บรรณานุกรม

กรมปศุสัตว์. 2553. ข้อมูลจำนวนเกษตรกรและปศุสัตว์ในประเทศไทยประจำปี 2553. ระบบ
  ฐานข้อมูลเกษตรกรผู้เลี้ยงสัตว์ (TH-LiFDS) ศูนย์สารสนเทศกรมปศุสัตว์ กระทรวงเกษตร
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  2553.

จรัญ จันทลักษณา. 2515. การผลิตโคเนื้อ. ภาควิชาสัตวบาล. คณะเกษตรศาสตร์.
  มหาวิทยาลัยเกษตรศาสตร์.

จุฑารัตน์ เศรษฐกุล และญาณิน โอภาสพัฒนกิจ. 2548. คุณภาพเนื้อโคภายใต้ระบบการผลิตและ
  การตลาดของประเทศไทย. กรุงเทพมหานคร: บริษัทสุพีเรียพริ้นดิ้งเฮ้าส์ จำกัด. 85 หน้า.

สุรชัย ชาครียรัตน์. 2545. การสืบพันธุ์และการผสมเทียมโค-กระบือ. กรุงเทพฯ: สำนักส่งเสริมและ
  ฝึกอบรม มหาวิทยาลัยเกษตรศาสตร์. 439 หน้า

ศรเทพ ธัมวาสร.2548. การเลี้ยงโคเนื้อ: แนวทางการพัฒนาอาชีพของเกษตรกรไทย. พิมพ์ครั้งที่ 3
  (ปรับปรุงใหม่). กรุงเทพฯ: อักษรสยามการพิมพ์. 452 หน้า.

Blakely, J., and D.H. Bade. 1994. The science of Animal Husbandry. 6th Edition. Prentice
  Hall Career and Technology, Prentice-Hall, Inc., New Jersey, USA.

Becker, P.L., Singer J.J., Walsh J.V.J.R. and Fay F.S. 1989. Regulation of calcium
  concentration in voltage clamped smooth muscle cells. Science. 244:211–214.

Berlin J.R. and Konishi M. 1993. Ca^{2+} transients in cardiac myocytes measured with high
  and low affinity Ca^{2+} indicators. Biophys.J. 65:1632–1647.

Betts, R., S. Weinsheimer, G.E. Blouse and J. Anagli. 2003. Structural determinants of the
  calpain inhibitory activity of calpastatin peptide B27-WT. J. Biol. Chem.
  278(10):7800-7809. Epub 2002 Dec 24.

Boccard, R., I. Buchter, M. Casteels, E. Cosentino, E. Dransfield and D.E. Hood. 1981.
  Procedures for measuring meat quality characteristics in beef production
  experiments. Lives. Prod. Sci. 8: 385-397.

Boehm, M.L., T.L. Kendall, V.F. Thompson and D.E. Goll. 1998. Changes in the calpains
  and calpastatin during postmortem storage of bovine muscle. J. Anim. Sci. 76:2415-
  2434.

Calkins C.R. and G. Sullivan. 2007. Ranking of beef muscles for tenderness. In Beef facts product enhancement. National Cattlemen's Beef Association. CO: Centenniel.

Cong, M., V.F.Thompson, G.E. Goll and P.B. Antin. 1998. The bovine calpastatin gene promoter and a new N-terminal region of the protein are targets for cAMP-dependent protein kinase activity. J. Biol. Chem. 273(1):660-666.

Crawford, C. 1990. Protein and peptide inhibitors of calpains. In: Intracellular Calcium-Dependent Proteolysis. Boca Raton. FL:CRC. p.75–89.

Cundiff, L.V., K.E. Gregory and J.E. Gosey. 1993. Cattle germplasm evaluation and germplasm utilization programs at MARC. Proc. Recip. Meat Conf. 46:45.

Dargelos, E., S. Poussard, C. Brule, L. Daury, and P. Cottin. 2008. Calcium-dependent proteolytic system and muscle dysfunctions: a possible role of calpains in sarcopenia. Biochimie. 90: 359-368.

Dransfield, E. 1994. Tenderness of meat, poultry and fish. in Pearson, A. M. and Dutson, T. R. Quality Attributes and their Measurement in Meat, Poultry and Fish Product. London: Blackie Academic and Professional. p. 289-315.

Delgado, E.F., G.H. Geesink, J.A. Marchello, D.E. Goll and M. Koohmaraie. 2001. The calpain system in three muscles of normal and callipyge sheep. J. Anim. Sci. 79: 398-412.

De Tullio, R., Averna M., Stifanese R., Parr T., Bardsley R.G., Pontremoli S., Melloni E. 2007. Multiple rat brain calpastatin forms are produced by distinct starting points and alternative splicing of the N-terminal exons. Arch. Biochem. Biophys. 465(1):148–156.

Ensminger, M.E. 1993. Dairy Cattle Science. 5th Edition. The Interstate Printer and Publisher, Inc. Danville, Illinois, USA. p. 550.

Emori, Y., H. Kawasaki, S. Imajoh, Y. Minami and K. Suzuki. 1988. All four repeating domains of the endogenous inhibitor for calcium-dependent protease independently retain inhibitory activity. Expression of the cDNA fragments in *Escherichia coli*. J. Biol. Chem. 263(5):2364-2370.

Ernst, C.W., A. Robic, M. Yerle, L. Wang and M.F. Rothschild. 1998. Mapping of calpastatin and three microsatellites to porcine chromosome 2q2.1-q2.4. Anim. Genet. 29(3):212-215.

Fries, R. and A. Ruvinsky. 1999. The Genetics of Cattle. CABI Publishing, New York.

Geesink, G.H. and M. Koohmaraie. 1999. Technical note: a rapid method for quantification of calpain and calpastatin activities. J. Anim. Sci. 77:3225-3229.

Geesink, G.H., D. Nonneman and M. Koohmaraie. 1998. An improved purification protocol for heart and skeletal muscle calpastatin reveals two isoforms resulting from alternative splicing. Arch. Biochem. Biophys. 356:19–24.

Geesink, G.H., A. Ouali and F.J.M. Smulders. 1992. Tenderisation, calpain/ calpastatin activities and osmolality of 6 different beef muscles. In: Proc. 38th Int. Cong. of Meat Sci. and Technol., Clermont Ferrand. pp 363-366.

Geesink, G.H., F.J. Smulders, H.L. van Laack, J.H. van der Kolk, T. Wensing and H.J. Breukink. 1993. Effect on meat quality of the use of clenbuterol in veal calves. J. Anim. Sci. 71:1161-1170.

Goll, D.E., Kleese W.C., Okitani A., Kumamoto T., Cong J., and Kapprell H.-P. 1990. Historical background and current status of the Ca2Γ-depen-dent proteinase system. In: Intracellular Calcium-Dependent Pro-teolysis, edited by Mellgren RL and Murachi T. Boca Raton, FL:CRC. p.3–24.

Goll, D.E., V.F. Thompson, H. Li, W. Wei and J. Cong. 2003. The calpain system. Physiol Rev. 83(3):731-801.

Hall, T. 2007. BioEdit: Biological Sequence Alignment Editor for Windows 95/98/NT/XP. [Online]. Available: http://www.mbio.ncsu.edu/BioEdit/bioedit.html.

Hoppe, B.L., Conti-Tronconi, B.M., and Horton, R.M. 1992. Gel loading dyes compatible with PCR. BioTechniques 12:679-680.

Huang, X. 1992. A contig assembly program based on sensitive detection of fragment Overlaps. Genomics. 14(1): 18-25.

Imajoh S., H. Kawasaki, Y. Emori and K. Suzuki. 1987. Calcium-activated neutral protease inhibitor from rabbit erythrocytes lacks the N-terminal region of the liver inhibitor but retains three inhibitory units. Biochem. Biophys. Res. Commun. 146:630–632.

Inazawa, J., H. Nakagawa, S. Misawa, T. Abe, S. Minoshima, R. Fukuyama, M. Maki, T. Murachi, M. Hatanaka and N. Shimizu. 1990. Assignment of the human calpastatin

gene (CAST) to chromosome 5 at region q14-q22. Cytogenet Cell Genet. 54(3-4):156-158.

Inomata M., Nakamura M., Imajoh-ohmi S, and Kawashima S. 1993. A variety of calpain/calpastatin systems in mammalian erythrocytes. Biochim. Biophys. Acta. 1178:207–214.

Inoue H., Nojima H., and Okayama H. 1990. High efficiency transformation of *Escherichia coli* with plasmids. Gene 96: 23-28.

Iwamoto, H., Miura, T., Okamura, T., Shirakawa, K., Iwatate, M., Kawamura, S., Tatsuno, H., Ikeda, Y., Matsuzaki, M. 1999. Calpain inhibitor-1 reduces infarct size and DNA fragmentation of myocardium in ischemic/reperfused rat heart. J. Cardiovasc. Pharmacol. 33(4):580-586.

Jiao W., McDonald D.Q., Coxon J.M. and Parker E.J. 2010. Molecular modeling studies of peptide inhibitors highlight the importance of conformational prearrangement for inhibition of calpain. Biochem.. 49(26):5533-5539.

Kapprell, H.P. and D.E. Goll. 1989. Effect of Ca^{2+} on binding of the calpains to calpastatin. J. Biol. Chem. 264:17888-17896.

Kawasaki H., Emori Y. and Suzuki K. 1993. Calpastatin has two distinct sites for interaction with calpain – effect of calpastatin fragments on the binding of calpain to membranes. Arch.Biochem.Biophys. 305:467–472.

Koohmaraie, M. 1988. The role of endogenous proteases in meat tenderness. Proc. Recip. Meat Conf. 41:89.

Koohmaraie, M., G. Whipple, D.H. Kretchmar, J.D. Crouse, and H.J. Mersmann. 1991. Postmortem proteolysis in longissimus muscle from beef, lamb and pork carcasses. J. Anim. Sci. 69:617-624.

Koohmaraie, M. 1992. Role of the neutral proteinases in postmortem muscle protein degradation and meat tenderness. Proc. Recip. Meat Conf. 45:63.

Koohmaraie, M. 1994. Muscle protinases and meat ageing. Meat Sci. 36: 93-104.

Koohmaraie, M. 1996. Biochemical factors regulating the toughening and tenderization process of meat. Meat Sci. 43:193-201.

Kunz, S., Niederberger, E., Ehnert, C., Coste, O., Pfenninger, A., Kruip, J., Wendrich, T.M., Schmidtko, A., Tegeder, I., Geisslinger, G. 2004. The calpain inhibitor MDL 28170 prevents inflammation-induced neurofilament light chain breakdown in the spinal cord and reduces thermal hyperalgesia. Pain. 110(1):409-418.

Laemmli, U.K. 1970. Cleavage of structural proteins during the assembly of the head of bacteriophage T4. Nature. 227:680 - 685.

Larkin, M.A., Blackshields, G., Brown, N.P., Chenna, R., McGettigan, P.A., McWilliam, H., Valentin, F., Wallace, I.M., Wilm, A., Lopez, R., Thompson, J.D., Gibson, T.J. and Higgins, D.G. 2007. ClustalW and ClustalX version 2. Bioinformatics. 23(21):2947-2948.

Lee, W. J., Hatanaka, M. and Maki, M. 1992a. Multiple forms of rat calpastatin cDNA in the coding region of functionally unknown amino-terminal domain. Biochim. Biophys. Acta. 1129:251-253.

Lee, W. J., Ma, H., Takano, E., Yang, H. Q., Hatanaka, M., and Maki, M. 1992b. Molecular diversity in amino-terminal domains of human calpastatin by exon skipping. J. Biol. Chem. 267:8437–8442.

Limaye, P.B., Bhave, V.S., Palkar, P.S., Apte, U.M., Sawant, S.P., Yu, S., Latendresse, J.R., Reddy, J.K., Mehendale, H.M. 2006. Upregulation of calpastatin in regenerating and developing rat liver: role in resistance against hepatotoxicity. Hepatology. 44(2):379–388.

Ma H.,Yang H.Q.,Takano E., Hatanaka M. and Maki M. 1994. Amino terminal conserved region in proteinase inhibitor domain of calpastatin potentiates its calpain inhibitory activity by interacting with the calmodulin-like domain of the proteinase. J. Biol. Chem. 269:24430–24436.

Maki M., Hatanaka M., Takano E. and Murachi T. 1990. Structure-function relationship of the calpastatins. In: Intracellular Calcium-Dependent Proteolysis, edited by Mellgren R.L. and Murachi T. Boca Raton, FL:CRC. p 37–51.

Melloni E., de Tullio R., Averna M., Tedesco I., Salamino F., Sparatore B. and Ponttremoli S. 1998. Properties of calpastatin isoforms in rat brain. FEBS Lett. 431:55–58.

Murachi T. 1989. Intracellular regulatory system involving calpain and calpastatin. Biochem. Int. 18:263–294.

Nakamura Y., Fukiage C., Ma H., Shih M., Azuma M. and Shearer T.R. 1999. Decreased sensitivity of lens-specific calpain Lp82 to calpastatin inhibitor. Experimental Eye Research. 69(2):155-162.

Nishimura T. and Goll D.E. 1991. Binding of calpain fragments to calpastatin. J. Biol. Chem. 266:11842–11850.

O'Connor, S.F., J.D. Tatum, D.M. Wulf, R.D. Green and G.C. Smith. 1997. Genetic Effects on Beef Tenderness in *Bos indicus* Composite and *Bos taurus* Cattle. J. Anim. Sci. 75:1822-1830.

Otsuka Y. and Goll D.E. 1987. Purification of the Ca^{2+}-dependent proteinase inhibitor from bovine cardiac muscle and its interaction with the millimolar Ca2+-dependent proteinase. J. Biol. Chem. 262:5839–5851.

Ouali, A., and A. Talmant. 1990. Calpains and calpastatin distribution in bovine, porcine and ovine skeletal muscle. Meat Sci. 28:331-348.

Parr, T., P.L. Sensky, R.G. Bardsley and P.J. Buttery. 2001. Calpastatin expression in porcine cardiac and skeletal muscle and partial gene structure. Arch. Biochem. Biophys. 395(1):1-13.

Pfaffl, M.W. 2004. Quantification strategies in real-time PCR. In: Bustin SA (ed), A–Z of Quantitative PCR. pp. 87–120. La Jolla, CA: IUL Biotechnology Series, International University Line.

Pringle, T.D., S.E. Williams, B.S. Lamb, D.D. Johnson and R.L. West. 1997. Carcass characteristics, the calpain proteinase system, and aged tenderness of Angus and Brahman crossbred steers. J. Anim. Sci. 75(11):2955-2961.

Posmantur, R., Kampfl, A., Siman, R., Liu, J., Zhao, X., Clifton, G.L., Hayes, R.L. 1997. A calpain inhibitor attenuates cortical cytoskeletal protein loss after experimental traumatic brain injury in the rat. Neurosci. 77(3):875-888.

Rao, M.V., Mohan, P.S., Peterhoff, C.M., Yang, D-S., Schmidt, S.D., Stavrides, P.H., Campbell, J., Chen, Y., Jiang, Y., Paskevich, P.A., Cataldo, A.M., Haroutunian, V., and Nixon R.A. 2008. Marked calpastatin (CAST) depletion in Alzheimer's disease accelerates cytoskeleton disruption and neurodegeneration: Neuroprotection by CAST overexpression. J. Neurosci. 28(47):12241–12254.

Raynaud, P., M. Gillard, T. Parr, R. Bardsley, V. Amarger and H. Leveziel. 2005a. Correlation between bovine calpastatin mRNA transcripts and protein isoforms. Arch. Biochem. Biophys. 440:46-53.

Raynaud, P., C. Jayat-Vignoles, M.P. Laforet, H. Leveziel, and V. Amarger. 2005b. Four promoters direct expression of the calpastatin gene. Arch. Biochem. Biophys. 437:69-77.

Sambrook, J., Fritsch, E.F. and Maniatis, T. 1989. Molecular Cloning: A Laboratory Cold Spring Harbor, New York: Cold Spring Habor Press.

Sazili, A.Q., Parr, T., Sensky, P.L., Jones, S.W., Bardsley, R.G. and Buttery, P.J. 2005. The relationship between slow and fast myosin heavy chain content, calpastatin and meat tenderness in different ovine skeletal muscles. Meat Sci. 69(1):17-25.

Shackelford, S.D., M. Koohmaraie, G. Whipple, T.L. Wheeler, M.F. Miller, J.D. Crouse, and J.O. Reagan. 1991. Predictors of beef tenderness: Development and verification. J. Food Sci. 56:1130-1140.

Shackelford, S.D., M. Koohmaraie, L.V. Cundiff, K.E. Gregory, G.A. Rohrer and J.W. Savell. 1994. Heritabilities and phenotypic and genetic correlations for bovine postrigor calpastatin activity, intramuscular fat content, Warner-Bratzler shear force, retail product yield, and growth rate. J. Anim. Sci. 72:857.

Shackelford, S.D., T.L. Wheeler, and M. Koohmaraie. 1995. Relationship between shear force and trained sensory panel tenderness ratings of 10 major muscles from bos-indicus and bos-taurus cattle. J. Anim. Sci. 73:3333-3340.

Spencer, M.J. and Mellgren R.L. 2002. Overexpression of a calpastatin transgene in mdx muscle reduces dystrophic pathology. Hum. Mol. Genet. 11(21):2645-2655.

Souaze F, Ntodou-Thome A, Tran CY, Rostene W, Forgez P. 1996. Quantitative RT-PCR: limits and accuracy. Biotechniques. 21(2): 280–285.

Takano E., Ma H., Yang H.Q., Maki M. and Hatanaka M. 1995. Preference of calcium-dependent interactions between calmodulin-like domains of calpain and alpastatin subdomains. FEBS. Lett. 362:93–97.

Takano, E., M. Maki, H. Mori, M. Hatanaka, T. Marti, K. Titani, R. Kannagi, T. Ooi and T. Murachi. 1988. Pig heart calpastatin: identification of repetitive domain structures

and anomalous behavior in polyacrylamide gel electrophoresis. Biochemistry. 27(6):1964-1972.

Takano, E. and Murachi T. 1982. Purification and some properties of human erythrocyte calpastatin. J. Biochem. 92:2021–2028.

Takano, J., M. Watanabe, K. Hitomi and M. Maki. 2000. Four types of calpastatin isoforms with distinct amino-terminal sequences are specified by alternative first exons and differentially expressed in mouse tissues. J. Biochem. 128(1):83-92.

Tradewell, M.L. and Durham H.D. 2010. Calpastatin reduces toxicity of SOD1G93A in a culture model of amyotrophic lateral sclerosis. Neuroreport. 21(15): 976-979.

Wang L.F., Wei S.G., Miao S.Y., Liu Q.Y. and Koide S.S.Calpastatin gene in human testis. Biochem. Mol. BiolInt. 33:245–252.

Warriss, P.D. 2000. Meat Science. UK: CAB International.

Wheeler, T.L., Savell, J.W., Cross, H.R., Lunt, D.K. and Smith, S.B. 1990. Mechanism associated with the variation in tenderness of meat from Brahman and Hereford cattle. J. Anim. Sci. 68:4206-4220.

Whipple, G., M. Koohmaraie, M.E. Dikeman, J.D. Crouse, M.C. Hunt and R.D. Klemm. 1990. Evaluation of attributes that affect longissimus muscle tenderness in *Bos taurus* and *Bos indicus* cattle. J. Anim. Sci. 68:2716.

Wulf, D.M., J.D. Tatum, R.D. Green, J.B. Morgan, B.L. Golden and G.C. Smith. 1996. Genetic influences on beef longissimus palatability in Charolais- and Limousin-sired steers and heifers. J. Anim. Sci. 74:2394.

ภาคผนวก

ภาคผนวก ก

ลำดับนิวคลีโอไทด์ของยีนคาลปาสเตติน

ลำดับนิวคลีโอไทด์ของยีนคาลปาสเตตินจากโคสายพันธุ์ชาโรเลส์ BT-CAST1 (NM_001030318), BT_CAST2 (NM_001030319) และ BT_CAST4 (NM_001030320) เปรียบเทียบกับโคพื้นเมืองไทยและโคกำแพงแสน

จุดไข่ปลา (...) แสดงนิวคลีโอไทด์ที่ไม่มีความแตกต่างเมื่อเทียบกับลำดับนิวคลีโอไทด์ของยีน BT_CAST1, กรอบสี่เหลี่ยม แสดงขอบเขตของโดเมน I - IV

แถบสีเหลือง แสดงตำแหน่ง start codon (ATG), แถบสีเทา แสดงตำแหน่ง stop codon (TAA), แถบสีฟ้า แสดงลำดับนิวคลีโอไทด์ที่ทำสำแหน่งได้จากงานวิจัยนี้

```
BT_CAST1      GGAGCTCTCGCGGGTCGGGGGTCGGGGGTCGGAAAAGCTGCCTCACAGGCGCGCCGCCCCTCCGCGCCCTCGCTCCCTCCCAGCCGCTCCCGGCTCCA  100
BT_CAST2      .....................................................G..TTAG....AGT.AG.TCTG.GG.AG.T.GAGTG..AAC..G   44
BT_CAST3      ------------------------------------------------------------------------------------------------    1
BT_CAST4      ------------------------------------------------------------------------------------------------    1
Thai_Native   ------------------------------------------------------------------------------------------------    1
Kampaengsaen  ------------------------------------------------------------------------------------------------    1

BT_CAST1      GCCTCCCTTCCAGGCTCCGCGCCGCCCGCCCGGAGGCAGCGCTCGCACCGGCCTCGCACCGGCCTCCCCGCCTCCCCGCGGCCC                  200
BT_CAST2      TGGC.T..-------...G..T.A....TGTCCTC..C..G.C.T.G.T...T.GC.CGT.TCT..AA..CAT.AT.. .G.AT...AT            135
BT_CAST3      ------------------------------------------------------------------------------------                1
BT_CAST4      ------------------------------------------------------------------------------------                1
Thai_Native   ------------------------------------------------------------------------------------                1
Kampaengsaen  ------------------------------------------------------------------------------------                1

BT_CAST1      CGGCCGCGCTGCCCGCCACACCCAGGAGCCATGTCAGTAGAAAAACCAGTGGATCGCCTTCCAAGTCAGGAGAAAAGAAGGATCAGATGAGAAAAAGCAA  300
BT_CAST2      TT..AA...GTG.T......AC...................................................................            231
BT_CAST3      ------------------------------------------------------------------------------------------------    1
BT_CAST4      ------------------------------------------------------------------------------------------------    1
Thai_Native   ------------------------------------------------------------------------------------------------    1
Kampaengsaen  ------------------------------------------------------------------------------------------------    1
```

BT_CAST1

```
CAAGCCTTGGGAGCAGTCAGCTCTCCAGAACTCAGGTGATGAAAAGCCCTGGTCCCCAAGGTAACTACTTCCTCTGCGTCAGCCAGCAAGTCTTCCAG   400
```

BT_CAST2   331
BT_CAST3   1
BT_CAST4   1
Thai_Native   1
Kampaengsaen   1

BT_CAST1

```
TATGAATCCCACAGAAGCCAAGGCTATTCCAGGCAGCAAACAGCTGGAAGGACACCGGCCATTCTCCTAACAGGAAAAGACACAAAAAACAGGCTGTAAAAACA   500
```

BT_CAST2   ...ATG...   431
BT_CAST3   ...ATG...   99
BT_CAST4   1
Thai_Native   1
Kampaengsaen   1

BT_CAST1

```
GAACCTGAGAAGAAGCCACAATCATCTAAGCCATCTGTGGTTCATGAGAAAAAAGAGTAAAGCCAAGGAACACACAGAGCCAAAAAGCCTAC   600
```

BT_CAST2   531
BT_CAST3   199
BT_CAST4   1
Thai_Native   1
Kampaengsaen   1

BT_CAST1

```
CCAAGCACTCATCAGATACAGGAAGCAAGCATGCCTCCTAAGGAAAAAGCCGGTTTCCAAATCAAGTGAGCAGCCACCATCAGAGAAATCAACAAAACCAAA   700
```

Domain I

BT_CAST2   631
BT_CAST3   299
BT_CAST4   1
Thai_Native   1
Kampaengsaen   1

BT_CAST1

```
GACCAAGTCACAGGACAAGATCTCCGGTGGTGAAAGAGCACTGTTCCTGCCGACATCGTTCCTGCTGCTGCAGCCAGCTGACAAGAATAAAGAGAAAT   800
```

Domain I

BT_CAST2   731
BT_CAST3   399
BT_CAST4   1
Thai_Native   1
Kampaengsaen   1

BT_CAST1 / BT_CAST2 / BT_CAST3 / BT_CAST4 / Thai_Native / Kampaengsaen sequence alignment

Block 1 (positions ending 900, 831, 499, 1, 1, 1)

Domain I

```
BT_CAST1      ...AAATTGTTAACATCGGCCGTACCAGCTGAATCTAAACCAAGTAAACCAAGTCAGACATGGACACTGCTCTGAGTGACTTAATAGACACTTTAG  900
BT_CAST2      831
BT_CAST3      499
BT_CAST4      1
Thai_Native   1
Kampaengsaen  1
```

Block 2 (positions ending 1000, 931, 599, 1, 1, 1)

Domain I

```
BT_CAST1      GAGAAACCTGAAAGAGATGAAAGAGAAGATAACACAATATACCGGACCGGAAGTGTCGGATCCAATGAGTTCTACCTACATAGAGGAACTGGGTAAAAGAGA  1000
BT_CAST2      931
BT_CAST3      599
BT_CAST4      1
Thai_Native   1
Kampaengsaen  1
```

Block 3 (positions ending 1100, 1031, 699, 1, 1, 1)

Domain I

```
BT_CAST1      ATCCACACTTCCTCCAAAATATAAGGAACTTCTGAATAAAGAAGAAGGATCGCAGGGCCCTCCTCCAGACTCCTTGAAACCCTGGGGCCCAATGATGCC  1100
BT_CAST2      1031
BT_CAST3      699
BT_CAST4      1
Thai_Native   1
Kampaengsaen  1
```

Block 4 (positions ending 1200, 1131, 799, 58, 1, 1)

Domain II

```
BT_CAST1      ATCGATGCCTTGTCATCCGACTTCACCTGCAGTTCCCTACAGCTGATGCAAAGAAAATCTGAGAAAGAGAGCTTTAAAAGCTCAGT  1200
BT_CAST2      1131
BT_CAST3      799
BT_CAST4      58
Thai_Native   1
Kampaengsaen  1
```

Block 5 (positions ending 1300, 1231, 899, 147, 1, 1)

Domain II

```
BT_CAST1      CAGCTGGGGTGATCAGAAGTGCTGCTCCACCCCAAGAGAGAAAAAGGAAAGTGGAAAAGGATGCCATGACTGAGCAGCCCTGTCAGCCTC  1300
BT_CAST2      1231
BT_CAST3      899
BT_CAST4      147
Thai_Native   1
Kampaengsaen  1
```

BT_CAST1
BT_CAST2
BT_CAST3
BT_CAST4
Thai_Native
Kampaengsaen

Domain II
Domain II
Domain III
Domain III
Domain III

Block 1 (positions): 1400, 1331, 999, 240, 1
Block 2 (positions): 1500, 1431, 1099, 340, 1, 1
Block 3 (positions): 1600, 1531, 1199, 440, 1, 1
Block 4 (positions): 1700, 1631, 1299, 540, 85, 85
Block 5 (positions): 1800, 1731, 1399, 640, 185, 185

Domain IV

```
BT_CAST1     CCCGGGAAGCCAAAGGCATCAGAGAAGCCCAAGGCATCAGAGAGAAACCTGCAGGTGCCCAGGACCCATTGATGCCCCTCAGGGGGACTTTGACAGCTGTC  2400
BT_CAST2     ..................................................................................................  2331
BT_CAST3     ..................................................................................................  1999
BT_CAST4     ..................................................................................................  1240
Thai_Native  ..................................................................................................   785
Kampaengsaen ..................................................................................................   785

BT_CAST1     CCTCGACTACAGAGAAACCTGACAGACACACCCAAAGGACCTGCTTCCAGTGCCGAAGCACCTAGGAATGGCGGGAAAGCAAAGGAATTC  2500
BT_CAST2     .........................................................................................  2431
BT_CAST3     .........................................................................................  2099
BT_CAST4     .........................................................................................  1340
Thai_Native  .........................................................................................   885
Kampaengsaen .........................................................................................   885

BT_CAST1     CACAAAGGCAAAGGAGGAAACTTCCAAGCCAAAAGCTGATGGAAAAAGTACAAGTTAAAGTTCACACTATTTGGTATCTGCATATAAAATCTTCAGCGGGG  2600
BT_CAST2     ..................................................................................................  2531
BT_CAST3     ..................................................................................................  2199
BT_CAST4     ..................................................................................................  1440
Thai_Native  ..................................................................................................   985
Kampaengsaen ..................................................................................................   985

BT_CAST1     TAGATGGTGGTGACTTCTGAAGAAGAAGGCTTTGATAACAGAAACAATTTCTGGGTGGCTTGGAGACAGTGGTATTGCTGAGTCTTTTGACCTCCTAAAC  2700
BT_CAST2     .................................................................................................  2631
BT_CAST3     .................................................................................................  2299
BT_CAST4     .................................................................................................  1540
Thai_Native  .................................................................................................  1002
Kampaengsaen .................................................................................................  1002

BT_CAST1     ATTGTCTGTTATCTTTTCCTGAAAAGAAACTGAATTGTCTGGTTCACCTGTGTTATTCTACTGAGTATGATAAACTTTAAAATTTTTAAAAATTGCCTT  2800
BT_CAST2     ................................................................................................  2731
BT_CAST3     ................................................................................................  2399
BT_CAST4     ................................................................................................  1640
Thai_Native  ................................................................................................  1002
Kampaengsaen ................................................................................................  1002
```
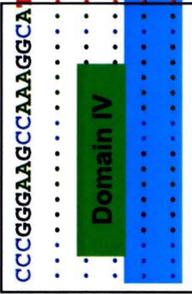
Sequence alignment figure.

Block 1 — row labels and end positions:
Label	Position
BT_CAST1	2900
BT_CAST2	2831
BT_CAST3	2499
BT_CAST4	1740
Thai_Native	1100
Kampaengsaen	1100

BT_CAST1: CAGTTGGGAGAGAAAGGAACTTTATATTTCTAAGAGATACATTTGATAGTGTTCTTAAAGCAGCACACAAAAAAGGAAAAACCTTTGCAAACTTTTGCACA

Block 2 — row labels and end positions:
Label	Position
BT_CAST1	3000
BT_CAST2	2931
BT_CAST3	2599
BT_CAST4	1840
Thai_Native	1200
Kampaengsaen	1200

BT_CAST1: TTCTCCCCACAGTGCCTGTAAATCTCATTAGTATTTTCAATTTGCACTTATTTTGTGTTAGCATTTGGAAAACGATGCCTCACGTGTTCTTCAGTGTT

Block 3 — row labels and end positions:
Label	Position
BT_CAST1	3100
BT_CAST2	3031
BT_CAST3	2699
BT_CAST4	1940
Thai_Native	1300
Kampaengsaen	1300

BT_CAST1: CTGATTTCTCATGACCCCTTTCCTCTTAGACTTGTGTGGACTGTGTTGATGTTTCTTTGGGTTGTGTTGTTATAAGTCAGTCATAAAATACTGTGCATTGGG

Block 4 — row labels and end positions:
Label	Position
BT_CAST1	3200
BT_CAST2	3131
BT_CAST3	2799
BT_CAST4	2040
Thai_Native	1325
Kampaengsaen	1325

BT_CAST1: CACATGTCTCCTCTTGAGCTGCTAATCGTAGAGACCCTGGACAGACCAGGAAGCGCCGCACCCCCCTTTCAGGTTGAACTTCTCTTTGCCAGAGGTTAGG

ภาคผนวก ข
ข้อมูลและผลการวิเคราะห์ทางสถิติ

ค่าแรงตัดผ่านเนื้อในกล้ามเนื้อสันนอก

ตารางที่ ข1 ค่าแรงตัดผ่านเนื้อในกล้ามเนื้อสันนอกของโคพื้นเมืองและโคกำแพงแสน

ตัวอย่างที่	ค่าแรงตัดผ่านเนื้อ (กก.ต่อตร.ซม.)	
	โคพื้นเมือง (NT)	โคกำแพงแสน (KU)
1	10.32	7.26
2	12.37	7.58
3	12.45	10.27
4	10.17	11.26
5	12.49	11.10
6	18.34	11.38
7	17.51	10.31
8	19.86	11.15
9	15.21	11.36
10	15.30	9.99
11	19.33	12.16
12	15.81	12.06
13	17.50	13.47
14	16.71	11.68
15	16.71	10.97
Mean	**15.34**	**10.80**
SD	**3.12**	**1.62**

การแสดงออกของยีนคาลปาสเตตินในกล้ามเนื้อและอวัยวะโคพื้นเมือง

ตารางที่ ข2 ผลการประเมินประสิทธิภาพการเพิ่มปริมาณยีนคาลปาสเตตินโดยเทคนิค Realtime PCR

สัดส่วนการเจือจาง	ซ้ำที่	ค่า E	ค่า CT	ค่า Tm
1:100	1	1.62	9.42	80.8
	2	1.70	9.62	80.6
1:1000	1	1.72	12.00	80.6
	2	1.58	12.11	80.8
1:10000	1	1.60	15.35	80.4
	2	1.54	15.14	80.6
1:100000	1	1.60	18.19	80.4
	2	1.55	18.07	80.4
ค่าเฉลี่ย		1.61		80.6

ตารางที่ ข3 ค่าเฉลี่ยปริมาณการแสดงออกของยีนคาลปาสเตตินในกล้ามเนื้อของโคพื้นเมือง*

ชนิดกล้ามเนื้อ	ค่า $E^{\Delta CP(actin-cast)}$	relative expression to LD
LD	168.54	1.00
TA	202.61	1.202
LT	179.86	1.067
SM	57.96	0.344
ST	169.23	1.004
RA	392.86	2.331

* n = 3

LD = Longissimus dorsi, LT= Longissimus troracis, TA = Transversus abdominis,

SM = Semimembranosus, ST = Semitendinosus and RA = Rectus abdominis

ตารางที่ ข4 ค่าเฉลี่ยปริมาณการแสดงออกของยีนคาลปาสเตตินในอวัยวะของโคพื้นเมือง*

อวัยวะ	ค่า $E^{\Delta CP(actin-cast)}$	relative expression to LD
อัณฑะ (T)	6.51	0.039
ลำไส้ (I)	28.40	0.169
ไต (K)	6.11	0.036
หัวใจ (H)	28.62	0.170
ม้าม (S)	5.60	0.033
สมอง (B)	0.39	0.002
ตับ (L)	1.18	0.007
กล้ามเนื้อสันนอก (LD)	168.54	1.000

* n = 3

ตารางที่ ข5 ค่า $E^{\Delta CP(actin-cast)}$ ในกล้ามเนื้อสันนอกของโคพื้นเมืองและโคกำแพงแสน

ตัวอย่างที่	ค่า $E^{\Delta CP(actin-cast)}$	
	โคพื้นเมือง (NT)	โคกำแพงแสน (KU)
1	142.43	116.29
2	150.44	116.29
3	140.71	71.78
4	72.36	98.88
5	124.09	193.05
6	216.26	98.08
7	205.15	86.67
8	220.86	141.28
9	162.16	70.62
10	113.50	71.49
11	161.50	114.42
12	201.85	90.81
13	264.86	108.55
14	189.17	152.59
15	162.82	92.30
Mean	**168.54**	**108.21**
SD	**48.793**	**33.518**

ค่ากิจกรรมการทำงานของเอนไซม์คาลปาสเตติน

ตารางที่ ข6 ค่าการดูดกลืนแสงของตัวอย่างที่ชะออกจากคอลัมน์ DEAE-Sephacel™

fraction no.	ค่าการดูดกลืนแสงที่ 280 nm
1	0.000
2	0.000
3	0.000
4	0.000
5	0.056
6	0.133
7	0.767
8	2.217
9	3.353
10	1.403
11	0.925
12	0.546
13	0.383
14	0.297
15	0.137
16	0.077
17	0.043
18	0.017
19	0.000
20	0.000

ตารางที่ ข7 ค่ากิจกรรมการทำงานของเอนไซม์คาลปาสเตตินในกล้ามเนื้อสันนอกของโคพื้นเมือง

โคพื้นเมือง (NT)	ค่ากิจกรรมการทำงานของเอนไซม์คาลปาสเตติน		
	Unit/100µl	Unit/17ml	Unit/g of meat
1	0.292	49.58	24.79
2	0.300	51.06	25.53
3	0.304	51.68	25.84
4	0.268	45.50	22.75
5	0.267	45.45	22.72
6	0.295	50.21	25.10
7	0.282	47.88	23.94
8	0.299	50.77	25.39
9	0.309	52.47	26.24
10	0.291	49.47	24.74
Mean	**0.291**	**49.41**	**24.70**
SD	**0.014**	**2.428**	**1.216**

ตารางที่ ข8 ค่ากิจกรรมการทำงานของเอนไซม์คาลปาสเตตินในกล้ามเนื้อสันนอกของโคกำแพงแสน

โคกำแพงแสน (KU)	ค่ากิจกรรมการทำงานของเอนไซม์คาลปาสเตติน		
	Unit/100μl	Unit/17ml	Unit/g of meat
1	0.193	32.81	16.41
2	0.272	46.18	23.09
3	0.265	44.99	22.50
4	0.284	48.34	24.17
5	0.264	44.88	22.44
6	0.292	49.58	24.79
7	0.289	49.13	24.57
8	0.286	48.68	24.34
9	0.262	44.60	22.30
10	0.303	51.51	25.76
11	0.297	50.43	25.22
12	0.285	48.51	24.25
13	0.296	50.26	25.13
Mean	**0.276**	**46.92**	**23.46**
SD	**0.028**	**4.805**	**2.402**

การวิเคราะห์ค่าความแตกต่างทางสถิติของโคพื้นเมืองและโคกำแพงแสนโดย T-test

1) ค่าแรงตัดผ่านเนื้อในกล้ามเนื้อสันนอก

Variable	breed	N	Lower CL Mean	Mean	Upper CL Mean	Lower CL Std Dev	Std Dev	Upper CL Std Dev	Std Err
sf	KU	15	9.9056	10.8	11.694	1.1825	1.6151	2.5472	0.417
sf	NT	15	13.613	15.339	17.064	2.2814	3.1161	4.9144	0.8046
sf	Diff (1-2)		-6.395	-4.539	-2.682	1.9695	2.4818	3.3565	0.9062

T-Tests

Variable	Method	Variances	DF	t Value	Pr > |t|
sf	Pooled	Equal	28	-5.01	<.0001
sf	Satterthwaite	Unequal	21	-5.01	<.0001

Equality of Variances

Variable	Method	Num DF	Den DF	F Value	Pr > F
sf	Folded F	14	14	3.72	0.0194

2) ปริมาณการแสดงออกของยีนคาลปาสเตตินในกล้ามเนื้อสันนอก [ค่า $E^{\Delta CP\ actin-cast}$]

Variable	breed	N	Lower CL Mean	Mean	Upper CL Mean	Lower CL Std Dev	Std Dev	Upper CL Std Dev	Std Err
exp	KU	15	89.645	108.21	126.77	24.539	33.518	52.861	8.6543
exp	NT	15	141.52	168.54	195.56	35.722	48.793	76.951	12.598
exp	Diff (1-2)		-91.65	-60.34	-29.03	33.218	41.858	56.611	15.284

T-Tests

Variable	Method	Variances	DF	t Value	Pr > |t|
exp	Pooled	Equal	28	-3.95	0.0005
exp	Satterthwaite	Unequal	24.8	-3.95	0.0006

Equality of Variances

Variable	Method	Num DF	Den DF	F Value	Pr > F
exp	Folded F	14	14	2.12	0.1723

3) ค่ากิจกรรมการทำงานของเอนไซม์คาลปาสเตตินในกล้ามเนื้อสันนอก

Variable	breed	N	Lower CL Mean	Mean	Upper CL Mean	Lower CL Std Dev	Std Dev	Upper CL Std Dev	Std Err
acti	KU	13	22.008	23.459	24.911	1.7223	2.4019	3.9648	0.6662
acti	NT	10	23.834	24.704	25.574	0.8364	1.2159	2.2198	0.3845
acti	Diff (1-2)		-2.979	-1.245	0.4894	1.5252	1.9825	2.8331	0.8339

T-Tests

Variable	Method	Variances	DF	t Value	Pr > |t|
acti	Pooled	Equal	21	-1.49	0.1504
acti	Satterthwaite	Unequal	18.6	-1.62	0.1224

Equality of Variances

Variable	Method	Num DF	Den DF	F Value	Pr > F
acti	Folded F	12	9	3.90	0.0487

ผลการวิเคราะห์ค่าสหสัมพันธ์ Pearson Correlation Coefficient

1) The CORR Procedure โคพื้นเมือง

```
                    3  Variables:    sf      exp      acti

                          Simple Statistics

Variable      N         Mean      Std Dev        Sum     Minimum     Maximum
sf           15     15.33867      3.11612    230.08000    10.17000    19.86000
exp          15    168.54400     48.79271         2528    72.36000   264.86000
acti         10     24.70400      1.21592    247.04000    22.72000    26.24000

                     Pearson Correlation Coefficients
                       Prob > |r| under H0: Rho=0
                         Number of Observations

                             sf          exp         acti

                 sf     1.00000      0.73633     -0.28037
                                     0.0017       0.4327
                            15           15           10

                 exp    0.73633      1.00000     -0.35621
                        0.0017                    0.3124
                            15           15           10

                 acti  -0.28037     -0.35621      1.00000
                        0.4327       0.3124
                            10           10           10
```

2) The CORR Procedure โคกำแพงแสน

```
                    3  Variables:    sf      exp      acti

                          Simple Statistics

Variable      N         Mean      Std Dev        Sum     Minimum     Maximum
sf           15     10.80000      1.61512    162.00000     7.26000    13.47000
exp          15    108.20667     33.51793         1623    70.62000   193.05000
acti         13     23.45923      2.40186    304.97000    16.41000    25.76000

                     Pearson Correlation Coefficients
                       Prob > |r| under H0: Rho=0
                         Number of Observations

                             sf          exp         acti

                 sf     1.00000      0.04562      0.65533
                                     0.8718       0.0150
                            15           15           13

                 exp    0.04562      1.00000     -0.17575
                        0.8718                    0.5657
                            15           15           13

                 acti   0.65533     -0.17575      1.00000
                        0.0150       0.5657
                            13           13           13
```

ภาคผนวก ค
ผลงานวิจัยที่เผยแพร่

การประชุมนักวิจัยรุ่นใหม่พบเมธีวิจัยอาวุโส สกว. ในปี พ.ศ. 2550

Calpastatin Gene Study in Thai Native Cattle (*B. indicus*) and Kampaengsaen Cattle (*B. indicus* x *B. taurus*)

Jirajaroenrat K.*, Opatpatanakit Y., Sethakul J.

Department of Animal Production, Faculty of Agricultural Technology,
King Mongkut's Institute of Technology Ladkrabang, Bangkok, Thailand 10520
*e-mail: kjkanya@kmitl.ac.th

Introduction

Calpastatin is a specific protein inhibitor of the calcium dependent proteinases μ- and m- calpain[1,2]. This enzyme system is known to be involved in meat tenderization, which is especially important in beef quality[3]. Four alternatively spliced mRNA transcripts of calpastatin gene (Type I, II, III and IV) have been reported in *Bos taurus* cattle[4]. The difference between them resides in their 5'-termini. The corresponding data for *B. indicus* has not been reported yet.

Objective

To study the calpastatin mRNA transcription in Thai native cattle (*B. indicus*) and Kampaengsaen cattle, the Thai developed *B. indicus* x *B. taurus* crossbred, in order to understand the relationship between calpastatin regulation and meat quality.

Method

The calpastatin cDNAs were cloned from skeletal muscle (Longissimus dorsi) by RT-PCR. Specific primers were designed based on the four *B. taurus* calpastatin mRNAs (Figure 1). Methodology flow chart is shown in Figure 2.
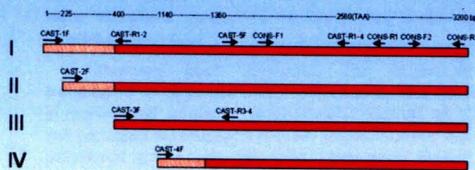
Figure 1 Primer design based on *B. taurus* mRNA transcripts (type I-IV)
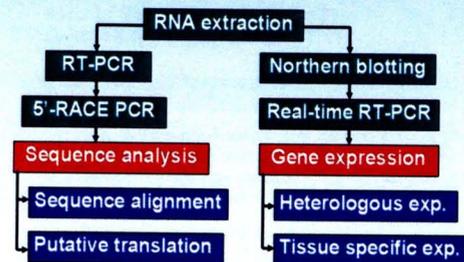
Figure 2 Research methodology flowchart

Conclusion

The 3'-termini of calpastatin mRNA transcripts in *B. taurus* and *B. indicus* share highly conserved sequence. Therefore this gene region may be important for enzyme function. The 5'-termini of the mRNAs in both Thai breeds and their expression in different muscles and tissues are under further investigation.

Acknowledgement

Result

Two cDNA fragments of Thai native and Kampaengsaen breeds were amplified (Figure 3) and sequenced (Figure 4), labelled CONS1 and CONS2 fragments, corresponding to the 3'-end of coding region and the 3'-untranslated region of calpastatin gene respectively. The CONS1 fragments of both breeds (1,002 bp) shared 100% of the nucleotide identity with the *B. taurus* transcripts and the CONS2 fragments (323 bp) shared 98% and 99% of nucleotide identity with the *B. taurus* transcripts respectively.
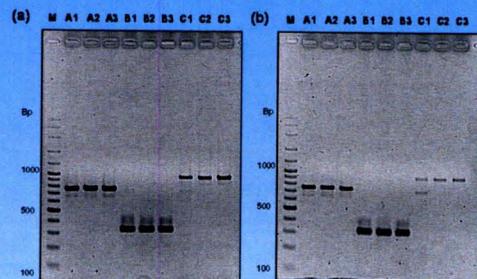

Figure 3 RT-PCR products of Thai native (a) and Kampaengsaen (b) cattle no. 1, 2 and 3 with primer sets: CONS-F1/CONS-R1 (A), CONS-F2/CONS-R2 (B) and CAST-5F/CAST-R1-4 (C).
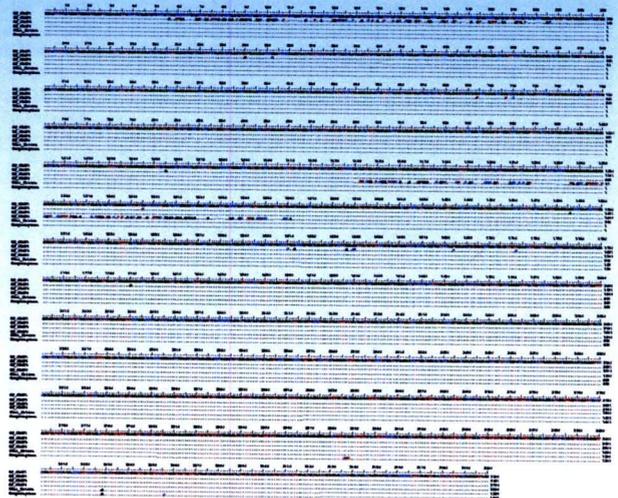

Figure 4 Calpastatin sequence alignment of *Bos taurus*: BT-CAST1 (NM_001030318), BT_CAST2 (NM_174003), BT_CAST3 (NM_001030319) and BT_CAST4 (NM_001030320), with Thai native and Kampaengsaen breeds

Reference

1. Koohmaraie M. 1988. *Proc. Recip. Meat Conf.* 41:89.

2. Koohmaraie M. 1992. *Proc. Recip. Meat Conf.* 45:63.

3. Whipple G., *et al.* 1990. *J. Anim. Sci.* 68:2716.

4. Raynaud P., *et al.* 2005. *Arch. Biochem. Biophys.* 437:69-77.

Calpastatin Expression in Different Muscles of Thai Native Cattle

Jirajaroenrat, K.[1*], Opatpatanakit, Y.[2] and Sethakul, J.[1]

[1]Department of Animal Production, Faculty of Agricultural Technology, KMITL, Bangkok, Thailand 10520
[2]Department of Animal Technology, Faculty of Agricultural Production, Maejo University, Chiangmai, Thailand 50290
*e-mail: kjkanya@kmitl.ac.th

Introduction

Calpastatin is a specific protein inhibitor of the calcium dependent proteinases μ- and m- calpain[1-2]. This enzyme system is known to be involved in meat tenderization, which is especially important in beef quality[3]. Four alternatively spliced mRNA transcripts of calpastatin gene (Type I-IV) have been reported in European breed (*Bos taurus*) cattle. Three calpastatin isoforms migrated at 145, 125 and 70 kDa were detected[4]. Thai native cattle were identified to be a member of *Bos indicus* specie. Little molecular information on this specie has been reported.

Objective

To characterize the pattern of calpastatin expression in different muscles of Thai native beef cattle (*B. indicus*) in order to understand the relationship between calpastatin regulation and meat quality.

Method

Muscle Extraction: The protein were extracted from six muscles; semimembranosus (SM), rectus abdominis (RA), semitendinosus (ST), longissimus troracis (LT), longissimus dorsi (LD) and transversus abdominis (TA) according to the method described by Delgado *et al.* [5].

SDS-PAGE: Protein concentration in the supernatant fraction was diluted to 3 mg/mL. Protein amount of 50 μg were separated by SDS-PAGE using 7 x 10 cm minigels. After staining, the protein bands were visualized comparing to prestained molecular weight marker (Invitrogen).

Western blot analysis: The seperated proteins were transferred to Hybond-P PVDF membrane. The 1st antibody used was a monoclonal antibody (Anti-DomIV: Sigma) at 1:500 dilution The polypeptides labeled with the 1st antibody were detected by using an polyclonal anti-mouse IgG (Fab fragments; Roche) conjugated with horseradish peroxidase at a 50 mU/ml. The 2nd antibody was detected by using TMB solution (3,3',5,5'- tetramethyl-benzidine: Roche). The image of the membrane was captured using gel document system (Syngene).

Result

We previously isolated the calpastatin mRNA transcripts by RT-PCR from skeletal muscles of Thai native cattle. About 1,500 bp of 3'-termini cDNA was isolated. The sequences shared 98-99% to that of the European cattle.

The muscle extracts were separated by SDS-PAGE (Figure 1). The expression of calpastatin isoforms in six muscles analysed by Western blotting showed different patterns. The 125 kDa and 47 kDa isoforms corresponding to the translation of Type III transcript, the erythrocyte isoform, respectively, were detected in SM and RA muscles, whereas LT, LM and TA muscles contained only 47 kDa isoform. Interestingly, the ST muscle contained the 36 kDa isoform the C-terminal proteolytic product of the calpastatin catalyzed by calpain enzymes (Figure 2).
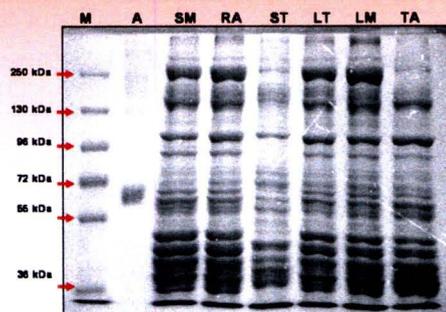

Figure 1. SDS-PAGE of protein extracts from 6 muscles of Thai native beef cattle: semimembranosus (SM), rectus abdominis (RA), semitendinosus (ST), longissimus troracis (LT), longissimus dorsi (LD) and transversus abdominis (TA) respectively. Lane M is a prestained protein molecular weight and lane A is bovine serume albumin 2 ug.
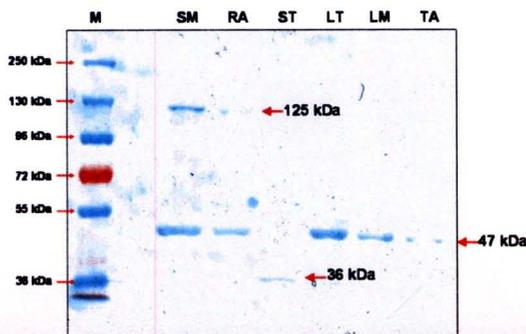

Figure 2. Western blotting of protein extracts from 6 muscles of Thai native beef cattle: semimembranosus (SM), rectus abdominis (RA), semitendinosus (ST), longissimus troracis (LT), longissimus dorsi (LD) and transversus abdominis (TA) respectively, against anti-calpastatin domain IV. Lane M is a standard protein molecular weight.

Conclusion

Different patterns of the calpastatin expression in different muscles were confirmed by Western blotting. Three calpastatin isoforms: 125, 47 and 36kDa bands were detected. Relationship of the calpastatin expression and its mRNA transcription will be further investigated.

Acknowledgement
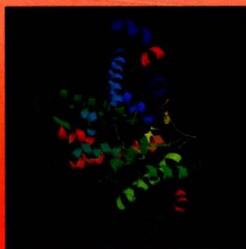
References

[1] Koohmaraie. 1988. Proc. Recip. Meat Conf. 41:89.

[2] Koohmaraie. 1992. Proc. Recip. Meat Conf. 45:63.

[3] Whipple et al. 1990. J. Anim. Sci. 68:2716.

[4] Raynaud et al. 2005. Arch. Biochem. Biophys. 437:69-77.

[5] Delgado et al. 2001. J. Anim. Sci. 79:398-412.