Veterinary Medicine Journal

Vol. 26 No. 1 January 2010

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Veterinary Medicine Journal published by the Association of Indonesia Veterinarians (PDHI)
and the Faculty of Veterinary Medicine, Airlangga University, Surabaya,
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Panhysterectomized Effect on Calcium and Phosphorus Retention in Sprague Dawley Rats Consuming Unsalted Teri Fish for 4 Weeks

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Abstract

The objectives of the research were to study the effect of panhysterectomized on calcium (Ca) and phosphorus (P) retention (Ca and P consumption, urinary and fecal Ca and P excretion) in Sprague Dawley rats that were fed unsalted teri fish which ratio of Ca:P is 0.5:0.7 for 4 weeks. Ten female of Sprague Dawley rats, 6 weeks old randomly divided into two groups (control and panhysterectomized group) of five each. At 8 weeks of age, the rats of panhysterectomized group were panhysterectomized. At 16 weeks of age, they were placed into individual metabolic cages for balance study. Every morning, from day 4 to 8 of the balance study, the remaining of feed was collected for Ca and P analyses. Urine and fecal samples were also collected at the same time. The research results showed that Ca retention, Ca consumption and urinary Ca excretion in panhysterectomized group were not significantly, while fecal Ca excretion was a significantly reduced (p < 0.01) in panhysterectomized group compared with the control group. Phosphorus consumption, P retention, fecal and urinary P excretion were not significantly (p<0.05) in panhysterectomized group compared with the control group. Based on the results could be concluded that panhysterectomized in rats were not affect the Ca and P retention.

Key words: Panhysterectomized, teri fish, Ca and P retention

Introduction

Besides a role in intestinal Ca absorption (Xu et al., 2003), the hormone estrogen also increases Ca reabsorption by renal tubules (McKane et al., 1995). The fall in the hormone estrogen may be the cause of decline in Ca and P absorption in the intestine (Colin et al., 1999; Xu et al., 2003); increased urinary Ca excretion (Dick et al., 2004) and lower urinary excretion of P (Dick and Prince, 2001; Dick et al., 2004). Decline in intestinal Ca absorption and renal Ca reabsorption efficiency causes patches of negative Ca (Heaney et al., 1978), and loss of bone mass in both post-menopausal individuals (Holzher et al., 2000; Hastrup et al., 2000) and post ovariectomy rats (Watanabe et al., 2001; O’Loughlin and Morris, 2003). Teri bargaining may be used as Alternative food for their daily protein and minerals. Teri bargaining besides containing high protein and minerals with minerals, especially the ratio of Ca: P (1:1), also contain vitamin D. Until now, a study on the utilization of freshwater bacteria to meet the needs of the individual minerals of menopause is still a bit done. Research Sprague Dawley rats fed with fresh anchovy feed ratio Ca: P = 3:1 (0.9%: 0.3% or 90:30 mg/100 g feed) for 12 weeks post-panhysterectomy lower Ca and P retention due to increased Ca excretion in feces, and increased P excretion in feces and urine (Hartiningingsih et al., 2004). While research on male Wistar rats were given feed containing Ca and P 0.6%: 0.6% or a ratio of Ca: P = 1:1 for 4 weeks to excrete Ca and P in feces and urine are lower than rats that consumed the feed containing P with a higher ratio (Ca: P = 1:2 and Ca: P = 1:2.5) (Tani et al., 2002). The utilization of feed were containing anchovy Ca and P 0.5%: 0.7% of Ca and P excretion in feces and urine of individuals after panhysterectomy not been done. This study aimed to assess the effects of retention of panhysterectomy Ca and P (the consumption of Ca and P, Ca and P excretion in feces and urine) in rats that consumed freshwater fish.
containing Ca and P 0.5% and 0.7%, respectively for 4 weeks. So expected to obtain information about benefits and food security minister in the prevention of bone demineralization.

Materials and Methods

Ten Sprague Dawley female rats 4 weeks old, 60-70 grams weight are included in individual cages with room temperature about 27-28°C. Each rat was given food containing 20% protein (20 grams of feed grain/100, Ca 0.5% (50 mg/100 g feed) and P 0.7% (70 mg/100 g of feed) and drinking water are aquabidestilata ad libitum. Composition of feed (% of grain/100 g) given during the study consisted of 78% cornstarch, 17% fresh water fish flour, 2.3% molasses, 1.3% CaCO3, 0.5% NaH2PO4, and 0.9% of vitamin mineral. At the age of 8 weeks, rats randomly divided into 2 groups (control and panhysterectomy or treatment) of each 5 rat. Operation panhysterectomy (taking the uterus and ovaries) done in accordance with the method described Wantor et al. (1992) is by making incision on the linea alba from the umbilicus to the kaudal, and as it anesthetic used ketamin and xylazine mixture injected by intramuscular. The same is done in the control rats, although not done taking the uterus and ovaries.

Compensation study to determine the retention at Ca and P, the consumption of Ca and P, Ca and P excretion in feces and urine tests performed at 12 weeks old rat (4 weeks post-panhysterectomy). During the exchange studies, each rat was placed in individual metabolic cages, given food and drink 15 grams aquabidestilata 120 ml each day. The study began after the rats patches adapted for 4 days. On days 4-8 of immunity studies, conducted every morning collecting the remaining food, feces and urine. Urine is collected, after the volume is measured and added 37% HCl solution that had a pH of 1, stored in a temperature of -5°C. Feces and food remains collected, after drying by drying and weighted also be stored in a temperature -5°C. For the Ca and P in food and feces, 3 gram sample of 6 grams of feces and feed samples dusty at a temperature 60°C according to method of Harris (1970). Examination of urine Ca and P after 5 ml urine sample was prepared by evaporation at a temperature of 60°C, dissolving with HCl 37% and the dilution method according to Harris (1970). Calcium, feed, feces and urine checked with the method cresolphtheline-a-complexone (Barker and Chauman Ray, 1967). The examinations of P in food, feces and urine have performed by absorbency atomic-spectrometry (AAS). Data results Ca and P were analyzed by t test.

Results and Discussion

Consumption of Ca of panhysterectomy rats did not differ with the control rats (Table 1), as well as the amount of food consumed by these two groups of rats (feed consumption data are not included). While research on Wistar rats aged 8 weeks to be done Liang et al. (2002) showed an increase in feed consumption as much as 12% within 3 weeks post-panhysterectomy feed consumption dropped 10-20% after the rats were given estradiol for 2 weeks. Consumption of Ca of panhysterectomy rats did not differ from control rats not likely associated with different levels of estrogen are two groups of rat. Hartningsih et al. (2006) reported that panhysterectomy Wistar rats fed fresh fish feed for 1.5 months showed no difference in consumption levels of estrogen and Ca compared with control rats. Estrogen levels of panhysterectomy rats do not decrease the possibility of having to do with body fat factor. This is based on weight gain of rats panhysterectomy is higher than the control rats. Within one month after panhysterectomy, the average weight increase in rats panhysterectomy is 8.5 grams or 2.5 times than the average weight gain of control rats ± 3.2 grams. Wade et al. (1985) and Meli et al. (2004) reported the occurrence of weight gain and fat tissue in animal experiments conducted in ovariectomy and premenopausal women associated with the loss of ovarian function because of either age or surgery. Korchuk et al. (2003) also reported a weight gain in post-cast gonadectomy due to increased body fat. According to Labrie et al. (1997) most of the estrogen produced in the gonadal tissue. The same thing was reported Goji (1993) that more than 95% of women premenopausal estrogen produced in the ovary. During menopause, the ovaries stop producing estrogen, and all circulating estrogen produced extragonadal networks such as fat tissue, osteoblast, and bone dendrois, vascular endothelium and some parts of the brain.

Ca excretion in feces of rats and lower panhysterectomy is significantly different (p<0.01) with control rats (Table 1). More low Ca excretion in the feces also occurred in Wistar rats panhysterectomy who consume freshwater fish for 1.5 months but did not differ significantly with control rats (Hartningsih et al., 2006). Ca excretion in feces showed a lower increase in Ca absorption in the intestine. According to Schulz-Ahrens et al. (2007) value of mineral absorption (Ca and P) is the difference of the amount of mineral (Ca and P) is consumed with the amount of mineral (Ca and P) is excreted in the feces. Some researchers reported that the intestinal absorption of Ca is actively or transcellular among others controlled
by the 1,25-dihydroxyvitamin D3 (Wasserman and Fullmer, 1995) and estrogen (Chen and Kalu, 1998). According to Chen and Kalu (1998) direct role of estrogen in the absorption transport intestinal Ca is active through estrogen receptor found on intestinal mucosal cells. In this study, estrogen was not different between Sprague Dawley rats compared with control panhysterectomy illustrates that the higher absorption of Ca Sprague Dawley rats is probably due panhysterectomy higher 1,25-dihydroxy vitamin of D3. Reportedly Tani et al. (2007) that consumption of feed containing Ca: P = 0.6%: 0.9% significantly increased 1,25-dihydroxy vitamin of D3 than Wistar rats that consumed the feed containing Ca: P = 0.6%: 0.6% but did not cause differences in Ca excretion in feces. Zhang et al. (2007) also reported that low Ca consumption significantly increased 1,25-dihydroxy vitamin of D3 of rats and ovarectomy rat superficial surgery (not ovariotomy). Calvo et al. (1988) reported that the young men who consumed the feed containing a low Ca or low ratio of Ca:P led to higher levels of 1,25-dihydroxyvitamin D3 than young women. More high of 1,25-dihydroxy vitamin D3 of young men (an analogue of the rats panhysterectomy) causes higher intestinal absorption of Ca was suggested low Ca excretion panhysterectomy rat in feces, while a lower 1,25-dihydroxyvitamin D3 of young women (analogous to the control rats) causes lower intestinal Ca absorption. This is characterized higher Ca excretion in feces of control rats. According to Van Croomen et al. (2004) and Van de Graaf et al. (2003), 1,25-dihydroxyvitamin D3 causes increased intestinal absorption of Ca. Several researchers also reported that the 1,25-dihydroxyvitamin D3 increases intestinal absorption of Ca suggested increase transport carriers of Ca trans cellula like epithelial Ca channel (ECaC1) at the apex membrane of intestinal epithelial cells (Hoenderop et al., 1999); duodenal mRNA calbindin D9K (Song et al., 2003; Bronner, 2003; Stepchenko and Bronner, 2001) and plasma membrane Ca2+-ATPases (PMCA1b) (Kip and Strehler, 2004). However, in this study was not carried out checks on 1,25-dihydroxy vitamin D3.

Ca excretion in the urine of rats panhysterectomy also lower although not significantly different with control rats (Table 1). Tani et al. (2007) also reported a decrease in urinary Ca excretion and increased 1,25-dihydroxyvitamin D3 male Wistar rats given feed containing Ca: P = 0.6%: 0.9%. According to Hoenderop et al. (2001) and Hoenderop et al. (2002), 1,25-dihydroxy vitamin D3 increases Ca reabsorption by the kidney thereby reducing urinary excretion of Ca.

Ca retention panhysterectomy rats tended to be higher than the control rats (Table 1). According to Scholz-Ahrens et al. (2007) retention of minerals (Ca and P) is the difference of the amount of minerals consumed by the amount of minerals (Ca and P) is excreted in feces and urine. Meanwhile, according Tomonari et al. (1997) the difference of the amount of minerals consumed by the amount of mineral excreted in feces and urine patches is defined as mineral. In this study, retention of Ca panhysterectomy rats tended to be higher than the control rats seemed related to the efficiency of Ca absorption in the intestine and kidney of rats panhysterectomy tended to be higher. In Table 1, shows that the consumption of Ca panhysterectomy rats and control rats are not different, panhysterectomy rats showed levels of Ca excretion in feces and urine which tend to be lower than the control rats. Retention of higher Ca can be advocates for the improvement of bone. Ca, Gueguen and Pointillart (2000) reported, that there is a link between the intestinal absorption of Ca, Ca excretion in feces and urine with Ca deposition in bone. Also reported that Ca is mobilized from the bone when a decline in Ca absorption in the intestine. O'Loughlin and Morris (1994) also reported a link between compensation or retention of Ca by the accumulation of mineral in bone. Wood (2000) reported that the retention or compensation to reflect the occurrence of Ca balance between the process of bone formation and resorption during bone remodeling process. Ca retention showed higher positive bone formation than bone resorption, and vice versa.

Consumption of P, P excretion in feces and urine, and P retention panhysterectomy rats did not differ with the control rats (Table 2). Consumption of P diet rats that are panhysterectomy not different from control rats were associated with a different amount of feed consumed by the two groups of these rats (feed consumption data are not included). While the P excretion in feces did not differ from control rats, probably due to P content in feed is higher than P Ca so that absorption in the intestine may occur by passive diffusion. P excretion in feces is not different from control rats thus showed a rat intestinal absorption of P panhysterectomy rats no different from the control. Reportedly Cross et al. (1994) that the majority (60-80%) in the intestinal absorption of P
Table 1. Average Consumption, Ca Excretion in Feces and Urine, and Ca Retention Sprague-Dawley Rats Who Are the Ancepcy Panhysterectomy Bargaining with the Ratio Ca:P = 0.5:0.7 (50 mg Ca: 70 mg P/100 g feed)

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<tr>
<td>Ca consumption</td>
<td>230.09±29.71</td>
<td>234.52±44.40</td>
<td>ns</td>
</tr>
<tr>
<td>Ca retention</td>
<td>200.13±27.13</td>
<td>188.51±41.95</td>
<td>ns</td>
</tr>
<tr>
<td>Ca fecal</td>
<td>29.85±5.66</td>
<td>44.08±6.02</td>
<td>**</td>
</tr>
<tr>
<td>Ca urine</td>
<td>0.50±0.10</td>
<td>1.12±0.33</td>
<td>ns</td>
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Description: ns = non significant (p>0.05), ** highly significantly different (p<0.01); BW, body weight

Table 2. Average Consumption, P Excretion in Feces and Urine, and P Retention Sprague-Dawley Rats Who Are the Ancepcy Panhysterectomy Bargaining with the Ratio Ca:P = 0.5:0.7 (50 mg Ca: 70 mg P/100 g feed)

<table>
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<th>Parameter</th>
<th>Panhysterectomy</th>
<th>Normal</th>
<th>Significances</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ca consumption</td>
<td>313.16±40.69</td>
<td>318.33±61.94</td>
<td>ns</td>
</tr>
<tr>
<td>Ca retention</td>
<td>206.63±66.62</td>
<td>204.67±54.64</td>
<td>ns</td>
</tr>
<tr>
<td>Ca fecal</td>
<td>24.84±2.29</td>
<td>31.89±3.60</td>
<td>ns</td>
</tr>
<tr>
<td>Ca urine</td>
<td>76.70±9.26</td>
<td>81.77±28.62</td>
<td>ns</td>
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Description: ns, non significant; BW, body weight

occurs through panhysterectomy rats passive diffusion, and according to Lee et al. (1986) only a small portion P absorbed by active transport. Pico et al. (1978) reported that P absorption in the intestine through the active transport with sodium-phosphate (NaPi-IIb) as a co-transport media are regulated by the hormone estrogen. Reported Xu et al. (2003) that estrogen plays a role in intestinal absorption of P through the stimulation of co-transporter-IIb pump marked by increase in mRNA and protein expression of NaPi-IIb female rat. Based finding suggests that the possibility: panhysterectomy treatment did not cause the decline of estrogen thus has no effect on P absorption in the gut that are not marked differently in local P-excretion by control rats. 

P excretion in the urine of rats did not differ panhysterectomy control rats (Table 2). No different the urinary excretion of P in panhysterectomy rats, with control rats did not appear related to different consumption and absorption of P in the intestine into two groups of animals were not different marked P excretion in feces. Tani et al. (2007) reported that P absorption in the intestine, excretion of P in feces and urine depends on the amount of P in feed consumed. 

The higher P is consumed, the higher the P absorption in the intestine, and the higher P is excreted in feces and urine, and vice versa. Higdon (2005) also reported that the number of P in feed consumed will easily absorbed by the intestine, whereas excess P in the blood due to the high P absorption in the intestine is excreted through the kidneys. Research on male Wistar rats by Tani et al. (2007) showed that P excretion in the urine increased when the rats were given feed containing higher phosphorus. Moreover, Takeda et al. (2000), Takeda et al. (2004) and Miyamoto et al. (2004) also reported that the kidney through ketransporter NaPi-IIa is responsible for the reabsorption of phosphate. In addition to dietary P, Napi-IIa expression is also controlled by estrogen (Farquhar et al., 2008, Dick and Prince, 2003, Dick et al., 2005, Farquhar et al., 2008, Dick and Prince, 2001, and Dick et al., 2005) reported that the regulation of estrogen decrease prisoners in transport kidney, increasing P excretion in the urine. Based on the above description, then no differences in urinary P excretion between rats panhysterectomy with control rats showed no association with the possibility of different estrogen levels are two groups of rats. Does
not differ in it estrogen rate causes not to differ in it regulation co transporter NaPi kidney thereby also causes not to differ in it excretion P in urine. In this study indicate that pannysterectomy no effect on urinary excretion of P in Sprague Dawley rats that consumed the feed in the ratio of fresh anchovy Ca: P = 0.5 : 0.7 for 4 weeks.

Retention of P of pannysterectomy rat’s value is not significantly different with P retention of the control rats (Table 2). Consumption of P and P excretion in the urine of rats pannysterectomy not significantly different with the control rats a factor P retention was not different (Table 2). These results are consistent with the results of research conducted (Miyama 2006) that the rats have pannysterectomy P retention did not differ significantly with P retention of the control rats.

Compensation study results showed that rats who consumed anchovy pannysterectomy fresh for 4 weeks still have the value of Ca and P retention and the ability of positive Ca and P absorption in the intestines is good. The same thing was reported (Hartingsh et al. 2001) that rats Sprague Dawley adult acute nephrosis patients who consume freshwater fish for 1.5 months has continued retention of Ca and P is positive, and supports bone mineralization. Similarly, Widyono et al. (1999) reported that healthy adult rats and osteopaty patient rats who consume fresh anchovy has a value of Ca and P retention positively marked by the retention and absorption of Ca and P are high.

Conclusion

From the results can be concluded that research on pannysterectomy rats that consumed freshwater fish in the ratio Ca: P = 0.5 : 0.7 or 80 : 70 mg/100 g of feed for 4 weeks did not result in changes in the retention of Ca and P.

Acknowledgments

This research is partly the result of research financed from public funds budget of Gadjah Mada University. Thanks are submitted to the Institute for Research and Corporate Citizenship Gadjah Mada University who provided research funds according to the agreement 2323R/P.12/SET.R./2004 number of research implementation.

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