P-450 has the ability to mediate the formation of bile acids so the amount of bile acids was decreased.

Bile acid decreasing is the main route to eliminate the cholesterol. The presence of flavonoids is also believed to play a role in suppressing the concentration of triglycerides (Table 2). Flavonoids which can activated cAMP synthesis, will increase the kinase protein and also increased the triglyceride hydrolysis, which decreased the blood triglycerides (Olivera et al. 2007).

The decreased lipid profile related with Galleano et al. (2012) study, that flavonoids may have beneficial role in regulating the fatty oxidation and improve adipocyte function, also flavonoids act as antioxidants, thus decreased the cholesterol levels in the blood (Oluremi et al. 2007), by release one hydrogen atom from the group and reducing the formation of free radicals, resulting in the synthesis of 3-hydroxy-3-metilglutaril-CoA (HMG CoA) which serves as a precursor in obstruction of cholesterol formation (Reynertson 2007). Naringenin flavonoid in citrus has been reported prevented the accumulation of adipose, adipocyte hypertrophy and dyslipidemia (Mulvihill et al. 2010).

The cholesterol from food, was entered into the digestive tract and small intestine then absorbed by enterocytes of the small intestine mucosa and will esterified into cholesterol esters. After that, with phospholipids and apolipoprotein, lipoprotein cholesterol esters will forming chylomicrons then get into the flow of lymph and the ending in the bloodstream (Linder 2006).

In addition, the saponin in the gastrointestinal tract forming a complex bond with difficult cholesterol absorbed blood cholesterol, but has no significant difference to blood cholesterol and triglycerides, even there are tendency to decrease from 69.6 mg/dL to 64.00 mg/dL (Hernawan E & Adriani L 2014) by the intestine so most of cholesterol was release with feces). The presence of flavonoids are also believed to inhibit the absorption of cholesterol by inhibiting the formation of micelles

Table 4. Effect of Citrus sinensis on meat cholesterol, meat fat and rumen pH

Parameter	T1 (0%)	T2 (7%)	T3 (12%)	T4 (17%)
Meat Cholesterol				
Thigh (µg/mg)	11.71±0.03^{b}	9.43±0.04^{a}	10.86±0.15^{ab}	10.43±0.20^{ab}
Breast (µg/mg)	11.14±0.04^{b}	9.71±0.04^{a}	10.43±0.17^{a}	10.43±0.21^{a}
Meat Fat				
Thigh (µg/mg)	25.68±1.48^{b}	13.85±1.38^{a}	14.51±1.18^{a}	9.70±3.98^{a}
Breast (µg/mg)	24.13±0.41^{c}	16.76±1.46^{b}	16.32±0.98^{b}	10.48±1.85^{a}
pH				
Rumen	6.80±0.01	6.82±0.03	6.78±0.04	6.50±0.02

Means within the row with different superscripts are significantly different (P<0.05)

ration so the absorption of cholesterol tend to settle and can be decreased (Olivera et al. 2007; Gropper et al. 2009).

Flavonoids and essential oil is thought to have a role to inhibit the synthesis of cholesterol in the liver. Hesperidin which is the dominant compound in the citrus flavonoid work through the mechanism to inhibite the activity of the HMG-CoA reductaseenzyme involved in cholesterol biosynthesis. Essential oils are also believed have a role inhibiting the reductase enzyme HMG-CoA.The essential oils in the skin of the fruit of Citrus plants genus are limonene, citronellal, geraniol, linalol, α-pinene, mirsen, β-pinene, sabinen, geranil acetate, nonanal, geranial, β-kariofilen, and α-terpineol (Chutia et al. 2009).

In this study, the saponin levels in each treatment using *Citrus sinensis* waste are 0.059, 0.109, and 0.143% respectively; tannin levels 0.067, 0.114, and 0.162% respectively, and the levels of flavonoids 0.032, 0.055, and 0.078% respectively (calculated in 7, 12, and 17%). The levels of these compounds is still tolerate, such as tannin only used not more than 2-3%, and saponins can be used up to 0.2% (Oluremi et al. 2007).

Another study, about the addition of sweet orange waste more than 20% in the diet, has increased the level of saponin, so the ration becomes less palatable, even the rations can not be consumed (Callaway et al. 2010). Tannins will coated the intestine membrane and inhibits the absorption of nutrients include the cholesterol

The essential oils have also been reported to decrease the blood triglyceride levels by decreasing the activity of Glycerol-3- Phosphate dehydrogenase (GPDH) enzyme, which is involved in the biosynthesis of triglycerides, according He et al. 2009. Fat tissue is made up of fat cells in a matrix of connective tissue. Triglycerides content in fat cells make up around 85% of the fat tissue, and triglyceride is made up of three fatty acids; can be any of many combinations of saturated, mono-unsaturated and polyunsaturated acids.

Most of ruminant, lipids consumed will hydrolysed in the intestine with helped bile salts and pancreatic lipase to fatty acids and glycerol. Lipids are already partially digested mainly in the form water-soluble, forming micelle is stable, especially content long chain fatty acids, monoglycerides and bile acids are diffused into the surface of mucosal cells and release the material to be absorbed (Linder 2006).

Additionally, apart from the effect on meat cholesterol and fat, the effect of citrus waste on rumen pH was also studied. The results revealed that the values ranged from 6.80±0.01 to 6.50±0.02 among various treatment groups including control group (Table 4). There was no statistical difference in rumen pH among all treatments, indicating that adding of citrus waste up to 17% level had no detrimental effect on rumen ecology.

Results of this study are very important because it can decrease the cholesterol meat level until 19.47% in thigh meat compared to the control using 7%, and also decreased the meat fat 62.22% when using 17% *Citrus sinensis* waste.

CONCLUSION

It could be concluded that the dietary added with *Citrus sinensis* waste in sheep had beneficial effects with regard to its ability in reducing the meat cholesterol and fat levels which could be attributed to various active ingredients present in it. Further, the inclusion of *Citrus sinensis* waste up to 17% level in the diet of sheep had no detrimental effect on rumen ecology in terms of rumen pH levels. However, further studies in this regard with different inclusion levels are warranted.

ACKNOWLEDGEMENT

The authors would like to express the most appreciation to a group of students who worked on *Citrus sinensis* and supported by KemenristekDirjen Dikti, PUPT Project Universitas Padjadjaran, 2015.

REFERENCES

[AOAC] Association of Official Analytical Chemists. 2006. Crude fat determination-soxhlet method. Meat Technology Information Sheet. Association of Official Analytical Chemists.

Bandiati KPS, Rahmat D, Heriyadi D, Damayanti T, Kuswaryan S, Andrian D. 2012. Aplikasi biomolekuler (Polimorphis Gen PIT1) dan bioteknologi reproduksi (SE dan IB) dalam upaya standarisasi sifat produksi yang berorientasi pasar domba pedaging Padjadjaran-1. Fakultas Peternakan, Universitas Padjadjaran.

Butterfield RM. 1988. New concepts of sheep growth. The Departement of Veterinary Anatomy. Sydney (Australia): University of Sydney.

Callaway T, Edrington T, Ricke S, Crandall P. 2010. Orange Pulp and Peel as Feedstuffs to Reduce E. coli O157:H7 and Salmonella Populations in Ruminants. USDA ARS, and University of Arkansas. USA.

Chutia M, Bhuyan DP, Pathak MG, Sarma TC, Boruah P. 2009. Antifungal activity and chemical composition of Citrus reticulata blanco essential oil against phytopathogens from North East India. Food Sci Technol. 42:777-780.

Fadilah YR, Hernawan E. 2011. Pengaruh Pemberian Tepung daun Salam (Syzygium polyanthum, Wight) dalam ransum terhadap kadar kolestrol dan trigliserida darah puyuh. Bandung (Indones): Fakultas Peternakan, Universitas Padjadjaran.

Francis G, Kerem Z, Harindes P, Makkar, Becker. 2002. The biological action of saponins in animal system: a review. J Nutr. 88:587-605

Galleano M, Calabro V, Prince PD. 2012. Flavonoids and metabolic syndrome. Ann New York Acad Sci. 1259:87-94.

Gaspersz V. 2006. Tehnik analisis dalam penelitian percobaan 1. Cetakan ke 3. Bandung (Indones): Penerbit Tarsito.

Gropper SS, Smith JL, Groff JL. 2009. Advanced nutrition and human metabolism. 5th edition. Wadsworth (USA): Cengage Learning. p. 131-175

He ML, Yang WZ, You JS, Chaves AV, Mir PS, Benchaar C, McAllister TA. 2009. Effect of garlic oil on fatty acid accumulation and glycerol-3-phosphate dehydrogenase activity in defferentiating adipocytes. Asian-Aust J Anim Sci. 22:1686-1689.

Hernawan E, Adriani L. 2014. The impact of sweet orange waste in rations on blood profile and weight gain of Padjadjaran rams. University of Agricultural Sciences and Veterinary Medicine Iasi.

[LKO Unpad] Laboratorium Kimia Organik Universitas Padjajaran. 2013. Kandungan senyawa aktif dalam limbah jeruk manis. Bandung (Indones): Fakultas MIPA, Universitas Padjadjaran.

Lambuth TR, Kemp JD, Glimp HA. 1970. Effect of the rate of gain and slaughter weight on lamb carcass composition. J Anim Sci. 30:27-35.

Linder MC. 2006. Biokimia nutrisi dan metabolisme. Jakarta (Indones): UI Press.

Malinow MR, Concocn W, Mclaugblin P, Trafford, Lin D, Livingstone A, Kohler G, McMulty W. 1987. Cholesterol and bile acid balance in macasa fasicularis effect of Alfalfa saponin. J Clinic Invest.

Tripoli E, La Guardia M, Giammanco S, Di Majo D, Giammanco M. 2007. Citrus flavonoids Molekular structure, biological activity and nutritional properties: A review. Elsevier J Food Chemist. 104:466-479.

Ministry of Agriculture. 2010. Prospects and Directions Citrus Agribusiness Development. (Accessed on August 10th 2015). http://www.litbang.pertanian.go.id/special/komoditas/b3jeruk.

Mirzae AA, Naser MS. 2008. Nutritive value of some agro-industrial by-products for ruminants. World J Zool. 3:40-46.

Mulvihill EE, Assini JM, Sutherland BG. 2010. Naringenin decreases progression of atherosclerosis by improving dyslipidemia in high-fat-fed low-density lipoprotein receptornull mice. Arteriosclerosis, Thrombosis, and Vascular Biology. 30:742-748.

Oluremi OIA, Ojighen VO, Ejembi EH. 2006. The nutritive potentials of sweet orange (Citrus sinensis) rind in broiler production. Int J Poult Sci 5:613-617.

Oluremi OIA, Ngi J, Andrew IA. 2007. Phytonutriens in citrus fruit peel meal and nutritional implication for livestock production. Nigeria: University of Agriculture Makurdi Benue State.

Olivera T, Ricardo KFS, Almeida MR, Costa MR, Nagem TJ. 2007. Hypolipidemic effect of flavonoids and cholestyramine in rats. Latin Am J Pharm. 26:407-410.

Oloche J, Ayoade JA, Oluremi OIA. 2015. Haematological and serum biochemical characteristics of West African dwarf goats fed complete diets containing graded levels of sweet orange peel meal. Nigeria: University of Agriculture Makurdi Benue State.

Peterson JJ, Dwyer JT, Beecher GR, Bhagwat SA, Gebhardt SE, Haytowitz DB, Holden JM. 2006. Flavanones in oranges, tangerines (mandarin), tangors and tangelos: a compilation and review of the data from analytica literature. J Food Comp Anal. 19:66-73.

Prajoga SBK, Permana I, 2009. Genetic evaluation of priangan sheep using multivariate maternal genetic effect and their variation of sheep mitochondrial-DNA. Biotechnol Anim Husb. 25:917-924.

Purbowati E, Suryanto E. 2000. Komposisi kimia otot Longissimus dorsi dan Biceps femoris domba yang diberi pakan dasar jerami 6 padi dan aras konsentrat yang berbeda. JPPT. 25:66-72.

Reynertson KA. 2007. Phytochemical analysis bioactive constituens from edible myrtaceae fruit (Thesis). [New York (USA)]: City University of New York.

Richmond W. 1973. In: Kasturi SR, Singhania PR. 2014. Clin Chemist. 19:1350-1356. Preparation and properties of a cholesterol oxidase from *Nocardia* sp. and its application to the enzymatic assay of total cholesterol in serum. Clin Chemist. 19:1350-1356.

Rusell DW. 2009. Fifty years of advances in bile acid synthesis and metabolism. J Lipid Res. 120-125.

Shepherd J. 2001. The role of the exogenous pathway in hypercholesterolaemia. Eur Heart J. 3:2-5

Steel RGD, Torrie JH. 2006. Principles and procedures of statistics, a biometrical approach book company.