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Expression Analysis of Galectin-1 from Fat in Berkshire Pigs

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ABSTRACT : Galectins are a group of animal lectins consisting of galectin-type carbohydrate recognition domains (CRD) with relatively minor domains. The biological properties of galectins include the regulation of inflammation, intercellular adhesion, cell differentiation and cell death. The diverse kinds of galectin suggest variety in their biological roles. Galectin-1 is released during adipocyte differentiation and is associated with fat which is one of the important factors for meat quality. To verify expression level, a 0.5 kb clone of galectin-1 was obtained from cDNA prepared from back fat tissue of a Sancheong Berkshire pig with good quality meat, and the galectin-1 gene identified. The deduced amino acid sequence of the galectin-1 gene was compared with those obtained from other species. By using RT-PCR and Real time-PCR, an attempt was made to determine the expression level of galectin-1 and to compare with various tissues (tenderloin and back fat) taken from pigs in different groups. Grouping of pigs was based on growth-stage (weighing 60, 80, and 110 kg) and the sub-speciation (Yorkshire and Sancheong Berkshire pigs). We attempted to determine influences of pig species, growth stages and tissue variations on the expression level of the galectin-1 gene and it was revealed that the expression pattern of the galectin-1 gene was significantly different ($p<0.01$ or $p<0.05$). Galectin-1 genes were expressed more highly in the back fat tissues of pigs weighing 110 kg than in those weighing 60 kg or 80 kg. However, the lowest expression was seen in the tenderloin tissues of pigs weighing 110 kg. Sancheong Berkshire pigs showed higher expression of the galectin-1 gene compared to Yorkshire pigs. Accordingly, it is considered that the expression pattern of the galectin-1 gene influences the growth of back fat tissues and the pig speciation relationship. Previous studies suggested that different expression of galectin-1 genes represents variety among the breeds and is closely related to fat tissue growth, conjugation and catabolism. Further, this study suggests that the expression of galectin-1 at a specific growth stage and tissue contributes significantly to the overall meat quality of Sancheong Berkshire pigs. (**Key Words** : Galectin-1, Back Fat, Sancheong Berkshire, RT-PCR, Meat Quality)

INTRODUCTION

Galectins are a family of proteins which have been widely distributed and conserved in all kinds of animals, plants and even fungi. They are defined by the structural similarity among specific polyactosamine-enriched glycoconjugates in CRDs and amino acids. Mammalian galectins are well-known in a variety of tissues from several species. Currently, it is known that there are fourteen galectins in mammals. Based on their domain structures, galectins are classified into three subgroups: proto-type galectins (galectin-1, -2, -5, -7, -10, -11, -13 and -14), chimera-type galectin (galectin-3) and tandem-repeat type galectins (galectin-4, -6, -8, -9 and -12) (Melissa et al., 2005). In general, galectin groups composed of at least a kind of galectin play a role in cell expression in any animals. This suggests that individual galectins are expressed in cell- and tissue-specific manners. Some galectins contribute broadly to the expressions of different cells and their high levels are found in various organs of animals. Their various functions suggest that they function as multi-functional proteins associated partially with the physiological functions of immune reactions. As a result of investigating the biological functions of galectins on the basis of such the reaction regulation, it was reported that the galectin proteins in mice act as an auto-regulatory growth inhibitor (Walls and Mallucci, 1991), and that galectin-3 plays an important role in mast cells, IgE-mediated neutrophils and eosinophils and allergic inflammation and activates the defensive mechanism of host antagonizing infections by bacteria and parasites (Liu, 1993; Truong et al., 1993). In addition, galectin-1 is expressed in epithelial cells, endothelial cells, fibroblasts, smooth muscle cells and nerves distributed primarily in the lung, liver, lymphocytes and lymph nodes. Other galectins are remarkably expressed at the level of

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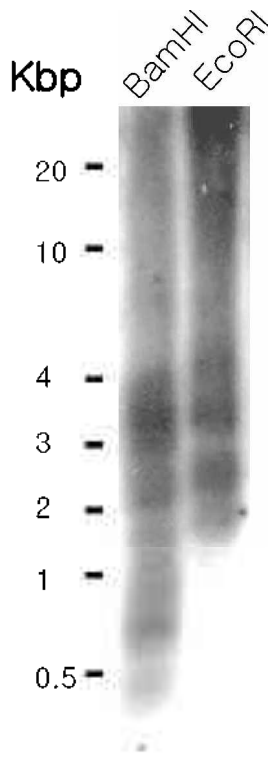

Figure 1. Genomic southern blot analysis. Ten micrograms of DNA were digested *BamH*I and *Eco*RI fractionated on a 0.8% agarose gel; and transferred onto a nylon membrane. The membranes were probed with the full-length of each gene, P^{32}-labeled back fat cDNA from swine. Size marker is shown on the left in kilobase pairs. Confirming existences and single copy number of the swine genes, obtained using cDNA.

individual cells and are discovered in only a few organs. This finding suggests that the distribution galectins varies with tissues and each galectin has its unique biological functions for specific cells during development or any other stages.

In 1986, Gitt and Barondes succeeded in producing human hepatoma cDNA library with antiserum which was characterized by specific binding with the β-galactoside-binding soluble lectins from human body, and isolating galectin-1 from human cDNAs by means of immunoscreening for the first time in the world (Gitt, Barondes, 1986).

It was found that *in vitro* the porcine galectin-1 functions as a modulator of progesterone production in the granulose cells derived from the porcine ovary (Walzel et al., 2004) as well as functions as an interaction between chondrocytes and a lactose-modified chitosan (Marcon et al., 2005). Galectin-4 was found as an isoform in the porcine small intestine (Melissa et al., 2005). Other than the above-mentioned studies, there are little or no studies on porcine galectins. Moreover, none of previous studies have addressed the effect of galectin-1 gene on the meat quality in pigs. It was revealed that although galectin-1 has various functions, it acts as secreted protein during adipocyte differentiation (Wang et al., 2004). Based on the information obtained from adipocyte secreted factors, we intended to verify the correlation between galectin-1 expression and fat tissues regarded as one of important determinants for porcine meat quality. To characterize the porcine galectin-1, we used Sancheong Berkshire pigs because they are famous for their good meat quality (Tadayosi, 2003). Galectin-1 gene was isolated from cDNA of the 30 kg-weight back fat tissue derived from Sancheong Berkshire pig. The gene was identical with the porcine galectin-1 gene and similar to those obtained from other organisms. Galectin-1 was expressed in each tissue of Sancheong Berkshire pig, and cDNA chip, RT-PCR, real time-PCR southern and western blotting was employed. Additionally, for the purpose of comparison between the porcine breeds, the same tissues with those extracted from Sancheong Berkshire were extracted from Yorkshire and used in this study.

The primary aim of this study is to clarify the biological role of affecting the porcine meat quality that the expression pattern of galectin-1 may be associated with adipose tissue by growth and formation.

MATERIALS AND METHODS

cDNA library construction and microarray analysis

Construction of cDNA library and analysis of cDNA microarray were carried out as described by the previous report (Kim et al., 2005).

Sampling of pig tissues

Sancheong berkshire (from Sungchuk Farm, the lineage formed with pigs from Kagoshima) and Yorkshire (GaYa Stockbreeding Ltd.) female pigs whose body weight reached 60, 80, and 110 kg were each butchered three times; Two farms have same feeding condition and similar management environments in the same area. Number of experimental was eighteen pigs, and their tissues were immediately taken, soaked into liquid nitrogen, and kept in a freezer at -80°C until RNA isolation.

Total RNA preparation

Total RNA was extracted with TRIzol reagent according to the manufacturer's protocol (Invitrogen, Carlsbad, CA). Trizol reagent 2 ml was added to 0.1-0.2 g of tissue grinded, mixed well by homogenizer, and a 1 ml aliquot of the mixture was transferred into 1.5 ml E-tube. Then, the mixture was left alone at room temperature for 10 minutes and centrifuged at 12,000 rpm for 10 minutes, and the

solution excluding the cell debris was saved into a new tube. A 200 μl chloroform was added to the supernatant, then 500 μl isopropanol was added to precipitate the RNA. The RNA sediment was washed with 70% ethanol, and the concentration was measured with absorbance, the confirmed by electrophoresis in 1.5% formamide gel.

Cloning of the galectin-1

Galectin-1 gene was cloned into pBluescript vector by using cDNA library prepared with cDNA Library Kit (Invitrogen, Carlsbad, CA). Transformation of *Escherichia coli* XL1-blue cells was done by the protocol of Mendel and Higa (1970). Plasmid Mini-prep Kit (QIAGEN Korea Ltd.) was used for isolation of the recombinant plasmid, which is followed by digestion with *Xho* I and *Eco*RI to see the expected insert on a 1% agarose gel by electrophoresis.

Analysis of galectin-1 expression by immunoblotting

After separation the lysates on 15% SDS-PAGE gels, the proteins were transferred to PVDF membranes. The membranes were saturated with 5% non-fat dry milk in TBS/Tween overnight at 4°C and, after washing with TBS/Tween, probed with a galectin-1 pAb (1:3,000 dilution in TBS/Tween with 5% BSA) for 3 h at room temperature. Then the blots were washed three times with TBS/Tween and incubated with a goat anti-rabbit secondary antibody conjugated to HRP (1:3,000 dilution in TBS/Tween). The washed blots were treated with ECL reagents according to the manufacturer's instructions, and the bands were visualized luminographically on X-ray films (Kodak).

Reverse transcription (RT) PCR

For RT-PCR, first strand cDNA was synthesized by using Superscript II Reverse Transcriptase, according to the manufacture's protocol (Invitrogen, Carlsbad, CA). Briefly, a 5 μg of extracted RNA was added to a reaction mixture consisting of 4 μl of 5× First strand Buffer (Invitrogen, Carlsbad, CA), 1 μl of 10 mM dNTPs dissolved DEPC-water, 2 μl 0.1 M DTT, 1 μl (200 U/μl) of SuperScript Reverse Transcriptase II (Invitrogen, Carlsbad, CA), 1 μl (0.5 μg/μl) of oligo-d(T) 12-18 primer, 1 μl of RNase Inhibitor (Invitrogen, Carlsbad, CA), and RNase-free water. Then, the RT step was carried out at 42°C for 1 h, followed by heating up at 70°C for 15 min and adding 1 μl RNase H at 37°C for 20 min before storage at 4°C.

The gene amplification for galectin-1 was achieved by using PCR with specific primers. Primers were 5'-AAAGA CAGCA ACAAC CTGTG CC-3' (forward primer) and 5'-AGGCA AATGC CTTTT AATTG GG-3' (reverse primer). In brief, 1 μl of reverse transcription mixture (cDNA) was added to the PCR mixture consisting of 1.5 μl 10× PCR-buffer, 1 μl 15 mM $MgCl_2$, 1.2 μl 2.5 mM dNTPs, 1 μl of each primer (10 pmol), 0.2 μl Taq-DNA-Polymerase (Promega, Madison, Wisconsin, USA), and 9.2 μl DEPC-H_2O. PCR was conducted in a condition; the first segment at 95°C for 3 min, 30 cycles (94°C for 30 sec, 55°C for 30 sec, 72°C for 1 min), and the final segment at 72°C for 10 min. A set of negative control was also included except the reverse transcription reaction. For internal control for assessing relative amount of target gene from different tissue samples, we used 18S rRNA gene for the comparison (Acc No. AF102857; Forward 5'-CTCGA TGCTC TTAGC TGAGT-3', Reverse 5'-CTAGT TAGCA TGCCG AGAGT -3'). RT-PCR products (approximately 649 bp) were separated on a 1% TAE agarose gel and visualized by UV after ethidium bromide staining.

Real time-PCR

Total RNA and cDNA was extracted as described above. After quantification of total RNA by spectrophotometer, cDNA dilutions of 100, 10, 1 ng, and 100 pg were prepared to create a quantitative reference standard with SYBR Green PCR Master Mix (Applied Biosystems). To obtain the maximum specificity in amplification, the primers were designed using the software Primer Express 3.0 (Applied Biosystems). The primers for real-time PCR were purchased from GenoTech Corp. Primer sequences for GAPDH (AF017079) was reported in pig. Primer sequences for Galectin-1 were 5'- CCTGC ACTTC AACCC TCGCT -3' (forward primer) and 5'- CCTCC ACGAC ACTTC CAGGC-3' (reverse primer). Real-time PCR was performed on cDNA generated by the RT reaction using the sequence detector 7500 ABI PRISM and SYBR Green I chemistry (Applied Biosystems). Forty cycles were performed at the following temperatures: 95°C for 10 min, then 95°C for 30 s and 60°C for 1 min. TaqMan GAPDH Control Reagents (Applied Biosystems) were used to evaluate the transcription of the glyceraldehyde-3-phosphatedehydrogenase (GAPDH: Acc No. AF017079) housekeeping gene as an endogenous reference. Primer concentrations were optimized for the RT-PCR to determine the minimum required to give the lowest threshold cycle and the maximal signal, while minimizing non-specific amplification. RT-PCR was run on ABI PRISM 7500. The Gene Amp 7500 SDS Software directly recorded the fluorescent light emission in real time. Based on the construction of a standard curve (validation test for GAPDH), semi-quantitative assessment of Galectin-1 was obtained.

Statistical analysis

The data are presented as the mean±SEM of three independent experiments with each choice of tissues (tenderloin and back fat) taken from total eighteen animals

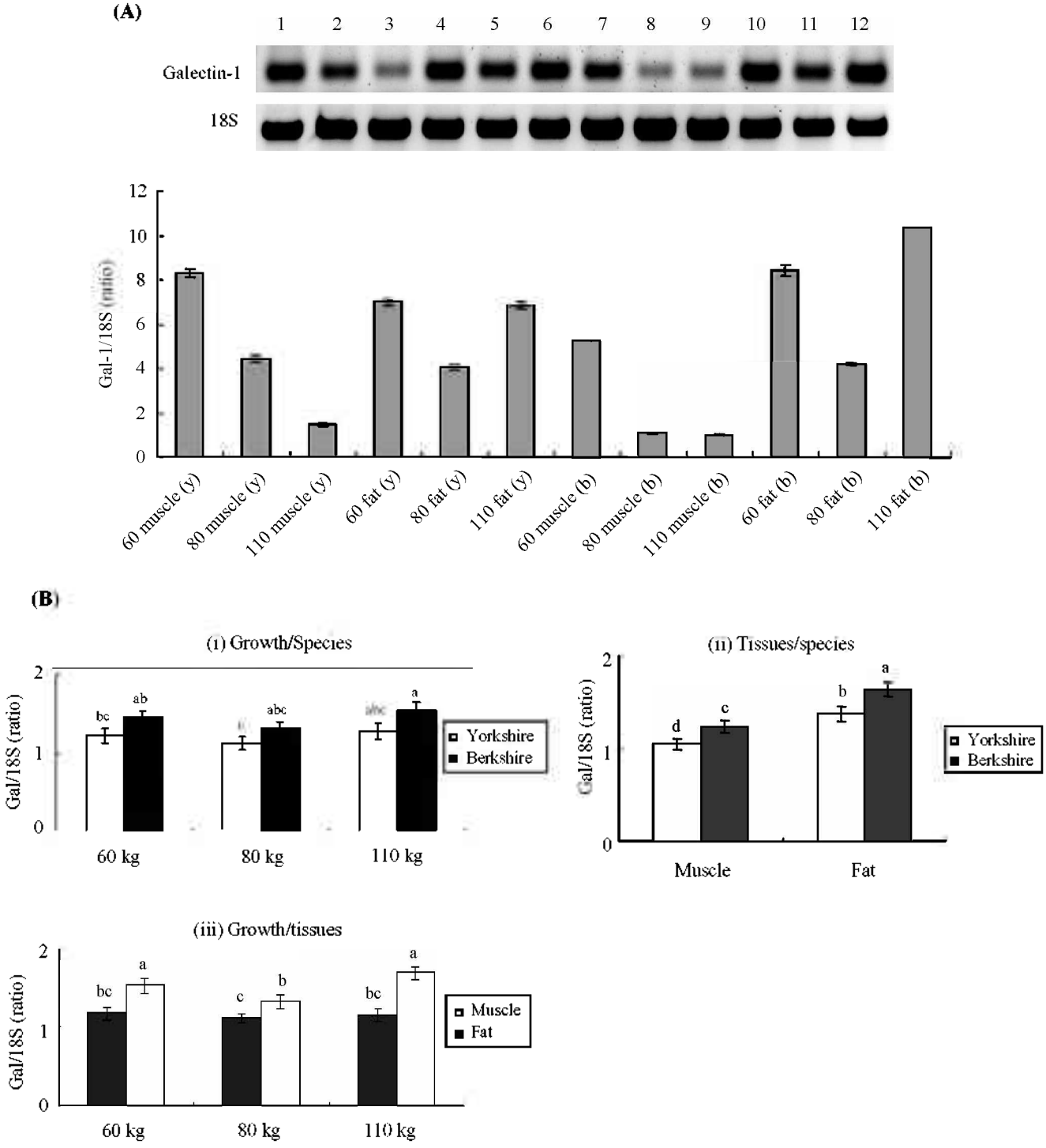

Figure 2. (A) Assessment of the expression level of the galectin-1 mRNA by RT-PCR with the muscle and fat tissues taken from Sancheong berkshire and Yorkshire pig in a different body weight. Lanes 1: 60 kg, muscle; 2: 80 kg, muscle; 3: 110 kg, muscle; 4: 60 kg, fat; 5: 80 kg, fat; 6: 110 kg, fat (lane1-6: Yorkshire). Lanes 7: 60 kg, muscle; 8: 80 kg, muscle; 9: 110 kg, muscle; 10: 60 kg, fat; 11: 80 kg, fat; 12: 110 kg, fat (lane7-12: Sancheong berkshire). (B) The expression levels of galectin-1 mRNA by interaction. (i) growth stage and species, (ii) species and tissues and (iii) growth stage and tissues. Data means±SE. The lowercase letters above the same bars indicate statistical significance; mean values with different are significantly different (i; $p<0.05$, ii and iii; $p<0.01$). $^{a, b, c, ab, bc, abc}$ Means within a row with different superscripts differ at the p-values indicated.

(9 of Sancheong Berkshire and 9 of Yorkshire) grouped into 3 different growth stage (60, 80, 110 kg of body weight). To identify the significance for the difference in the expression levels of Galectin-1 assessed by RT-PCR, ANOVA analysis of variance was performed by means of SAS program, and the significance was verified with Duncan's multiple range test.

RESULTS

A 542-bp gene was isolated from the cDNA clones of the back fat in Sancheong Berkshire pig, by means of full-sequencing. It was found that the gene had the same sequence of previously reported porcine galectin-1 gene (Accession no. AY604429) and the 408-bp sequence encoded 135 amino acids. The gene includes CRD, and has the 65-bp sequence at 5'-truncated region and the 60-bp sequence at 3'-untranslated region.

The amino acid sequence of galectin-1 extracted from Sancheong Berkshire pig was same the known porcine galectin-1 protein (AAT37622.1) and showed high sequence homology to the proteins of bovine galectin-1 (CAA32508.1), human galectin-1 (NP_002296.1) and rodent galectin-1 (NP_032521.1) - 89%, 87% and 83%, respectively. Also the amino acid sequence of galectin-1 CRDs showed high homology in all the species.

To examine the copy number of galectin-1 in the porcine genome, Southern blot analysis was carried out with porcine genomic DNA digested with *Bam*H I and *Eco*R I. The hybridization was performed with the CRD of galectin-1 cDNA as a probe (Figure 3). Two bands were observed in the *Eco*R I digest, because the probe contains *Eco*R I digest site. The result suggests that galectin-1 existed in the single copy in pig genome. Although not stated in this study, the results of cDNA chip indicated its expression in the muscles and adipose tissue-specifically, higher expression in the latter. Similarly, as a result of RT-PCR with the tenderloin and back fat of Sancheong Berkshire pig, it was more significantly expressed in the back fat.

To clarify the expression level of galectin-1 with RT-PCR, the tenderloin and back fat tissues extracted from Sancheong Berkshire pigs by each growth stages (weighing 60 kg, 80 kg, and 110 kg) were used (Figure 2A). As a result of performing this analysis technique, the highest expression was found in the back fat tissue of the Sancheong Berkshire pig weighing 110 kg, whereas the lowest expression was observed in the tenderloin tissue of the Berkshire pig weighing 110 kg, rather than the pigs weighing 60 kg or 80 kg. A shown in Figure 2A, its expression in the tenderloin tissue decreased with growth, while its expression in the back fat tissue decreased during in the pig weighing from 60 kg to 80 kg but increased in the pig weighing 110 kg.

So as to account for the correlation between the expression pattern of galectin-1 and the sub-speciation of pig, we analyzed the expression level of galectin-1 extracted from Yorkshire pig in the same manner. To draw a comparison between Sancheong Berkshire and Yorkshire pigs, we extracted and used the tenderloin and back fat tissue of Yorkshire pig, same as Berkshire pig. As a result, the highest expression was observed in the tenderloin at the pig weighing 60 kg, while the lowest expression was seen in the tenderloin at the pig weighing 110 kg. The expression pattern was similar to that found in Sancheong Berkshire pigs however the highest expression was identified in the tenderloin at stage 60 kg, unlike Sancheong Berkshire pig. However, according to the findings in the fat tissue, the lowest expression was seen at the pig weighing 80 kg and the highest expression at the pig weighing 110 kg (Figure 2A). These results suggest that the expression of galectin-1 varies with breeds. In addition to that, the highest expression of galectin-1 gene in the back fat of Berkshire pig weighing 110 kg suggests that the expression pattern of galectin-1 gene is closely correlated with accumulation of fat at the pig weighing 110 kg.

A statistical analysis of RT-PCR results was carried out to verify the correlation with varieties (species/growth stage /tissue) parameters through attempting to test the statistical significance, in order to analyze the interrelationship between the expression of galectin-1 gene and two or more variables (Table 1). As shown, there is more significant

Table 1. Statistical analysis of the expression level of the galectin-1 gene by the variables and by the interaction of variables (growth stage, tissue and pig speciation)

Source (variable)	DF	Anova SS	Mean square	F value
Growth-stage	2	0.220	0.110	91.33**
Tissue	1	1.186	1.186	984.64**
Species	1	0.452	0.452	375.25**
Growth-stage×tissue	2	0.178	0.089	73.99**
Growth-stage×species	2	0.008	0.004	3.63*
Tissue×species	1	0.014	0.014	11.68**
Growth-stage×tissue×species	2	0.064	0.032	26.58**
Error	24	0.0289	0.0012	
Corrected total	35	2.152		

* $p<0.05$, ** $p<0.01$.

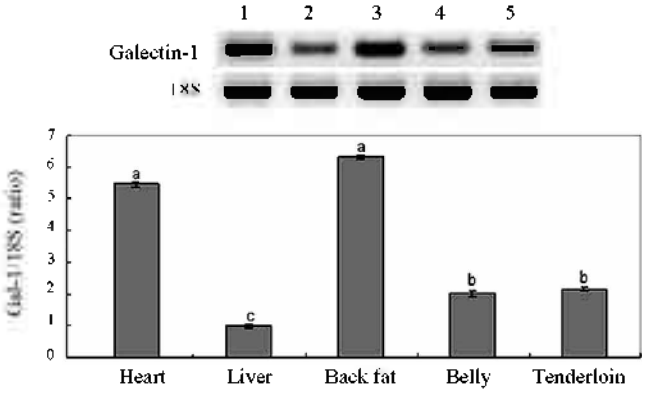

Figure 3. The expression levels of the galectin-1 mRNA in various tissues of Sancheong berkshire pigs weighing 110 kg. [a]p-[c]p indicates a significant difference ($p<0.01$). The lanes are designated as fallows: lane 1: heart; lane 2: liver; lane 3: back fat; lane 4: belly, lane 5: tenderloin.

correlation between growth stage and tissue variety ($p<0.01$). This finding present that the highest expression is seen in the back fat at the pig weighing 110 kg, while the lowest expression is observed in the tenderloin at the pig weighing 110 kg.

Correlation among variables was evaluated on the basis of the growth stage in Figure 2B ((i) and (iii)). This result suggests that the correlation among variables for determining the significance of the expression pattern of galectin-1 affects selectively its expression in the tissues and species (A; $p<0.05$, B; $p<0.01$). While Sancheong Berkshire pigs displayed the high expression in the tissues compared with Yorkshire pigs, the difference in the expression levels by growth stage between the two species was not significant. However, the expression in Yorkshire pig weighing 80 kg was less significant than in other cases (Figure 2B (i) and (iii)). The result of Figure 2B (ii) indicates the significant correlation between the breed of pig and tissue samples ($p<0.01$). Definitely, the expression level was more remarkable in Sancheong Berkshire pigs than in Yorkshire pigs and in the back fat than in the tenderloin.

Comparing different tissues derived from the pig weighing 110 kg Berkshire pig which exhibited the highest expression level of galectin-1, the highest expression was observed in the back fat, followed by the heart, tenderloin, belly and liver (Figure 3).

In order to compare quantitatively galectin-1 gene expression level of varieties (species/growth stage/tissue) parameters in pigs, a real-time PCR was performed. The relative expression of Galectin-1 in each sample was calculated from a calibration curve (Ct vs. concentration) obtained by measuring GAPDH fragment amplification after serial dilution of the same mRNA sample. The melting dissociation profiles performed on Galectin-1 and GAPDH cDNAs allowed confirmation of the specificity of the amplifications. The results of the relative quantification, normalized against GAPDH amplification, the result showed that the expression of Galectin-1 in pigs was similar to RT-PCR (Figure 4). The expression in the back fat tissue was higher than in the tenderloin tissue. But the expression in the pig weighing 60 kg of the back fat tissue was higher than 110 kg.

As a result of performing western blotting with galectin-1 to clarify the expression pattern of the protein, it was expressed in both the tenderloin and fat tissue and specifically, the expression level was higher in the fat than in the tenderloin (Figure 5). In addition, regarding the finding with different tissues derived from Sancheong Berkshire pig weighing 110 kg, the highest expression was found in the back fat followed by the heart, belly, tenderloin and liver. Interestingly, galectin-1 appeared in the form of tetramer and had a little different size in the muscles and fat tissues.

Collectively, the expression pattern of galectin-1 gene in Sancheong Berkshire pig seems closely correlated with the fat tissue.

DISCUSSION

Galectins have been identified in a wide variety of tissues of several species (Leffler et al., 1997), and has been studied by many researchers (Pieters, 2006). Also Galectin-1 shows the various expression patterns in different tissues. However, there are very little studies on the effect of galectin-1 on the meat quality of porcine. Fat is one of critically important determinants for better meat quality. As reported previously in the quality of meat (Tadayosi, 2003), Sancheong Berkshire pigs have been highly recognized regarding its good meat quality in Korea, and this study aimed to clarify the correlation between the expression pattern of galectin-1 gene and the quality of meat in Sancheong Berkshire pigs.

We isolated the gene encoding galectin-1 from cDNA in the back fat tissue of Sancheong Berkshire pig, and sequenced the entire gene. The DNA sequence of galectin-1 originated from Sancheong Berkshire pig was same with the previously known porcine sequence and highly sequence homology to that of other mammals. With the intention of verifying more various expression patterns of galectin-1 gene extracted from the porcine tissues at different growth stages, RT-PCR was mainly applied and additionally, cDNA chip, real time PCR, southern and western blotting were also conducted. Furthermore, we attempted to clarify the expression patterns of the gene in more various porcine tissues and overall, to search for the correlation between the expression pattern of galectin-1 and

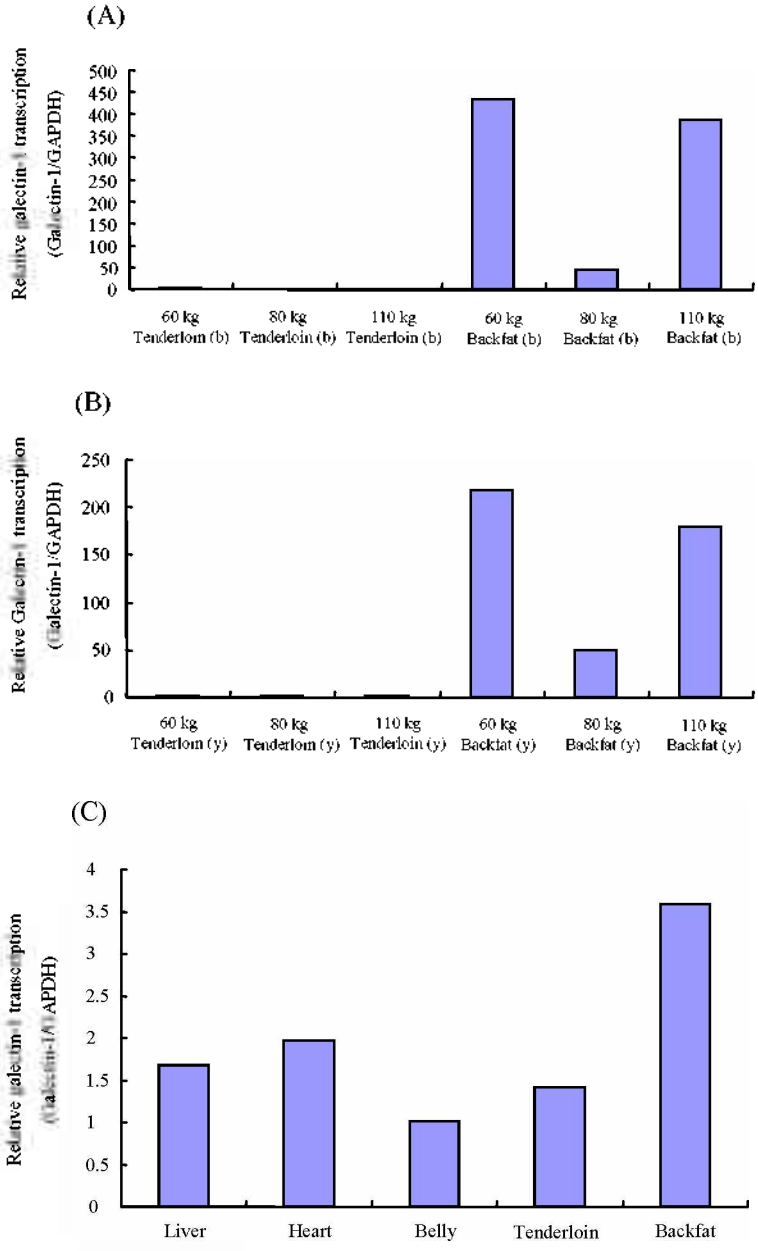

Figure 4. Analysis of transcription levels of galectin-1 gene by real time PCR in growth stage and various tissues. Values are reported as galectin-1 gene expression relative to the housekeeping transcript GAPDH. (A) The expression levels of the galectin-1 mRNA by real time-PCR with the muscle and fat tissues taken from Sancheong berkshire and Yorkshire pig in a different body weight. (B) The expression levels of the galectin-1 mRNA in various tissues of Sancheong berkshire pigs weighing 110 kg.

the meat quality.

The expression level of galectin-1 gene increased significantly fat tissue, which suggests that the expression of galectin-1 may related to mechanism of fat tissue. In addition, the expression level was higher in the back fat tissue than in the tenderloin tissue, suggesting that the presence of appropriate amount of fat in meat contributes to overall of meat quality. It is known that galectin-1 acts a modulator of adipocyte differentiation and other factors released from the fat (Wang et al., 2004). In this study, we analyzed the expression pattern of galectin-1 gene in the porcine tissues in order to account for the correlation between the expression level of galetcin-1 and the meat quality.

To clarify the differences in the expression pattern of galectin-1 gene in Sancheong Berkshire pigs and Yorkshire pigs were employed as a sort of control. Commonly, their expression levels were higher in the back fat than in the tenderloin. Comparing the two breeds, the expression level was higher in Sancheong Berkshire pigs than in Yorkshire pigs. Further, comparing the expression levels among different growth stages and various tissues, the highest

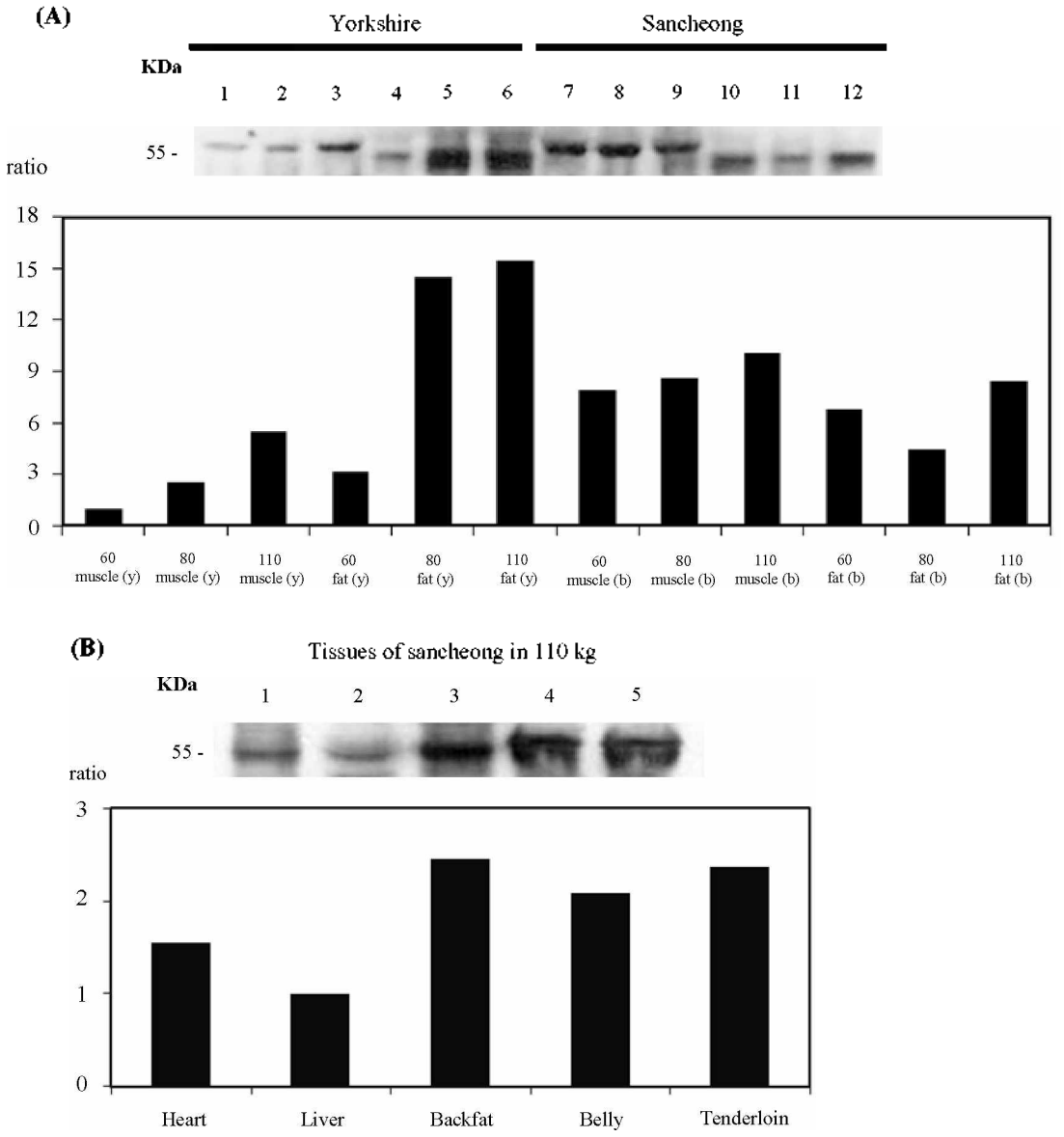

Figure 5. Western blot analysis of galectin-1 proteins extracted from pig. (A) Tenderloin and fat of galectin-1 proteins were analyzed in Yorkshire and Sancheong berkshire. (B) Various tissues of galectin-1 proteins were analyzed Sancheong berkshire pigs weighing 110 kg. (A) The lanes are designated as fallows: lane 1, tenderloin of 60 kg in Yorkshire; lane 2, 80 kg; lane 3, 110 kg; lane 4, back fat of 60 kg; lane 5, 80 kg; lane 6, 110 kg; lane 7, tenderloin of 60 kg in Sancheong; lane 8, 80 kg; lane 9, 110 kg; lane 10, back fat of 60 kg; lane11, 80 kg; lane 12, 110 kg. (B) The lanes are designated as fallows: lane 1, heart; lane 2, liver; lane 3, back fat; lane 4, belly; lane 5, tender loin.

expression level was seen in the back fat tissue of Sancheong Berkshire pig.

Western blot result of galectin-1 appeared in other form that monomer is not. In the research of Leffler et al. (2004), galectins reported that occur as monomer, dimer or higher order oligomers depending on specific case and conditions (concentration, ligand). In this study, galectin-1 was discovered specific tetramer. Despite sequence are equal in fat and muscular tissues, reason of the band size difference becomes could not know.

The proteins of galectin are bound with β-galactosides and their physiological functions are unknown. Recombinant galectin-1 plays an important role in physiological mechanisms in the whole organism including cell-cell or cell-extracellular matrix interactions, the regulation of cell growth, the maintenance of homeostasis, the induction of immunity (Perillo et al., 1998), in specific developmental processes (Hughes, 2004; Watt et al., 2004), and progression of cancer (Takenaka et al., 2004). However, it was reported that any abnormality of the primary phenotypes were not observed in galectin-1-deficient mice, probably by virtue of complementation (Poirier et al., 1993;

Colnot et al., 1998). In pigs, galectin-1 play a role in bridging agent between Chitlac and chondrocyte aggregates (Marcon et al., 2005) and regulation of progesterone production (Walzel et al., 2004).

Galectins might mediate a wide range of biological functions, but was no research about relation with fat metabolism. In our research, Galectin-1 found in backfat library and expresses the high amount in adipose tissue being secreted at fat cell differentiation. We are predicted that galectin is close connection with adipose tissue and is relation to be between fat cell and fat cell or fat cell and extracellular matrix. Like this, there are little or no studies on the roles of porcine galectin-1 as of now. According to the results of this study, it is considered that galectin-1 biosynthesis increases fat tissue in pigs which showed the highest expression.

In this study, we tried to evaluate the different expression patterns of galectin-1 gene and to verify the correlation between the expression levels and porcine meat quality. This study is meaningful as the first trial to establish such the correlation. Further, to achieve the production of better pork, we will strive constantly to search for the specific genes whose expression patterns are correlated with the meat quality.

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