Green Materials and Technology

International Symposium on Green Materials and Technology (ISGMT)

> Edited by Dr. Subaer

TTP TRANS TECH PUBLICATIONS

DISTRIBUTION & ACCESS FOR PUBLICATION SUPPLEMENTS ABOUT US CONTACT US $\, \psi \, \chi$

LOG IN

Main Themes

Actual Problems of Engineering Mechanics Vol. 968

Green Materials and Technology Vol. 967

Functional Properties of Modern Materials II Vol. 966

Carbon Dioxide: Problems and Decisions II Vol. 965

Seminar on Materials Science and Technology Vol. 964 Home Materials Science Forum Green Materials and Technology

Green Materials and Technology

Volume 967 doi: <u>https://doi.org/10.4028/www.scientific.net/MSF.967</u>

Papers	Book								
mokosuli	1 2 3 4 5	>							
Paper Title		Page							
<u>Novel DNA Barcode Sequence</u> 59 <u>Discovery from Transcriptome of</u> <u>Acheta domesticus: A Partial</u> <u>Mitochondrial DNA</u>									

Green Materials and Technology p.2	
Authors: Yash Munnalal Gupta, Kittisak Buddhachat, Surin Peyachoknagul, Somjit Homchan Abstract: The potential of mitochondrial DNA (mtDNA) genes are well-known for	
<u>more</u> <u>Collection of Mitochondrial tRNA</u>	65
<u>Identification for Acheta</u> <u>domesticus</u> Authors: Yash Munnalal Gupta, Kittisak	
Abstract: The mitochondria are organelles found within eukaryotic cell, more	
<u>Philogenic Relationship of Wild</u> <u>Pigs and Local Pig from North</u> Sulawesi Based on the Growth	71
Hormone Gene (GH Gene) Authors: Alexius Mege Revolson, Yermia Semuel Mokosuli, Jacqueline Jochebed Rayer Debby, Ellen Hetie Adil, Christny Rompas, Nony Manampiring,	
Merry Montolalu Abstract: Growth hormone regulates reproduction and growth in mammals. A more	
	Green Materials and Technology p.2 Authors: Yash Munnalal Gupta, Kittisak Buddhachat, Surin Peyachoknagul, Somjit Homchan Abstract: The potential of mitochondrial DNA (mtDNA) genes are well-known formore Collection of Mitochondrial tRNA Sequences and Anticodon Identification for Acheta domesticus Authors: Yash Munnalal Gupta, Kittisak Buddhachat, Surin Peyachoknagul, Somjit Homchan Abstract: The mitochondria are organelles found within eukaryotic cell,more Philogenic Relationship of Wild Pigs and Local Pig from North Sulawesi Based on the Growth Hormone Gene (GH Gene) Authors: Alexius Mege Revolson, Yermia Semuel Mokosuli, Jacqueline Jochebed Rayer Debby, Ellen Hetie Adil, Christny Rompas, Nony Manampiring, Merry Montolalu Abstract: Growth hormone regulates reproduction and growth in mammals. Amore

Frontiers in Advanced Materials Vol. 956	Molecular Barcoding Based 16S rRNA Gene of Thermophilic Bacteria from Vulcanic Sites, Linow Lake, Tomohon Authors: Simanjuntak Suddin, Yermia Semuel Mokosuli, Welerubun Marcelina, Naharia Orbanus, Kapahang Ardi Abstract: Thermophilic bacteria live at temperatures above 45 ⁰ C. Many more	83
	Interaction of Physiology Character - Secondary Metabolic of <i>Fusarium oxysporum</i> on Tomatoes of Fusarium Wilt Symptom Authors: Abdul Azis Ambar, Henny Setyawati, Nur Ilmi Abstract: <i>Fusarium oxysporum (F.</i> <i>oxysporum</i>) is the pathogen that caused more	95
	<u>Consumption Value and Goat</u> <u>Livestock Weight Gain through</u> <u>Taiwanese Grass Feeding Using</u> <u>Morinda Bio-Activator Liquid</u> <u>Fertilizer</u> Authors: Rahmawati Semaun, Juliawati Rauf, Fitriani, Syamsuddin Hasan, Budiman Nohong	101

Abstract: Concentrate is required to be added for forage feeding in order to <u>...more</u>

Instant Ginger Beer, the Traditional Health Drinks from Buginese-Makassar Authors: Husain Halimah, Nur Raisah Maddeppungeng, Nurasmi Abstract: The present study aims to produce traditional health drinks based ...more

107

The Effect of Temperature and Duration Roasting of the Physical Characteristics of Arabica Coffee Authors: Bondaris Palungan Musa, Iradat Rapa Charnia, Salu Salma Abstract: This study aims to determine the effect of temperature and roasting more	113
Prevalence of Nematodes in Bali Cattle in Tamarunang Village, Gowa Regency, South Sulawesi Authors: Ahmad Hasyim, A. Bida Purnamasari, A. Dewi Rizka M. Ainulia, Muh. Rizaldi Trias Jaya N. Putra Abstract: The purpose of this research was to determine the prevalence of the more	118

The Used of Protease from Palado(Agave) Roots, and Palado Leaf inthe Making Process of VirginCoconut Oil (VCO)Authors: Sitti Rahmawati, Siti Nuryanti, KASMIR S. MaleAbstract: Virgin coconut oil (VCO) which is also known as coconut oil or virgin moreShowing 11 to 20 of 42 Paper Titles< 12345	123

DISTRIBUTION & ACCESS FOR PUBLICATION SUPPLEMENTS ABOUT US POLICY & ETHICS CONTACT US IMPRINT & PRIVACY POLICY SITEMAP

Scientific.Net is a registered brand of Trans Tech Publications Ltd © 2019 by Trans Tech Publications Ltd. All Rights Reserved

Preface

Dean's Greeting

Bismillahirrahmanirrahim. Assalamu alaikum warahmatullahi wabarakatuh Good Morning Ladies and Gentlemen.

I would like to take this opportunity to express my sincere appreciation to all keynote speakers and invited speakers for accepting our invitation to share their research findings and best practices here, in the 3rd ICMSTEA and 1st ISGMT. We do hope that the best practices in the field of Mathematics, Sciences, Technology, and Education can be well communicated among researchers and academics gathered in this conference. I would also like to take this opportunity to thank the Chair of ICMSTEA committee for organizing this special event to form a platform for networking as well as the exchange of lessons learnt and best practices.

Ladies and Gentlemen,

Implementation of the 3rd ICMSTEA and 1st ISGMT is not only expected to be a forum for disseminating the results of the latest findings in the field of each study, but also expected to be a networking forum for academics and researchers. We hope that the meeting of researchers and academics from various institutions in this place can be a starting point for future collaborations.

Behind the implementation of these activities, there are hard work, commitment and outstanding cooperation of the executive committee. Therefore, to the whole team of the executive committee, I really thank for your hard work and enthusiasm for the success of the 3rd ICMSTEA and 1st ISGMT.

Finally, as the Dean of the Faculty of Mathematics and Natural Sciences, Universitas Negeri Makassar, I want to congratulate you all for participating in ICSMTEA and ISGMT 2018. Welcome to the city of Makassar, hopefully this activity can bring benefit for all of us.

Wassalamualaikum Wr. Wb.

Remarks of the Chairman The 3rd ICMSTEA and the 1st ISGMT 2018

Assalamualaikum warahmatullah wabarakatuh.

Alhamdulillah, all praises belong only to *Allah Subhanahu Wa Ta'ala* which always give His grace and guidance so the 3rd ICMSTEA and the 1st ISGMT 2018 which organized by the Faculty of Mathematics and Natural Sciences, Universitas Negeri Makassar can be held. Greetings and blessings aimed to the Messenger of Allah, Prophet Muhammad and the entire family and friends.

As the chair of the committee of the 3rd ICMSTEA and the 1st ISGMT 2018, I would like to deliver my highest appreciation to the *Keynote Speakers*, Dato' Professor Dr. Zul Azhar Zahid Jamal, Dr. Saleh Abdurrahman, M.Sc, Assoc. Prof. Dr. Andriana Surleva and all the invited speakers who have been willing to spend their time to share their knowledge, perspectives, and best practices to all the participants of the 3rd ICMSTEA and the 1st ISGMT 2018.

The 3rd ICMSTEA and the 1st ISGMT 2018 aimed to provide the chances for researchers and academics to share and discuss the latest findings in their field of expertise, thus it will also provide the chance for students to gain more insight on the development of knowledge in the realm of science. Besides keynote speakers and invited speaker session, there will be a parallel session where researchers can share their latest findings. The parallel session will be divided into several groups based on the topic of their study, including Mathematics, Physics, Chemistry, Biology, Education, and related fields. The I do hope that the knowledge and experience of all the speakers who participated in this forum would be beneficial in supporting the dissemination of the latest research findings, thus strengthen the position of the field of study as one of the answers in solving problems in the community.

On behalf of the committee of the 3rd ICMSTEA and the 1st ISGMT 2018, I do apology for any inconvenience you may experience during your time in this event. Hopefully, all participants can enjoy the knowledge sharing of the conference as well as the atmosphere and the friendliness of Makassar.

May Allah bless our activities. Waalaikumussalam Warahmatullahi Wabarakatuh.

Chair of the committee,

Drs. Subaer, M.Phil., Ph.D.

Materials Science Forum ISSN: 1662-9752

Founding Editor Fred H. Wohlbier

Honorary Editor

Prof. Graeme E. Murch

SEND MESSAGE

University of Newcastle, PRC for Geotechnical Science and Engineering, School of Engineering; University Drive, Callaghan, Australia, NSW 2308;

Editor(s) in Chief

Prof. Iulian Antoniac

SEND MESSAGE

University Politehnica of Bucharest, Faculty of Materials Science and Engineering; 313 Splaiul Independentei, Bucharest, 060042, Romania;

Editorial Board

Prof. Dezső L. Beke

SEND MESSAGE

University of Debrecen, Department for Solid State Physics; Bem tér 18/b, Debrecen, 4026, Hungary;

Prof. Giorgio Benedek

SEND MESSAGE

University of Milano Bicocca, Department of Materials Science; U5, Universitá di Milano-Bicocca, Via R. Cozzi 55, Milano, 20125, Italy;

Prof. Anil K. Bhatnagar

SEND MESSAGE

University of Hyderabad, School of Physics and School of Engineering; Hyderabad, India, 500046;

Prof. Chi Ming Chan

SEND MESSAGE

Hong Kong University of Science and Technology, Department of Chemical and Biomolecular Engineering, Clear Water Bay; Kowloon, China;

Roberto B. Figueiredo

SEND MESSAGE

Federal University of Minas Gerais, Department of Metallurgical and Materials Engineering; Belo Horizonte, MG, 30, Brazil, 31270-901;

Prof. Hermann G. Grimmeiss

SEND MESSAGE

Lund University, Department of Solid State Physics; Box 118, Lund, 221 00, Sweden;

Prof. Jerzy Jedlinski

SEND MESSAGE

AGH University of Science and Technology, Faculty of Materials Science and Ceramics, Department of Physical Chemistry and Modeling of Processes and Surface Engineering; al. Mickiewicza 30, Kraków, 30-059, Poland;

Prof. Megumi Kawasaki

SEND MESSAGE

Oregon State University, School of Mechanical, Industrial and Manufacturing Engineering; 204 Rogers Hall, Corvallis, USA, 97331;

Prof. Pentti O. Kettunen

SEND MESSAGE

Tampere University of Technology, Department of Materials Science, Faculty of Automation, Energy and Materials Engineering; PO Box 589, Tampere, 33101, Finland;

Prof. Terence G. Langdon

SEND MESSAGE

University of Southampton, Faculty of Engineering and the Environment; Lanchester Building (Bldg. 7), Highfield Campus, Southampton, United Kingdom, SO17 1BJ;

Prof. Jai Sung Lee

SEND MESSAGE

Hanyang University, Department of Metallurgy and Materials Science; 55 Daehak-no, Sangnokgu, Ansan, Korea, South, 426-791;

Prof. Eric J. Mittemeijer

SEND MESSAGE

Max Planck Institute for Intelligent Systems; Heisenbergstrasse 3, Stuttgart, 70569, Germany;

Prof. Stephen J. Pearton

SEND MESSAGE

University of Florida, Department of Materials Science and Engineering; Gainesville, USA, 32611-6400;

Prof. Vassilis Pontikis

SEND MESSAGE

Commissariat à l'Energie Atomique et les Energies Alternatives (CEA), CEA-Saclay; Bdg. 524, Gif-sur-Yvette, 91191, France;

Prof. András Roósz

SEND MESSAGE

Hungarian Academy of Sciences, Miskolc University (HAS-MU); Miskolc-Egyetemváros, 3515, Hungary;

Prof. David N. Seidman

SEND MESSAGE

Northwestern University, Department Materials Science and Engineering; Cook Hall, 2220 Campus Drive, Evanston, USA, 60208;

Dr. Ching Hua Su

SEND MESSAGE

NASA/Marshall Space Flight Center, EM31 NASA/Marshall Space Flight Center; Huntsville, USA, 35812;

Prof. David Tomanek

SEND MESSAGE

Michigan State University, Physics and Astronomy Department; 567 Wilson Road, East Lansing, USA, MI 48824-6455;

Prof. A.S. Wronski

SEND MESSAGE

University of Bradford, School of Engineering, Design and Technology; West Yorkshire, Bradford, United Kingdom, BD7 1DP;

Prof. David J. Young

SEND MESSAGE

University of New South Wales, School of Materials Science and Engineering; Sydney, Australia, NSW 2052;

← → C					९ 🕁 🍮
Scopus Preview		Author sear	ch <u>Sources</u>	Help 🗸	Dr. Yermia Semuel Mokosuli, MSi 🗸
Source details					Feedback > Compare sources >
Materials Science Forum Scopus coverage years: from 1984 to 1986, from 1994 to Present Publisher: Trans Tech Publications Ltd			CiteScore 2018 0.33 Add CiteScore to y	your site	0
Subject area: (Harvis Steves Generi Menvis Steves) (Ergineering Medanial Ergineering) (Ergineering Medania of Menvish) (Physica and Astronomy Condensed Meter Physica) Viewall documents) Set document alest Journal Homesage	5JR 2018 0.173		0		
			SNIP 2018 0.299		٥
CiteScore CiteScore rank & trend CiteScore presets Scopus content coverage					
CiteScore 2018 Calculated using a	ata from 30 April, 2019	CiteScore rank 💿			
Citation Count 2018 2404 Citations >		Category			Rank Percentile
0.33 = * Documents 2015 - 2017* 7,206 Documents >		Materials Science General Materials Science			#345/439 💻 20th
*CtaScere indudes all en lable document types View CtaScere methodology	> CiteScore FAQ >	Engineering Mechanical Engineering			#462/579 💻 20th
CiteScoreTracker 2019 © Last updat	ed on <i>12 August, 2019</i> Updated monthly	Engineering Mechanics of Materials			#299/352 1 4th
$0.18 = \frac{4}{ $$ $$ Documents 2016 - 2018 $$ $$ $$ $$ $$ $$ $$ $$ $$ $$ $$ $$ $$$		View CiteScore trends >			
😵 Metrics displaying this icon are compiled according to Snowball Metrics 7, a collaboration between industry and academia.					





Philogenic Relationship of Wild Pigs and Local Pig from North Sulawesi Based on the Growth Hormone Gene (GH Gene)

REVOLSON Alexius Mege^{1,a*}, MOKOSULI Yermia Semuel^{2,3,b}, DEBBY Jacqueline Jochebed Rayer^{3,c}, ELLEN Hetie Adil^{3,d}, CHRISTNY Rompas^{3,e}, NONY Manampiring^{2,f} and MERRY Montolalu^{2,g}

^{1*}Professor of Animal Physiology, Department of Biology, Faculty of Mathematics and Science, State University of Manado, Tondano, Sulawesi Utara, Indonesia

²Laboratory of Bioactivity and Biomolecular, Dapartent of Biology State University of Manado, Tondano, Sulawesi Utara, Indonesia

³Departments of Biology, Faculty of Mathematics and Science, State University of Manado, Tondano, Sulawesi Utara, Indonesia

^{a*}ramege@unima.ac.id; ^byermiamokosuli@unima.ac.id, ^cdebbyrayer@unima.ac.id ^dellenhetieadil@unima.ac.id ^echritnyrompas@unima.ac.id ^fnonnymanampiring@gmail.com ^gmerrymontolalu@gmail.com

Keywords : growth hormone gen, local pigs, phylogeny tree, Sulawesi Utara

Abstract. Growth hormone regulates reproduction and growth in mammals. A study was conducted to obtain the characteristics of the GH gene, in local pigs in North Sulawesi. Pig samples were obtained from traditional farmers, from four districts in North Sulawesi. DNA extraction and purification, using pig pituitary tissue. Amplification of GH gene, performed by PCR method. Visualization of CO1 gene amplikon, performed by electrophoresis technique. Sequencing, conducted through the First BASE Singapore sequencing service. The results show that there is a variation of local pigs CO1 gene in North Sulawesi. Variations are also found in the amino acid sequence encoded by the GH gene. Knowledge of the characteristics of local pig gh gene, the basics of selection of local pigs superior to North Sulawesi.

Introduction

Local pigs are wild pigs, which have been domesticated for a long time and have experienced high adaptation to certain environments. Indonesia has five species of local pigs, from eight existing species in the world¹. Some areas that have long been breeding local pigs are Minahasa, North Tapanuli, Bali, Sumba, Toraja and Papua. Local pig farm, done conventionally. Local pigs are fed agricultural wastes, human food waste, and non-planted crops. Local pig farmers do not use factory-made feed, so the growth of pigs takes place naturally. This makes local pork taste more savory and with a higher unsaturated fatty acid composition than hybrid pigs².

Local Indonesian pigs, endangered, among others, due to breeders prefer to breed imported pigs and hybrid pigs, which produce more meat, making it more economically profitable. The population of some local Indonesian pigs is the local pig of Jawa (Sus verrucosus), Local pig of Kalimantan (Sus barbatus), local pig of Sulawesi (Sus celebensis) and Babirusa (Babyroussa babyrusa). The population of all local pigs, already in the vulnerable to endangered stages are almost threatened to perish^{3,4,5}

The need for pork in Asia is high, therefore the potential of Indonesia to be a pork exporter country is very large. In 2016, Indonesia's pig production is at 342,346 metric tons ⁶. Data from the Central Bureau of Statistics showed that live pig exports to Singapore in July 2016 reached the US \$ 4.58 million (Rp61.77 billion), up 11.61%, compared to June's value of U \$ 4.10 million (Rp55.3 billion). Pork consumers prefer local pigs compared to hybrid pigs. The productivity of local pigs is relatively smaller compared to imported pigs. However, it is necessary to make a superior local pig selection, based on both phenotypic and genotypical characters.

The productivity of local pigs, influenced by the process of reproduction, hormone production, and feed given⁷. Growth hormone (GH) is a hormone synthesized and secreted by the anterior

pituitary gland section. GH plays a role in regulating metabolism and growth in mammals⁸. GH genes provide the biological effects on individual growth after birth, the rate of growth and milk production^{9,10}. GH gene plays an important role in reproduction, embryogenesis, lactation, and growth of pigs^{7,11,12}. Injections of pregnant mare serum gonadotropin (PMSG) and human chorionic gonadotropin (hCG), increase production of growth hormone in ⁷. On the other hand, increased expression of growth hormone increases the performance and growth of pigs. The characteristics of the GH gene owned by local pigs are potentially developed into molecular markers, for superior local pig selection. A study was conducted to obtain superior local pigs in North Sulawesi based on the characteristics of the GH gene.

Materials and Methods Samples

Local pig samples obtained from several districts in North Sulawesi are northern minahasa, minahasa, mongondow and sanger balls (Figure 1). The pituitary gland is used as a tissue source, for DNA extraction. The organs of the pituitary gland are preserved in 70% ethanol, further down to the laboratory.



Figure 1. Location of local pigs sampling in Sulawesi island (map source : www.google.co.id/maps/place/Sulawesi/.com).



Figure 2. The Sulawesi local pigs, sampling location Langowan, Minahasa.

DNA Extraction, PCR Amplification and Sequencing

Total genomic DNA was extracted from pituaitary gland tissue using Genomic DNA Mini Kit (Tissue) according to the manufacturer's protocol. Amplification of GH gene using PCR method, was applied MyTaq HS Red Mix Bioline (Table 1 and Table 2).

PCR Component	Volume (µL)
2x MyTaq HS Red Mix Bioline	25
Primer Forward : : 5'-	
TGGTGTTTGGCACCTCAGAC-3'	1
(Miao Zhiguo et al. 2012)	
Primer Reverse : HCO2198 : 5'-	
CGTCATCACTGCGCAAGTTT-3'	1
(Miao Zhiguo et al. 2012)	
DNA babi lokal	2
ddH ₂ O	21
Total	50

Ta	ble 2. The Condition	s of PCR.	
Cycle	Time (Second)	Temperatur (°C)	Phase
25	60	94	Denaturation
	30	50	Annealing
33 X	30	72	Ekstension
	60	72	Final Ekstension

PCR products were purified using Wizard SV Gel and PCR Clean-Up System (Promega Corp). Purified PCR products were analyzed by electrophoresis in 1% agarose gel. The molecular size of the amplified products was estimated using 1 kbp DNA ladder (Biometra). PCR products were sequenced using AB1 PRISM Dye Terminator Cycle Sequencing Ready Reaction System, version 1.1. (Applied Biosystems) in FIRST BASE Singapura

Sequences Analyses and Phylogeny trees reconstructed

Obtained sequences were aligned using BioEdit, Geneious v11.0.3 and MEGA 6.0 software. Sequences were subjected to Basic Local Alignment Search Tool (BLAST) in order to perform sequence similarity searches (www.ncbi.nih.gov.com). Nucleotide frequencies were calculated using BioEdit software. Phylogenetic trees were reconstructed using MEGA 6.0 software.

Results and Discussion

Amplification of GH gene areas, local pigs from five districts in North Sulawesi were successful. PrimaryGH gene with forward:: 5'-TGGTGTTTTGGCACCTCAGAC-3 'and reverse 5'-CGTCATCACTGCGCAAGTTT-3' ¹¹, show clearly visible band patterns in the electrogram, electrophoresis results. The thickness of the band formed, indicating the number of GH amplicon genes, which were successfully amplified by the PCR method. Based on a 1 kb ladder DNA size as a comparison, it shows the length of the GH gene, from each local pig sample being in the range 300 bp - 400 bp (Figure 2). It can be concluded that extraction using local pig pituitary gland, successfully isolated total DNA well.



Figure 3. Visualization of amplicons of GH gene fragments, local pigs from Sulawesi.

GH gene sequencing results, visualized in a chromatogram using the Geneious v11.0.3 program. The mean HQ score was 89.3%, indicating the ouput quality of the GH gene sequence, local pigs from Sulawesi were excellent. GH forward and reverse sequence genes, aligned to obtain a consensus area. Alignment is done by Bioedit program in order to obtain an accurate sequences, to reduce the risk of errors in the results BLAST, and fault tree phylogeny reconstruction. Characteristics of the GH gene, local pigs of North Sulawesi, have been obtained based on sequence analysis. The length of the GH gene, local pigs are 351 bp (MGH), 438 bp (BGH), 356 bp (RGH), 432 bp (UGH) and 379 bp (SGH). The difference in the length of the GH consensus gene area has shown variations in the GH gene, local pigs in North Sulawesi (Table 3).

No	Comple	Sequences characteristics
1	Dahi Lakal Mirahaas	DNA molecule: MGH
1	(MGH)	Length = 351 base pairs Molecular Weight = 107819,00 Daltons, single stranded Molecular Weight = 214318,00 Daltons, double stranded G+C content = 65,24% A+T content = 34,76%
2	Babi Lokal Bolaang Mongondow	Nucleotide Number Mol% A 69 19,66 C 126 35,90 G 103 29,34 T 53 15,10 DNA molecule: BGH
-	Utara (BGH)	Length = 438 base pairs Molecular Weight = 133110,00 Daltons, single stranded Molecular Weight = 267180,00 Daltons, double stranded G+C content = 61,64% A+T content = 37,90%
		Nucleotide Number Mol% A 79 18,04 C 125 28,54 G 145 33,11 T 87 19,86 R 1 0,23 K 1 0,23
3	Babi Lokal Bolaang Mongondow (RGH)	DNA molecule: RGH Length = 356 base pairs Molecular Weight = 109258,00 Daltons, single stranded Molecular Weight = 217366,00 Daltons, double stranded G+C content = 65,17% A+T content = 34,83%
4	Babi Lokal Minahasa Utara (UGH)	Nucleotide Number Mol% A 69 19,38 C 126 35,39 G 106 29,78 T 55 15,45 DNA molecule: UGH Length = 432 base pairs Molecular Weight = 132068,00 Daltons, single stranded Molecular Weight = 263439,00 Daltons, double stranded G+C content = 59,72% A+T content = 38,66%
		Nucleotide Number Mol% A 85 19,68 C 137 31,71 G 121 28,01 T 82 18,98
5	Babi Lokal Sulawesi Tengah (SGH)	DNA molecule: SGH Length = 379 base pairs Molecular Weight = 115950,00 Daltons, single stranded Molecular Weight = 231042,00 Daltons, double stranded G+C content = 58,05% A+T content = 40,11%
		Nucleotide Number Mol% A 73 19,26 C 123 32,45 G 97 25,59 T 79 20,84

Alignment of the GH gene, local pig North Sulawesi, has shown many sites where there are differences in nucleotides. Alignment with the Multalin program, obtained the consensus of the GH gene, from five regions in Sulawesi (Figure 4).

•••••••••••••••••••••••••••••••••••••••	1	10	20	30	40	50	60	70	80	90	100	110	120	130
ConsensusMGH ConsensusRGH ConsensusUGH CConsensusSGH ConsensusBGH Consensus	CT CACCAG	ATCGTCAT(Atcgtcat(CACTGCGCA Cactgcgca Tt	AGTTTGTGGAAA AGTTTGTGTCAA GGTGTTTGGCAC ,gt,t,tggca,	ITTTTCATA Ittgtcgta Ittgacci Itcagacci	TTGF GATGCTTGF GGTTTGCTTGF GTTTTGCTTGF GCGTC-TACGF gttgcttGF	AGGA-CTGTC AGGA-CTGTC AGGATCTGTC AGGATCTGTC AGAACCTGAA AGgA.CTGLC	CTGCCCGGGG CTGCCCGGGG CTGCCCGGGG CTGCCCGGGG GGACCTGGAG ctgCCcGGgG	GCTGCCATCCT GCTGCCATCCT GCTATCCT GCTGCCATCCT GAGGGCAT GCtgatCcT	CCAGCTCCT(CCAGCTCCT(CCAGT-CT(CCAGCTCCT(CCAGCCCCT(CCAGCCCCT(CCAGcCT(GCAAAAGGGAG GCAAAAGGGAG GCAAAAGGGAG GCAAAAGGGAG GCAAAAGGGAG GCAaaaGGGGAG	GAGAGAGACA GAGAGAGACA GAGAGAGACA GAGAGAGA	GAGGCCGAI GAGGCCGAI GAGGACGA CAGAGC CTGCGGI caGCg.	GTCCTTGGC GTCCTTGGC -TCCTTGGC -TGAATTAC GTC gTctc
	131	140	150	160	170	180	190	200	210	220	230	240	250	260
ConsensusMGH ConsensusRGH ConsensusUGH CConsensusSGH ConsensusBGH Consensus	AGCGGC AGCGGC AGCGGC AGCGGC CCGC agCgGC	GCTCTCCT(GCTCTCCT(GCTCTCCT(-CTCTA-T(ACACTG-G(-CLCTL(ATCCCCTCC ATCCCCTCC ATCCCCTC- ATCCC	ATCTTCCACCCT GTCTTCCACCCT GTCTTCCACCCT -TCTTTCCACCA -CCTTTCCCACT -GCCGGCTCTCT .LCLL.C.C.CT	CCCCTCCA CCCCTCCA CCCCTCCA CCCCTCCA CCCCTCTG CCCGGCTG CCCGGCTG CCC_LCLg	FGCTTGGAGAA FGCTTGGAGAA FGCTTGGAGAA FGCTGGAGAA FGCGAGAAC FGCGGAGC FGCGGAGG	AGGACAAAGA AGGACAAAGA AGGACAAAGA GAGACAAAGA GAGACAAAGA GAGGCGAGA GGTGGGGGGGA gggacaaaGA	GGGAAGACTC GGGAAGACTC GGGAAGACTC GGAAACTC CGCACGTG gG.Aag.cTc	CGCAGGGGGAA CGCAGGGGGGAA CGCAGGGGGGAA C-CAGGGGGGAA CGCTGGGGGGAA CgCaGGGGGAA	ATGAGGAAAA Atgaggaaa Atgaggaaa Atgaggaaa GagaAa GagaAa Tgggtccca Gaaa	AGATTICTCCG AGATTICTCCG AGATTICTCCG AGATTICCCCG AGTTITCCCCG AGTCTCTCTCG Ag, TLTCLCcG	GGTCGAGAG GGTCGAGAG GGTCGAGAG GGTCGAGAG GGTGAG GTAGCAGTTC igcgag	TGAACTGC TGAACTGC TGAACTGC TGATGC TGATGC HCTCTCGA tgatGcl	TACAGAGAGA TACAGAGAGAG TACAGAGAGAG TACAGAGAGAG TACAGAGAGAG CCCGGAGAA LaCaGAGAGAg
	261	270	280	290	300	310	320	330	340	350	360	370	380	390
ConsensusMGH ConsensusRGH ConsensusUGH CConsensusSGH ConsensusBGH ConsensusBGH	AGCATG AGCATG AGCATG AGCATG AGCATG ATCTTT AgCaTg	GGACCCCCT GGACCCTCT GGACCCTCT GGACCCTCT GGACCCTCT TCCTCATTT gg.acCcTcT	TCCCCCAGC TCCCCCAGC TCCCCCAGC TCCCCCAGC TCCCCCAGC TCCCCCTGC TCCCCCAgC	CCACGTGCGT CCACGTGCGT CCACGTGCGT CCTGGT GGAGTCTTCCCT cctcgT.c.t	CCCCCCAC CCCCCCAC CCCCCCAC CCCCCCAC CTTTGTCC CCTTTGTCC	CGCTCCGCTCF CGCTCCGCTCF CGCTCCGCTCF CGCTCCTCC CGCTCCTCC TTCTCCAAGCF cgCTCCLCa	RGCCGGGAGA RGCCGGGAGA RGCCGGGAGA GACCCGGAGA GACCCGGAGA Atggagga a,cc,ggaga	GAGC GAGC GAGC GGGTGGAAGA GGGTGGAAGA GAgc	CGGCATGGGCC CGGCATGGGCC CGGAATGGGCC CGGAGGCC TGGAGGGGGCACA CGGagGgCc	CCAGTGTGCC CCAGTGTGCC CCAGTGTGCC CCGGGGCG- IGGAGAGCGCC IGGAGAGCGCC ICCGG,GCGC	GGGACCCGCAG GGGACCCGCAG GGGACCCGCAG ACCC CGCTGCCAAGG ,g,acCC,g	iCGCCAT iCGCCAT iCGCC-T iCGCCCT iACTCGGCCT gCGcCcT	C C C C TGTCTCT C	CCCACCCGC CCCACCCGC CCCACCCGC CCCACCCGC CCCACCC CTCTCCCTT CccaCCC
	391	400	410	420	430	440	450	460	470	48482				
ConsensusHGH ConsensusRGH ConsensusUGH CConsensusSGH ConsensusBGH Consensus	AT-CAG AT-CAG AT-CAG -T-CAG TTGCAG .T.CAG	GGCCTGGA GGCCTGGA GGCCTGGA GGCCTGGA GAGCTGGA GGCCTGGA GgccTGgA	TGCCCT TGCCCT TGCCCT GATGGCAG	CCTCCAGTCCT CCTCCAGTCCT CCTCCAGGTCTT CCTCCAGTGCTT CCCCCGGGCAGG CCLCCAGgcct	IGC-TTCTCI TCATTTTC ACAGATCC .cat.cl	STAGACGCGG Icatacacgc Icaagcaaacc Icaagcaaacc	-TCTGAGGTG -TCTGAGATG CTACGACAAA ,t.,ga.a.,	CCAAACACCA Tg TTTGACACAA tacac.a	ATTTT ACTTGCGCAGT a.tt	GATGACGA				

Figure 4. Align the GH gene with the Multalin program. (http://multalin.toulouse.inra.fr/multalin/cgi-bin/multalin.pl).

The dots indicate the same amino acid encoded by all the GH gene from Sulawesi. If any sequence has a difference in amino acids, then the amino acid name symbol is written. Based on translations, amino acids encoded by the GH gene, have shown many differences. Sites where transition and transversion mutations occur in the GH gene sequence (Figure 5) have affected the type of amino acid translation. Thus, there has been a variety of amino acids encoded by the GH gene, local pigs in Sulawesi.

72	1 10 CACCMKATCG His Xaa lle	20 TCATCACTGC Val lie Thr Ala	24 30 GCAAGTTTGT GI1 Val Cys	40 GTCAAATTTG Val Lys Phe	50 TCGTAGGTTT Val Val Gly Le	60 GCTTGAGGAT Leu Gu Asp	70 CTGTCCTGCC Leu Ser Cys	80 CGGGGGGCTGC Pro Gly Ala Ala	90 CATCCTCCAG Ile Leu Gin	10 CTCCTGCAAA Leu Leu Gin) 110 Agggagagag Lys Gly Igiul Ar	AGACAGAGAG	CCGAGTCCTT Pro Ser Pro	140 Gocagegee Trp Gin Arg Arg	150 CTCTCCTGTC Ser Pro Val	160 CCCTCCRTCT Pro Ser
ConsensusUGH	ст			·G-····T Gly- Asn - Phe	···A·····	Ala + Gly	Ser Val Leu Pro	Gly Gly	-Tyr Pro Pro	-Val - Cys Lys	Arg Glu Arg	Gui Thr Glui - II	A····· Asp Asp ·			Ser Ser
ConsensusSGH	Gin			- <mark>Asn</mark> - Cys	Arg Ser Phe	Ala 🛨 Gly	Ser Val Leu Pro	Gly Gly Cys	His Pro Pro	Ala Pro Ala Ly:	Gly Arg Glu	Arg His Arg	Ala Gu	TA·····-	Pro Leu Cys	Pro Ser
ConsensusRGH					GA ·	·····	Cys Pro Ala	Arg Gly Leu	Pro Ser Ser Ser	ser Cys Lys	Arg Gu Arg	GU Thr GU - II	Ala Glu Ser Le	Ala Ala Ala	Leu Ser Cys	Pro Leu Arg Leu
ConsensusMGH							Cvs Pro Ala	Are Giv Leu	Pro Ser Ser Ser	Ser Cvs Lvs	Are Glu Are	GU Thr GU - I	Ala Glu Ser Le	Ala Ala Ala	Leu Ser Cvs	Pro Leu His Leu
ConsensusBGH				· · ·								· · -Gl)	Arg Val Leu	Gly Ser Gly	Ala <mark>Leu Leu</mark> Ser	Pro Pro Ser
J2	TCCACCCTCC Phe His Pro Pro	CCTCCATGCT	19 TGGAGAAGAG Trp Arg Arg	0 200 ACAAAGAGGG Aspillys Gloi Gly	210 AAGACTCCGC Arg Leu Arg	220 AGGGGGAAAT Arg Gly Lys	GAGGAAAAGA	240 TTTCTCCGGG Phe Ser Gly	D 250 TCGAGAGTGA Ser Arg Val) 26 ACTGCTACAG Asn Cys Tyr Ar) 270 AGAGAGCATG g Glu Ser Met	280 GGACCCTCTC Gly Pro Ser) 290 CCCCAGCCCA Pro Pro Ala His	300 CGTGCGTCCC Val Arg Pro	310 CCCACCGCTC Pro Thr Ala	320 CGCTCAGCCG Pro Leu Ser Arg
ConsensusUGH	Ser Thr Leu	Pro Ser Met Le	Gly Closed	y Gin Arg Gly	Lys Thr Pro	G <mark>in Giy Giu M</mark> è	t Arg Lys Arg	Phe Leu Arg	Val Glu Ser Glu	Leu Leu Gin	Arg Glu His	Gly Thr Leu Ser	Pro Ser Pro	Arg Ala Ser	P <mark>ro His Arg</mark> Ser	Ala Gin Pro
ConsensusSGH	T.C.A Phe Pro Leu-	Pro - + + +			- A		AT	•••••C••••				- Asp Pro Leu	A·····C···- Thr His Pro -	Trp Ser	Pro Pro Leu	GA··C
ConsensusRGH	Pro Pro Ser	Pro Pro Cys	Leu Glu Lvs -													
ConsensusMGH	Pro Pro Ser	Pro Pro Cus	Au Glu Ive -								······	·····C···	·····			
ConsensusBGH	Ser Thr Leu	Pro Ser Met Le	Gly Glu -Gly	Gin Arg Giy	Lys Thr Pro	G <mark>in Giy Giu M</mark> é	t Arg Lys Arg	Phe Leu Arg	Val Glu Ser Glu	ı Leu Leu Gin	Arg Gu Hs	Gly Thr His Ser	Pro Ser Pro	Arg Ala Ser	P <mark>ro His Arg</mark> Ser	Ala Gin Pro
JS	330 GGAGAGAGCC GOL Arg Ala	0 340 GGCATGGGCC Gly Met Gly	35 CCAGTGTGCG Pro Ser Val An	0 360 GGACCCGCAG g Aspi Pro Gin	370 CGCCATCCCC Arg His Pro	380 ACCCGCATCA His Pro His Gir	GGGCCTGGAT	GCCCTCCTCC Ala Leu Leu) 410 AGTYCTTTCA Gin SVIII Phe Gin) 42 GCTTCTCGTA Leu Leu Val	GACGCGGTCT ASPLAIA Val	440 GAGGTGCCAA Giy Ala Lys	ACACCAATTT His Gin Phe	451 T		
ConsensusUGH	Gly Glu Ser	Arg Asn Gly Pro	Gin Cys Ala	Gly Pro Ala	Ala Pro-				• GT • • • • •							
ConsensusSGH		Val -Ala	Pro Gig	C	····C·-···	Thr teu	Are Ala -	Cvs	Cvs His	TT····A··		Are Cvs				
ConsensusRGH								·····	· · · C · ·			-0/-				
ConsensusMGH									····C··							
ConsensusBGH	Gly Gly Ser	Are His Gly Pro	Gin Cvs Ala	Giv Pro Ala	Ala Pro Ser Pro	Pro Ala Ser	Gly Pro Gly	Cvs Pro Pro Pro	GIV Pro Ser	Ala Ser Are	Are Are Gly Le	Arg Cvs Gin	Thr Pro			

Figure 5. Comparison of amino acid composition translated by GH gene, local pigs in North Sulawesi

The results of basic local alignment searching tools, on the NCBI website, showed the GH gene, local pigs from Minahasa, Bolaang Mongondow Utara, and Sulawesi Tengah, have the closest similarity, with *Sus scrofa* [FJ766018.1], (99%). On the other hand, the GH gene, local pig Bolaang Mongondow and Minahasa Utara, has the closest similarity to *Sus scrofa* [EU684439.1], (99%). Compared with previous research, that local pigs of Sulawesi, have species names, *Sus celebensis*^{4,5}, it was confirmed BLAST results on the NCBI website, July 4, 2018, no data was found on the gene bank NCBI, similarity of GH gene with *Sus celebensis* (Table 4).

No	Samples	Similarity Sequence	% Similarity	Location
1	MGH	<i>Sus scrofa</i> isolate GHp08TW growth hormone (pGH) gene, complete cds GenBank: FJ766018.1	99 %	Sichuan 625014, China
2	BGH	<i>Sus scrofa</i> isolate GHp06FO growth hormone (pGH) gene, complete cds GenBank: FJ766017.1	99 %	Sichuan 625014, China
3	SGH	<i>Sus scrofa</i> isolate GHp06FO growth hormone (pGH) gene, complete cds GenBank: FJ766017.1	99 %	Sichuan 625014, China
4	RGH	<i>Sus scrofa</i> haplotype H110 growth hormone gene, complete cds GenBank: EU684439.1	99%	East Asia
5	UGH	<i>Sus scrofa</i> haplotype H110 growth hormone gene, complete cds GenBank: EU684439.1	99%	East Asia

Table 4. GH gene sequence, which has the closest similarity, BLAST results on the NCBI website.

The reconstruction of the phylogeny tree, based on the GH gene sequence, has been done to obtain the status of species, and the evolutionary relationships, local pigs in North Sulawesi. Phylogeny construction is carried out with 24 sequences of the bank gene, BLAST results, which have a similarity of 99%. Phylogeny tree construction is done with two models, namely: Neighbor-Joining and Minimum Evolution. In both models, the same phylogenetic tree topography is produced. This reinforces the construction of phylogeny trees that have been formed. The phylogenetic tree forms two monophyletic groups. The first group is the GH gene sequence, local Sulawesi pigs namely: samples from Bolaang Mongondow, Sulawesi Tengah, Minahasa Utara and Minahasa. The second monophyletic group was 24 BLAST-related sequences and samples from Bolaang Mongondow Utara (Figure 6).



a.



Figure 6. The result of reconstruction of phylogeny tree, local Sulawesi pig, based on GH gene, with 24 sequences of BLAST results. The phylogeny tree is built with MEGA 6.0; bootstrap 1000 x. (a). Neighbor Joining Model (b). Minimum Evolution Model.

This phylogenetic tree explains that based on the GH gene, local Sulawesi pigs originate from a common ancestor, except for local pigs from Bolaang Mongondow Utara. However, they form the same monophyletic group, but no one forms a node. Therefore, confirmed the variation of the GH gene, local pigs in North Sulawesi. From the phylogenetic tree formed, local pigs from Bolaang Mongondow are the oldest local pig, based on the evolutionary history of the GH gene. Molecular barcoding using the CO1 gene, also cannot confirm the status of local pig species in Minahasa, North Sulawesi¹³.

GH genes play a role in the growth and health of mammals^{14.} Growth hormone plays a role in postnatal growth regulation, stimulation of anabolic processes such as bone growth and protein synthesis¹⁵. The GH gene affects milk production, fat content and cow's milk protein¹⁶. With the vital role of the GH gene, the molecular characteristics of the GH gene greatly affect the growth and development of mammal animals. GH gene research, as a molecular marker for superior livestock selection, has been widely practiced on Cattle. GH gene is recommended by many researchers, as a

molecular marker in the selection of superior cows ^{17, 18,19}. Variations in the GH gene have been used as the basis for genetic selection of Balinese cattle ²⁰. Based on the results of this study, the proposed GH gene, potentially developed as a molecular marker for superior local pig selection.

Conclusion

GH gen variation has been found in local pigs of North Sulawesi. GH gene variation, affect the amino acid composition encoded. The GH gene, potentially developed as a molecular marker, for superior local pig selection.

References

[1]. M.F. Rothschild, A. Ruvinsky, G. Larson, J. Gongora, T, Cucchi, K. Dobney, L. Andersson, G. Plastow, F.W. Nicholas, and C. Moran. 2011. The genetics of the pig. 2nd ed. Rothschild MF, Ruvinsky A, editors. London: CAB International.

[2]. E.H. Adil, R.A.Mege and Y.S. Mokosuli. Characteristics of fatty acids, in local minahasa pigs, north Sulawesi. International Journal of Veterinary Sciences and Animal Husbandry 2018; 3(3): 04-07

[3]. K. Kawanishi, M. Gumal, W. Oliver. 2008. *Sus barbatus*. The IUCN red list of threatened species. Version 2014.2. IUCN Global Species Programme Red List Unit [Internet]. [cited 24 July 2018]. Available from: http://www.iucnredlist.org/details/41772/0

[4]. J. Burton, A.A. Macdonald. 2008. *Sus celebensis*. The IUCN red list of threatened species. Version 2014.2. IUCN Global Species Programme Red List Unit [Internet]. [cited 24 July 2018]. Available from: http://www.iucnredlist.org/details /41773/0

[5]. A. A. Macdonald, J. Burton, and K. Leus. 2008. *Babyrousa babyrussa*. The IUCN red list of threatened species. Version 2014.2. IUCN Global Species Programme Red List Unit.

[6]. Badan Pusat Statistik (BPS). Sulawesi Utara.

[7]. N. Manampiring, C. Sumantri, G. Maheswari and W. Manalu. Expression of Growth Hormone Gene in the Pituitary of Piglets Born to Gilts Injected with Pregnant Mare Serum Gonadotropin and human Chorionic Gonadotropin Prior to Mating *International Journal of Sciences: Basic and Applied Research (IJSBAR) (2016) Volume 30, No 4, pp 446-455*

[8]. R.A. Curi, D.A. Palmieri, L. Suguisawa, H.N. de Oliveira, A.C. Silveira, and C.R. Lopes. 2006. Growth and carcass traits associated with GH1/*Alu* I and POU1F1/*Hinf* I gene polymorphisms in Zebu and crossbred beef cattle. Genet Mol Biol 29 (1) (2006): 56-61.

[9]. A. Dybus. Associations between Leu/Val polymorphismof growth hormone gene and milk production traits in Black-and White Cattle. *Arch. Tierz.*, *Dummerstorf* 45(5) (2002): 421–428.

[10]. W. Ge, M.E. Davis, H.C. Hines, K.M. Irvin, and R.C. M. Simmen. Association of Single Nucleotide Polymorphism in the Growth Hormone and Growth Hormone Receptor Genes with Blood serum Insulin-like growth Factor I Concentration and Growth Trais in Angus Cattle. *J. Anim. Sci.* 81 (2003): 641–648.

[11]. A. M. Murphy, K.G. Meade, P.A. Hayes, S.D. Park, A.C. Evans, P. Lonergan and D.E. Machugh. "Transmission ratio distortion at the growth hormone gene (GH1) in bovine preimplantation embryos: An in vitro culture-induced phenomenon?". Mol Reprod Dev, 75: 715-722, (2008).

[12]. H.I. Yardibi, F. Hosturk, G. Paya, G. Kaygisiz, A. Ciftioglu, A. Mengi, and K. Oztabak. "Association of growth hormone gene polymorphisms with milk production traits in South Anatolian and East Anatolian Red Cattle". J Anim Vet Advan, 8:1040–1044, (2009) [13]. R. A. Mege and Y.S. Mokosuli. DNA Barcoding of local pigs in minahasa, north sulawesi. International Journal of Fauna and Biological Studies (2017); 4(5): 82-87.

[14]. A. Sutarno, A. Junaidi, T. Baharudin. "DNA polymorphism at locus-2 growth hormone gene of Madura cattle". Biodiversitas, 6:77-81, (2005).

[15]. L. G. G.Silveira, L. R. Furlan, R. A. Curi, A. L. J. Ferraz, M. M. de Alencar, L. C. A. Regitano, C. L. Martins, M. de Beni, Arrigoni, L. Suguisawa, A. C. Silveira and H. N. De Oliveira. Growth hormone 1 gene (GH1) polymorphisms as possible markers of the production potential of beef cattle using the Brazilian Canchim breed as a model. Genet. Mol. Biol. 3 (2008) 1: 874-879.

[16]. T. Hartatik, D. Kurniawati and Adiarto. Associations between polymorphism of growth hormone gene With milk production, fat and protein content in friesian Holstein cattle. Journal of the Indonesian Tropical Animal Agriculture (J. Indonesian Trop. Anim. Agric.) 40(3):133-137, (2015).

[17]. H. Nugroho, W. Busono, and S. Soejosopoetro. 2013. Peningkatan performans sapi bali melalui seleksi berbasis marka gen growth hormone dan *miostatin* pada *village breeding centre* untuk menghasilkan bibit berkualitas. Laporan Penelitian Unggulan Perguruan Tinggi. Universitas Brawijaya, Malang.

[18]. Jakaria, D. Duryadi, R.R. Noor, B. Tappa, & H. Martojo. Evaluasi Keragaman Genetik Gen Hormon Pertumbuhan (GH) padaSapi Pesisir Sumatera Barat Menggunakan Penciri PCR-RFLP. Media Peternakan, Vol. 30. No. 1. April 2007, hlm. 1-10

[19]. S. Rahayu, S. B. Sumitro, T. Susilawati, dan Soemarno. 2006. Identifikasi Polimorfisme gen GH (*growth hormone*) Sapi Bali Dengan Metode PCR-RFLP. *Berk. Penel. Hayati: 12 (7–11), 2006.*

[20]. P. P. Agung, S. Anwar, W.P.B. Putra, dan S. Syahruddin. Keragaman gen *Growth Hormone* (GH) pada beberapa rumpun sapi lokal Indonesia. Pros Sem Nas Masy Biodiv Indon. Volume 3. No. 3. Desember 2017).