

ISAG . 2019

37th International Society for
Animal Genetics Conference

July 7 - 12, 2019 - Lleida, Spain

**Applied Genetics and Genomics In Other Species of Economic Interest
Workshop**

Organised by a standing committee: YES

Meeting information

Date: July 8th 2019

Time: 2:30 – 6:00 pm

Number of participants: 53

Chair

Name: Leanne van de Goor

Affiliation: VHLGenetics

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Agenda

- Welcoming Remarks
- Pig Comparison test Discussion
- Dromedary Comparison test Discussion
- Alpaca/Llama Comparison test Discussion
- Pigeon Comparison test Discussion
- Election of Committee
- Oral presentations
- Close

Summary of the meeting

1. Welcoming Remarks

This was the third workshop with the name Applied Genetics and Genomics In other Species of Economic Interest. Comparison tests for Pig, Dromedary, Alpaca/Llama and Pigeon have been carried out.

2. Pig Comparison test Discussion

The Duty lab was the "Laboratorio di Biotecnologie Genetiche" – University of Perugia, Department of Agricultural, Food and Environmental Sciences (Italy). Thirty-one (31) labs requested samples. Twenty-seven labs reported results. Twenty-one samples (including 1 reference) were submitted to all participants. Only two labs requested a second samples shipment. Twenty-seven labs reported results for some or all markers of the Core Panel (15 markers: S0005, S0090, S0101, S0155, S0227, S0228, S0355, S0386, SW24, SW240, SW72, SW857, SW911, SW936, SW951) and the Additional Panel (7 markers: IGF1, S0002, S0026, S0215, S0225, S0226, SW632). Only markers from the Core Panel were included in the ranking system. Clerical errors such as 162/162 instead of 162/ or the genotype 210/200 instead of 200/210 were not considered as mistakes for the ranking system. However, labs need to pay attention to such nomenclature issues when reporting their results as it is a lot of extra work to correct for such mistakes.

Due to the unavailability of the final version of the compiled results from all the markers and the labs involved in this CT the performance data have to be considered as provisional.

The overall marker concordance among labs was good, ranging from a minimum of 92.39% (S0386) to a maximum of 100.0% (S0090); in this performance value is not included the locus S0101 that showed a lot of problems among the labs. Marker S0101 showed a high discrepancy among the labs about the allele 217 (genotyped as 218 in most of the labs). After the discussion, taking into account that this allele is reported for the first time, the workshop participants proposed that both genotypes 218 and 217 are correct (no votes against).

Marker S0386 showed discrepancy between the labs about the allele 177: after the discussion the workshop participants highlighted that the correct genotype for samples 04, 06, 14 and 20 is 167/177, 169/177, 169/177, and 169/177 respectively. This discrepancy was already discussed in previous CT rounds (and more precisely in 2014 and 2017): the low concordance for this marker could be explained for using primers unable to amplify this allele in some samples. It is recommendable to remind again using the correct primer sequences as proposed in DNA microsatellite analysis for parentage control in Austrian pigs. Nechtelberger D, Kaltwasser C, Stur I, Meyer JN, Brem G, Mueller M, Mueller S., Anim Biotechnol. 2001. Nov; 12(2):141-4. PMID: 11808629. The recommended primers are: Fw: 5'-GAA CTC CTG GGT CTT ATT TTC TA, Rv: 5'-GTC AAA AAT CTT TTT ATC TCC AAC AGT AT. Since this situation has previously arisen, missing this allele was considered a mistake in the final ranking system.

Marker SW24 showed discrepancy among labs for the sample 05: the lowest allele (104) was missed by several labs. The proposal after the workshop discussion was not to consider missing this allele as a mistake in calculations for the ranking (no votes against).

Not all participants answered the included parentage questions correctly, three labs failed.

3. Dromedary Comparison test Discussion

The Duty lab was Qatar Genetic Lab from Qatar (responsible: Dr. Hanaa Ahmed). Six of seven labs (2 Qatar, 1 USA, 1 Germany, 1 New Zealand and 1 Russia) that requested samples, reported results. Twenty samples plus 2 reference samples were submitted to all participants. All labs reported results for the Core Panel (LCA19, LCA37, LCA56, LCA65, LCA66, LCA8, YWLL29, YWLL44) and 4 of them for the Back Up



Panel (LCA24, LCA77, LCA99, LGU49, VOLP3, VOLP32, VOLP59, YWLL08, YWLL36). Some Labs reported up to 24 additional markers. Only the markers in the Core Panel were included in the ranking system. In the Draft compilation sent out by FASS the results were compiled exactly as reported by the labs. The overall marker concordance among labs was excellent (100%) for 6 of the 8 markers in the Core Panel. Markers with lower concordance were: YWLL44 (98%) because of the presence of a null allele (106) not detected by one of the labs, and LCA56 (99%). There were some clerical errors, not considered for the final score. Two parentage questions were included in the CT, and for both of them, exclusion was not detected with the Core Panel, while 1 marker from the Back Up showed discrepancy for case 1. Low exclusion power of the current panels lies on the absence of polymorphism with LCA19 (Core Panel) and LCA24, LCA77, VOLP59 and YWLL36 (Back Up) being mono allelic markers. Changes in the Current Panels are required with previous consensus among labs doing dromedary parentage testing.

4. Alpaca/Llama Comparison test Discussion

The Duty lab was the Veterinary Genetics Laboratory from UC Davis (Cecilia Penedo as organizer). Eight of nine labs that requested samples, reported results. Twenty samples plus 4 reference alpaca samples (llama was not available) were submitted to all participants. All labs reported results for the Core Panel (LCA19, LCA37, LCA5, LCA56, LCA65, LCA66, LCA8, LCA94, LCA99, LGU49, LGU50, YWLL29, WLL40, YWLL44) while additional markers were reported for several labs. Only the markers in the Core panel were included in the ranking system. In the Draft compilation sent out by FASS the results were compiled exactly as reported by the labs. The overall marker concordance among labs was very good (99,71%) for 11 of the 14 markers in the Core Panel. Markers with lower concordance were: LCA37 (96,88%) because to incorrectly named or missed alleles, LCA66 (95,63%) because to 2 labs with off-ladder alleles, otherwise the concordance was excellent, and LGU50 (93,75%) because 1 lab was unable to detect one allele (null allele, 184). There were some clerical errors, not considered for the final score. Two parentage questions were included in the CT, with good concordance among labs.

5. Pigeon Comparison test Discussion

The Duty lab was VHL from the Netherlands. Ten labs (Europe: 7 and Africa: 3) requested samples. Twenty samples and one reference sample were submitted to all participants. Seven labs reported results. Labs reported results for the 12 STR markers in the Core Panel (Cl μ D11, Cl μ T43, Cl μ D01, PIGN57, Cl μ T13, Cl μ D16, Cl μ D19, Cl μ T02, Cl μ D17, Cl μ D35, Cl μ T17, PIGN4) and the 4 STR markers in the Backup Panel (PIGN15, PIGN10, PIGN26, PIGN12). Only the markers in the Core panel were included in the ranking system. In the Draft compilation sent out by FASS the results were compiled exactly as reported by the labs. The overall marker concordance among labs was good (>98%) for nine of the twelve markers in the core panel. Only three markers showed a lower concordance: Cl μ D16 (95%) because of the presence of an extreme allele (130) and miscalling of this allele by three labs. Cl μ T17 (95%) because one lab called two alleles (237 and 245) incorrectly and PIGN4 because one lab did not detect a few large alleles (297, 305 and 309).

6. Oral Presentations

- Invited Workshop Presentation: American mink genome sequencing project. Y. Miar, Dalhousie University, Canada (presentation was postponed to a later point of time).
- Development of a new multiplex system for analysis of 16 microsatellite markers of reindeer (*Rangifer tarandus*). V.A. Orekhov, GORDIX Ltd, Russia.
- Association analysis of coat color in blue wildebeest. R. van Deventer, Unistel Medical Laboratories, South Africa.



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- Estimation of genomic breed composition of individual animals in composite beef cattle. Z. Li, Neogen Geneseek, USA.

Committee chair (the new chair)

Chair: Leanne van de Goor
Term of service: 2017-2021
Affiliation: VHLGenetics
E-mail address: Lgo@vhladmin.nl

Committee members (the new committee)

Other members	Term of service	E mail address
Amparo Martinez	2017-2021	amparomartinezuco@gmail.com
Ntanganedzeni Mapholi	2017-2021	maphon@unisa.ac.za
Younes Miar	2017-2021	miar@dal.ca
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Foluke Eunice Sola-Oja	2019-2023	solaojo.fe@unilorin.edu.ng

COMPARISON TEST (2018-2019) YES

Duty laboratory Pig

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Duty laboratory Dromedary

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Duty laboratory Alpaca/Llama

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Duty laboratory Pigeon

Contact person: Leanne van de Goor
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Comments (issues rising)

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List of recommended markers with primer information

Dromedary:

ISAG STR Core Panel - Dromedary

Locus	Forward	Reverse
LCA8	GCTGAACCACAATGCAAAGA	AATGCAGATGTGCCTCAGTT
LCA37	AAACCTAATTACCTCCCCCA	CCATGTAGTTGCAGGACACG
LCA56	ATGGTGTTTACAGGGCGTTG	GCATTACTGAAAAGCCCAGG
LCA65	TTTTTCCCCTGTGGTTGAAT	AACTCAGCTGTTGTCAGGGG
LCA66	GTGCAGCGTCCAAATAGTCA	CCAGCATCGTCCAGTATTCA
YWLL29	GAAGGCAGGAGAAAAGGTAG	CAGAGGCTTAATAACTTGCG
YWLL44	CTCAACAATGCTAGACCTTGG	GAGAACACAGGCTGGTGAATA

ISAG Additional Markers - Dromedary

Locus	Forward	Reverse
CVLR01	GAAGAGGTTGGGGCACTAC	CAGGCAGATATCCATTGAA
CVLR04	CCCTACCTCTGGACTTTG	CCTTTTGGGTATTTTCAG
CVLR05	CCTTGGACCTCCTTGCTCTG	GCCACTGGTCCCTGTCATT
LCA99	CAGGTATCAGGAGACGGGCT	AGCATTTATCAAGGAACACCAGC
LGU49	TCTAGGTCCATCCCTGTTGC	GTGCTGGAATAGTGCCCAGT
VOLP3	AGACGGTTGGGAAGGTGGTA	CGACAGCAAGGCACAGGA
VOLP32	GTGATCGGAATGGCTTGAAG	CAGCGAGCACCTGAAAGAA
VOLP59	CCTTCCTCAGAATCCGCCACC	CCC CGCACCAAGCAG
YWLL08	ATCAAGTTTGAGGTGCTTTCC	CCATGGCATTGTGTTGAAGAC
YWLL36	AGTCTTGGTGTGGTGGTAGAA	TGCCAGGATACTGACAGTGAT

Alpaca/Llama:

ISAG STR Core Panel - Llamas and Alpacas

Locus	Forward	Reverse
LCA5	GTGGTTTTTGCCCAAGCTC	ACCTCCAGTCTGGGGATTTC
LCA8	GCTGAACCACAATGCAAAGA	AATGCAGATGTGCCTCAGTT
LCA19	TAAGTCCAGCCCCACTCA	GGTGAAGGGGCTTGATCTTC
LCA37	AAACCTAATTACCTCCCCCA	CCATGTAGTTGCAGGACACG
LCA56	ATGGTGTTTACAGGGCGTTG	GCATTACTGAAAAGCCCAGG
LCA65	TTTTTCCCCTGTGGTTGAAT	AACTCAGCTGTTGTCAGGGG
LCA66	GTGCAGCGTCCAAATAGTCA	CCAGCATCGTCCAGTATTCA
LCA94	GTCCATTCATCCAGCACAGG	ACATTTGGCAATCTCTGGAGAA
LCA99	CAGGTATCAGGAGACGGGCT	AGCATTTATCAAGGAACACCAGC
YWLL29	GAAGGCAGGAGAAAAGGTAG	CAGAGGCTTAATAACTTGCG
YWLL40	CACATGACCATGTCCCCTTAT	CCAGTGACAGTGTGACTAAGA
YWLL44	CTCAACAATGCTAGACCTTGG	GAGAACACAGGCTGGTGAATA
LGU49	TCTAGGTCCATCCCTGTTGC	GTGCTGGAATAGTGCCCAGT
LGU50	CTGCTGTGCTGTCACCCTA	AGCACCATGCCTCTAAGT

ISAG Additional Markers - Llamas and Alpacas

Locus	Forward	Reverse
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LCA24	ACTCACGGGTGACATACAGTG	GAGCAGTGTTTGTTTGCATT
YWLL08	ATCAAGTTTGAGGTGCTTCC	CCATGGCATTGTGTTGAAGAC
YWLL36	AGTCTTGGTGTGGTGGTAGAA	TGCCAGGATACTGACAGTGAT
YWLL43 (X-linked)	ATACCTCTCTGCTCTCTCTC	CCTCTACAACCATGTTAGCCA
YWLL46	AAGCAGAGTGATTTAACCGTG	GGATGACTAAGACTGCTCTGA

Pigeon:

ISAG STR Core Panel - Pigeons

Locus	Forward	Reverse
Cl μ D11	CCAATCCCAAAGAGGATTAT	ACTGTCCTATGGCTGAAGTG
Cl μ T43	GGGAAAGGAAATTTGACACTG	ACTGTCGATGCCATTAAGAC
Cl μ D01	GATTCTCAAGCTGTAGGACT	GTTTGATTGTTGGGCCATC
PIGN57	CTCTGTATGTCCATCTGAAC	ACCCATTTACCACTCTCTAA
Cl μ T13	CTGTGAGCAGTAACAGTCC	GTTTGCAAGCCCTGTTATCTCA
Cl μ D16	GCAGTGATAAAGTTCTGGAACA	GTTTGCCTCACCGTGACATCA
Cl μ D19	CTGCCGTTTCTTCTAATGCAC	GTTTGGATTCTGGGAGTGTATG
Cl μ T02	AGTTTTAATGAAGGCACCTCT	TGTAGCATGTCAGAAATTGG
Cl μ D17	TCTTACACACTCTCGACAAG	GTTTCCACCAAATGAGCAAG
Cl μ D35	GGGAGCTTAAGGGATTATTG	ATTCCTGCATGCCTACTTA
Cl μ T17	ATGGGTTTGGAGATGTTTTG	GTTTGATGGAGTTGCTATTTTGCT
PIGN04	GGTTTTCTGTTTCCTCACG	GGGATTCTGGGATTATTTTTTC

ISAG Additional Markers - Pigeons

Locus	Forward	Reverse
PIGN15	TTTCTTTTCATTTGCTGTGG	AACCAGGCATTGGAGTCTTT
PIGN10	TTCCACTGAATGGGTCTCAG	CTGCCAGAAGGTAAATGACAC
PIGN26	TCACTGTATTACCAAAGTCTG	CAATGTGGGGGCGTCTATG
PIGN12	CAGATCCAGCAGTCTTGAAG	CCCATCTAATGCGATAAATCC

Pig:

ISAG STR Core Panel - Pig

Locus	Forward	Reverse
S0005	TCCTTCCCTCCTGGTAACTA	GCACTTCTGATTCTGGGTA
S0090	CCAAGACTGCCTGTAGGTGAATA	GCTATCAAGTATTGTACCATTAGG
S0101	GAATGCAAAGAGTTCAGTGTAGG	GTCTCCCTCACACTTACCGCAG
S0155	TGTTCTCTGTTTCTCCTCTGTTG	AAAGTGGAAAGAGTCAATGGCTAT
S0227	GATCCATTTATAATTTTAGCACAAGT	GCATGGTGTGATGCTATGTCAAGC
S0228	GGCATAGGCTGGCAGCAACA	AGCCACCTCATCTTATCTACACT
S0355	TCTGGCTCCTACACTCCTTCTTGATG	TTGGGTGGGTGCTGAAAAATAGGA
S0386	GAATCCTGGGTCTTATTTTCTA	GTCAAAAATCTTTTATCTCCAACAGTAT
SW24	CTTTGGGTGGAGTGTGTGC	ATCCAAATGCTGCAAGCG
SW240	AGAAATTAGTGCCTCAAATTGG	AAACCATTAAGTCCCTAGCAAA
SW72	ATCAGAACAGTGCGCCGT	TTTGA AAAATGGGGTGTITTC
SW857	TGAGAGGTCAGTTACAGAAGACC	GATCCTCCTCAAATCCCAT
SW911	CTCAGTTCTTTGGGACTGAACC	CATCTGTGGAAAAAAAAGCC



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SW936	TCTGGAGCTAGCATAAGTGCC	GTGCAAGTACACATGCAGGG
SW951	TTTCAAACTCTGGCACCAG	GATCGTGCCCAAATGGAC
ISAG Additional Markers - Pig		
Locus	Forward	Reverse
IGF1	GCTTGGATGGACCATGTTG	CATATTTTCTGCATAAATTGAACCT
S0002	GAAGCCCAAAGAGACAACCTGC	GTTCTTTACCCACTGAGCCA
S0026	AACCTTCCCTTCCAATCAC	CACAGACTGCTTTTTACTCC
S0215	TAGGCTCAGACCCTGCTGCAT	TGGGAGGCTGAAGGATTGGGT
S0225	GCTAATGCCAGAGAAAATGCAGA	CAGGTGGAAAGAATGGAATGAA
S0226	GCACTTTAACTTTCATGATACTCC	GGTTAACTTTTNCCTCAATACA
SW632	TGGGTTGAAAGATTCCCAA	GGAGTCAGTACTTTGGCTTGA

Duty laboratories for the next comparison test with contact details

Duty laboratory Pig

Contact person: Rebecca Bellone
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Duty laboratory Dromedary

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Duty laboratory Alpaca/Llama

Contact person: Rebecca Bellone
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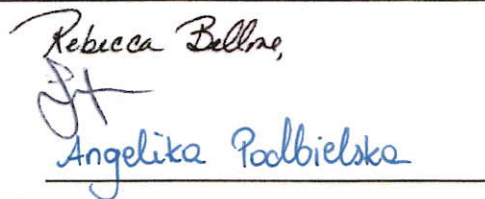
Duty laboratory Pigeon

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SIGNATURES



Chair



Duty laboratories