INTERNATIONAL SOCIETY FOR ANIMAL GENETICS

Applied Genetics and Genomics in other Species of Economic Interest

Organised by a Standing Committee: YES   ~~NO~~

Meeting information

Date: July 29, 2021
Time: 9:00 AM
Number of participants: 58

Chair

Name: Leanne van de Goor
Affiliation: VHLGenetics, The Netherlands
Contact email: Leanne.vandegoor@vhlgenetics.com

Co-Chair (optional)

Name: Marcela Martinez
Affiliation: Sociedad Rural de Argentina, Argentina
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Agenda

9:00 AM	Welcoming remarks
9:05 AM	Pig CT Discussion - Felipe Avila.
9:20 AM	Dromedary CT Discussion - Marcela Martinez.
9:35 AM	Alpaca/Llama CT Discussion - Felipe Avila
9:50 AM	Goat CT Discussion - Clementine Rodellar.
10:05 AM	Sheep CT Discussion - Agata Piestrzyńska-Kajtoch.
10:30 AM	Break
11:00 AM	Pigeon CT Discussion - Angelika Podbielska.
11:15 AM	Mutation frequency Pigeon STR marker PIGN26 - Leanne van de Goor.
11:30 AM	Workshop business meeting
	Selection of new Duty Labs for 2020-2021 Comparison tests
	Election of committee
	Any other business
11:45 AM	Evaluation of population structure alpacas maintained in Poland and identification of alpaca-llama hybrids based on microsatellite markers - Angelika Podbielska

| 11:50 AM | Rate of rejection of first-degree relationships for assigning parent-offspring relationships and estimation of genotyping errors with a high-density array in pigs - Luis Gomez-Raya |
| 11:55 AM | Molecular characterization and occurrence of variation within the promoter region of CASK gene in racing pigeons - Monika Stefaniuk-Szmukier |

Summary of the meeting

1. Welcoming Remarks

This was the fourth workshop with the name Applied Genetics and Genomics In other Species of Economic Interest. Comparison tests (CTs) for Pig, Dromedary, Alpaca/Llama, Pigeon, Sheep and Goat have been carried out. During previous conferences the CTs for Sheep and Goat were discussed during the Applied Sheep and Goat Genetics Workshop.

2. Pig CT Discussion

The UCDavis, Veterinary Genetics Laboratory (USA) was the Duty lab. Twenty-nine labs requested samples. Twenty-eight labs reported results. Twenty-two samples (including 2 reference samples) were submitted to all participants. The relative overall marker concordance among labs was good, ranging from a minimum of 95.7% (SW240) to a maximum of 100.0% (S0227, S0228, SW911, and SW936). Both parentage questions were answered correctly by twenty-seven labs, one lab didn't answer the parentage questions. Marker S0386 showed discrepancy between labs regarding allele 177 in the samples PCT08 and PCT16. This issue was already discussed during the previous CTs of 2014, 2017, and 2019. The low concordance for these samples could be explained by using primers unable to amplify allele 177. 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.

3. Dromedary CT Discussion

Qatar genetic lab (Qatar) was the Duty lab. Six labs requested samples and all six labs reported results. Twenty-four samples (including 2 reference samples) were submitted to all participants. The relative overall marker concordance among labs was good, ranging from a minimum of 93.2% (LCA37) to a maximum of 100.0% (LCA19, LCA56, LCA65, LCA66, LCA8, and YWLL29). Marker LCA19 of the core panel was mono-allelic. In the back-up panel markers LCA24, LCA77, YWLL36, and VOLP59) were also mono-allelic. This was also the case in the previous CT. The first parentage question was answered correctly by five out of six labs, the second parentage question was answered correctly by all six labs.

Because of low PE, changes in the current ISAG recommended panels are required. Rob Grahn from UCDavis offered to share data on allele frequencies from a project that is currently running in his lab. The committee will look into the data to see if it can contribute to improve the panels. Anybody who can share allele frequency data of Dromedary markers is asked to get in touch with the committee. The committee will come up with a proposal to improve the panels.

4. Alpaca/Llama CT Discussion

The UCDavis, Veterinary Genetics Laboratory (USA) was the Duty lab. Twelve labs requested samples. Ten labs reported results. Twenty-two samples (4 llama samples and 18 alpaca samples, including 2 alpaca

reference samples) were submitted to all participants. The relative overall marker concordance among labs was very good, ranging from a minimum of 97.0% (LGU50) to a maximum of 100.0% (LCA19, LCA5, LCA56, LCA65, LCA94, LCA99, YWLL29, YWLL40, and YWLL44).

The first parentage question was answered correctly by four out of ten labs. The reason for a high percentage of incorrect answers was the fact that it was a one marker exclusion parentage case. The second parentage question was answered correctly by nine out of ten labs.

It is recommended to include a reference sample with off-ladder allele 230 for marker LCA66 in the next CT.

5. Goat CT Discussion

The University of Zaragoza, LAGENBIO Lab (Spain) was the Duty lab. Thirty-two labs requested samples. Thirty labs reported results. Twenty samples (including 1 reference sample) were submitted to all participants. The relative overall marker concordance among labs was good, ranging from a minimum of 93.2% (SRCRSP08) to a maximum of 99.5% (ILSTS19). All labs answered the parentage question correctly.

Three genotypes were discussed. Sample GCT09 for marker MAF65 is heterozygous, the correct genotype is 125/155. The concordant genotype 99/ of samples GCT12 for marker OarFCB20 was not correct and will be counted as error in the ranking. The correct genotype for this samples is 99/121. The genotype of samples GCT20 for marker SRCRSP08 was not clear. Seventeen labs reported 240/ and thirteen labs reported 238/240. Angelika Podbielska and Agata Piestrzyńska-Kajtoch from the The National Research Institute of Animal Production in Poland offered to sequence this sample, results are not available yet. Because this issue was not discussed in previous CTs, for this year's CT it was decided that both the genotypes 240/ and 238/240 will not be counted as mistake for the ranking.

6. Sheep CT Discussion

The National Research Institute of Animal Production (Poland) was the Duty lab. Forty-five labs requested samples. Forty-two labs reported results. Twenty-one samples (including 1 reference sample) were submitted to all participants. The relative overall marker concordance among labs was good, with an exception for marker INRA172 which had an overall relative marker concordance of 84.2%. All other markers were above 93%. Marker INRA063 showed the highest overall relative marker concordance (99.8%). Both parentage questions were answered correctly by all forty-one labs. The genotypes of the samples OCT16 and OCT18 for marker INRA172 were discussed. For both samples, 22 labs reported 138/154 and 15 labs reported 140/154. The duty lab sequenced both samples and proved that the correct genotype for both samples is 140/154. Because this issue was not cleared out before, in this CT both the genotypes 138/154 and 140/154 will not be counted as errors in the ranking. For samples OCT15, marker INRA172 six different genotypes were reported. The duty lab sequenced this sample and proved that the smallest allele in this sample was allele 137 and therefore the correct genotype of this sample is 137/160. Because this issue was not discussed in previous CTs, for this CT an incorrectly reported 137 allele will not be counted as an error for the ranking.

7. Pigeon CT Discussion

The National Research Institute of Animal Production (Poland) was the Duty lab. Eleven labs requested samples. Nine labs reported results. Twenty-one samples (including 1 reference sample) were submitted to all participants. The relative overall marker concordance among labs was good, ranging from a minimum of 88.8% (CliμT43) to a maximum of 97.2% (PIGN4). Both parentage questions were answered correctly by eight labs, one lab didn't answer the parentage questions.

The genotype of samples PCT18 for marker CiμD35 was not clear. Three labs reported 175/177 and five labs reported 177/. Sequencing of this sample by Angelika Podbielska and Agata Piestrzyńska-Kajtoch from

The National Research Institute of Animal Production in Poland revealed that this sample is homozygous for allele 177 (27 repeats), see figure 1. Because this issue was not discussed in previous CTs, for this year's CT it was decided that both the genotypes 175/177 and 177/ will not be counted as mistake for the ranking.

Figure 1.
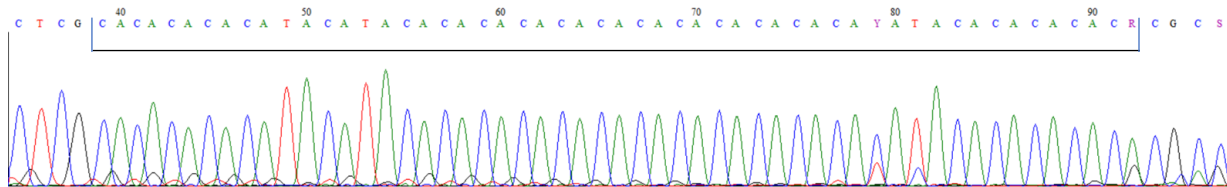
8. Mutation frequency Pigeon STR marker PIGN26
VHLGenetics presented results regarding one-marker mismatch cases in Pigeons. In a total of 4953 parentage cases, the cases with a mismatch in only one-marker and only an one-repeat difference were counted. This revealed a high mutation rate (0.73%) in marker PIGN26, see figure 2. This should be taken into account when verifying parentage in Pigeons.

Figure 2.
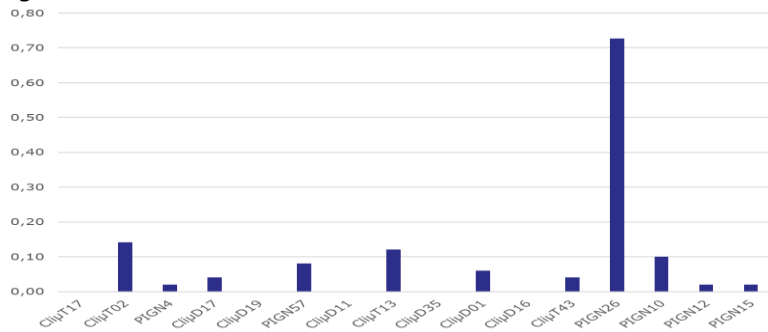
9. Pig, Goat, Sheep SNP panels for parentage verification
Participants expressed their interest in SNP panels for Pig, Goat and Sheep.
The committee will investigate the options to set-up SNPs panels for Pigs, Goats and Sheep. Labs that have allele frequency data that can be shared need to get in touch with the chair of the committee.

10. Enclosing disease markers in next CTs
All duty labs are encouraged to include a disease marker in the next CT. This is not required and is only possible if the duty lab has access to samples with heterozygous and/or homozygous mutant genotypes for a disease marker.

New Committee chair

Chair: Amparo Martinez
Term of service: first term of service 2017-2021, second term of service 2021-2025
Affiliation: University of Cordoba, Spain
E-mail address: amparomartinezuco@gmail.com

New Committee co-chair

Co-Chair: Marcela Martinez
Term of service: first term of service 2016-2019, second term of service 2019-2023
Affiliation: Sociedad Rural de Argentina, Argentina
E-mail address: mmartinez@sra.org.ar

New Committee members

Other committee members	First term of service	Second term of service	Email address
Ntanganedzeni Mapholi	2017-2021	2021-2025	maphon@unisa.ac.za
Younes Miar	2017-2021	2021-2025	miar@dal.ca
Emiliano Lasagna	2019-2023		emiliano.lasagna@unipg.it
Foluke Eunice Sola-Oja	2019-2023		solaojo.fe@unilorin.edu.ng
Angelika Podbielska	2021-2025		angelika.podbielska@iz.edu.pl

COMPARISON TEST (2020-2021)   YES   ~~NO~~
Duty laboratory Pig

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Duty laboratory Dromedary

Contact person: Hanaa Ahmed
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Duty laboratory Alpaca/Llama

Contact person: Rebecca Bellone
Affiliation: UCDavis, USA
E-mail address: rbellone@ucdavis.edu

Duty laboratory Pigeon

Contact person: Angelika Podbielska
Affiliation: National Research Institute of Animal Production, Poland
E-mail address: angelika.podbielska@iz.edu.pl

Duty laboratory Sheep

Contact person: Agata Piestrzyńska-Kajtoch

Affiliation: National Research Institute of Animal Production, Poland
E-mail address: agata.kajtoch@iz.edu.pl

Duty laboratory Goat

Contact person: Clementine Rodellar
Affiliation: Laboratorio de Genética Bioquímica (LAGENBIO). University of Zaragoza, Spain
E-mail address: rodellar@unizar.es

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	CAGAGGCTTAATAACTTGCAG
YWLL44	CTCAACAATGCTAGACCTTGG	GAGAACACAGGCTGGTGAATA

ISAG Additional Markers - Dromedary

Locus	Forward	Reverse
CVLR01	GAAGAGGTTGGGGCACTAC	CAGGCAGATATCCATTGAA
CVLR04	CCCTACCTCTGGACTTTG	CCTTTTTGGGTATTTTCAG
CVLR05	CCTTGGACCTCCTTGCTCTG	GCCACTGGTCCCTGTCATT
LCA99	CAGGTATCAGGAGACGGGCT	AGCATTTATCAAGGAACACCAGC
LGU49	TCTAGGTCCATCCCTGTTGC	GTGCTGGAATAGTGCCCAGT
VOLP3	AGACGGTTGGGAAGGTGGTA	CGACAGCAAGGCACAGGA
VOLP32	GTGATCGGAATGGCTTGAAA	CAGCGAGCACCTGAAAGAA
VOLP59	CCTTCCTCAGAATCCGCCACC	CCCGCGCACCAAGCAG
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	TAAGTCCAGCCCCACACTCA	GGTGAAGGGGCTTGATCTTC
LCA37	AAACCTAATTACCTCCCCCA	CCATGTAGTTGCAGGACACG
LCA56	ATGGTGTTTACAGGGCGTTG	GCATTACTGAAAAGCCCAGG
LCA65	TTTTTCCCCTGTGGTTGAAT	AACTCAGCTGTTGTCAGGGG
LCA66	GTGCAGCGTCCAAATAGTCA	CCAGCATCGTCCAGTATTCA
LCA94	GTCCATTCATCCAGCACAGG	ACATTTGGCAATCTCTGGAGAA

LCA99	CAGGTATCAGGAGACGGGCT	AGCATTTATCAAGGAACACCAGC
YWLL29	GAAGGCAGGAGAAAAGGTAG	CAGAGGCTTAATAACTTGCAG
YWLL40	CACATGACCATGTCCCCTTAT	CCAGTGACAGTGTGACTAAGA
YWLL44	CTCAACAATGCTAGACCTTGG	GAGAACACAGGCTGGTGAATA
LGU49	TCTAGGTCCATCCCTGTTGC	GTGCTGGAATAGTGCCCAGT
LGU50	CTGCTGTGCTTGTCACCCTA	AGCACCACATGCCTCTAAGT

ISAG Additional Markers - Llamas and Alpacas

Locus	Forward	Reverse
LCA24	ACTCACGGGTGACATACAGTG	GAGCAGTGTTTGGTTTGCATT
YWLL08	ATCAAGTTTGAGGTGCTTTCC	CCATGGCATTGTGTTGAAGAC
YWLL36	AGTCTTGGTGTGGTGGTAGAA	TGCCAGGATACTGACAGTGAT
YWLL43 (X-linked)	ATACCTCTCTTGCTCTCTCTC	CCTCTACAACCATGTTAGCCA
YWLL46	AAGCAGAGTGATTTAACCGTG	GGATGACTAAGACTGCTCTGA

Pigeon:

ISAG STR Core Panel - Pigeons

Locus	Forward	Reverse
CliμD11	CCAATCCCAAAGAGGATTAT	ACTGTCCTATGGCTGAAGTG
CliμT43	GGGAAAGGAAATTTGACACTG	ACTGTCGATGCCATTAAGAC
CliμD01	GATTTCTCAAGCTGTAGGACT	GTTTGATTTGGTTGGGCCATC
PIGN57	CTCTTGTATGTCCATCTGAAC	ACCCATTTACCACTCTCTAA
CliμT13	CTGTCGAGCAGTAACAGTCC	GTTTGCAAGCCCTGGTTATCTCA
CliμD16	GCAGTGATAAAGTTCTGGAACA	GTTTGCCTCACCGTGACATCA
CliμD19	CTGCCCGTTTCTTCTAATGCAC	GTTTGGATTTCTGGGAGTGTATG
CliμT02	AGTTTTAATGAAGGCACCTCT	TGTAGCATGTCAGAAATTGG
CliμD17	TCTTACACACTCTCGACAAG	GTTTCCACCCAAATGAGCAAG
CliμD35	GGGAGCTTAAGGGATTATTG	ATTCCTTGCATGCCTACTTA
CliμT17	ATGGGTTTGGAGATGTTTTG	GTTTGATGGAGTTGCTATTTTGCT
PIGN04	GGTTTTTCTGTTTCCTCACG	GGGATTCTGGGATTATTTTTTC

ISAG Additional Markers - Pigeons

Locus	Forward	Reverse
PIGN15	TTTCCTTTCATTTGCTGTGG	AACCAGGCATTGGAGTCTTT
PIGN10	TTCCACTGAATGGGTCTCAG	CTGCCAGAAGGTAAATGACAC
PIGN26	TCACTGTATTCACCAAAGTCTG	CAATGTGGGGGCGTCTATG
PIGN12	CAGATCCAGCAGTCTTGAAG	CCCATCTAATGCGATAAATCC

Pig:

ISAG STR Core Panel - Pig

Locus	Forward	Reverse
S0005	TCCTTCCCTCCTGGTAACTA	GCACTTCCTGATTCTGGGTA
S0090	CCAAGACTGCCTTGTAGGTGAATA	GCTATCAAGTATTGTACCATTAGG
S0101	GAATGCAAAGAGTTCAGTGTAGG	GTCTCCCTCACACTTACCGCAG
S0155	TGTTCTCTGTTTCTCCTCTGTTTG	AAAGTGGAAAGAGTCAATGGCTAT

S0227	GATCCATTTATAATTTTAGCACAAAGT	GCATGGTGTGATGCTATGTCAAGC
S0228	GGCATAGGCTGGCAGCAACA	AGCCCACCTCATCTTATCTACACT
S0355	TCTGGCTCCTACACTCCTTCTTGATG	TTGGGTGGGTGCTGAAAAATAGGA
S0386	GAACTCCTGGGTCTTATTTTCTA	GTCAAAAATCTTTTTATCTCCAACAGTAT
SW24	CTTTGGGTGGAGTGTGTGC	ATCCAAATGCTGCAAGCG
SW240	AGAAATTAGTGCCTCAAATTGG	AAACCATTAAGTCCCTAGCAAA
SW72	ATCAGAACAGTGCGCCGT	TTTGAAAATGGGGTGTTTCC
SW857	TGAGAGGTCAGTTACAGAAGACC	GATCCTCCTCCAAATCCCAT
SW911	CTCAGTTCTTTGGGACTGAACC	CATCTGTGGAAAAAAAAGCC
SW936	TCTGGAGCTAGCATAAGTGCC	GTGCAAGTACACATGCAGGG
SW951	TTTCACAACTCTGGCACCAG	GATCGTGCCCAAATGGAC

ISAG Additional Markers - Pig

Locus	Forward	Reverse
IGF1	GCTTGGATGGACCATGTTG	CATATTTTTCTGCATAACTTGAACCT
S0002	GAAGCCCAAAGAGACAACTGC	GTTCTTTACCCACTGAGCCA
S0026	AACCTTCCCTTCCCAATCAC	CACAGACTGCTTTTTACTCC
S0215	TAGGCTCAGACCCTGCTGCAT	TGGGAGGCTGAAGGATTGGGT
S0225	GCTAATGCCAGAGAAATGCAGA	CAGGTGGAAAGAATGGAATGAA
S0226	GCACTTTTAACTTTCATGATACTCC	GGTTAAACTTTTNCCCCAATACA
SW632	TGGGTTGAAAGATTTCCCAA	GGAGTCAGTACTTTGGCTTGA

Sheep:

ISAG STR Core Panel - Sheep

Locus	Forward	Reverse
AMEL	CAGCCAAACCTCCCTCTGC	CCCGCTTGGTCTTGTCTGTTGC
CSRD247	GGACTTGCCAGAACTCTGCAAT	CACTGTGGTTTGTATTAGTCAGG
ETH152	TACTCGTAGGGCAGGCTGCCTG	GAGACCTCAGGGTTGGTGATCAG
INRA005	TTCAGGCATACCCTACACCACATG	AAATATTAGCCAACTGAAAACTGGG
INRA006	AGGAATATCTGTATCAACCGCAGTC	CTGAGCTGGGGTGGGAGCTATAAATA
INRA023	GAGTAGAGCTACAAGATAAACTTC	TAACTACAGGGTGTTAGATGAACTC
INRA063	GACCACAAAGGGATTTGCACAAGC	AAACCACAGAAATGCTTGGAAG
INRA172	CCAGGGCAGTAAAATGCATAACTG	GGCCTTGCTAGCCTCTGCAAAC
MAF065	AAAGGCCAGAGTATGCAATTAGGAG	CCACTCCTCCTGAGAATATAACATG
MAF214	AATGCAGGAGATCTGAGGCAGGGACG	GGGTGATCTTAGGGAGGTTTTGGAGG
McM042	CATCTTTCAAAAGAACTCCGAAAGTG	CTTGGAATCCTTCCTAACTTTCGG
McM527	GTCCATTGCCTCAAATCAATTC	AAACCACTTGACTACTCCCCAA
OarFCB20	GGAAAACCCCCATATATACCTATAC	AAATGTGTTTAAGATTCCATACATGTG

Goat:

ISAG STR Core Panel - Goat

Locus	Forward	Reverse
CSRD247	GGACTTGCCAGAACTCTGCAAT	CACTGTGGTTTGTATTAGTCAGG
ILSTS008	GAATCATGGATTTTCTGGGG	TAGCAGTGAGTGAGGTTGGC
ILSTS19	AGGGACCTCATGTAGAAGC	ACTTTGGACCCTGTAGTGC
ILSTS87	AGCAGACATGATGACTCAGC	CTGCCTCTTTTCTTGAGAGC

INRA005	TTCAGGCATACCCTACACCACATG	AAATATTAGCCAACTGAAAACTGGG
INRA006	AGGAATATCTGTATCAACCGCAGTC	CTGAGCTGGGGTGGGAGCTATAAATA
INRA023	GAGTAGAGCTACAAGATAAACTTC	TAACTACAGGGTGTTAGATGAACTC
INRA063	GACCACAAAGGGATTTGCACAAGC	AAACCACAGAAATGCTTGGAAG
MAF65	AAAGGCCAGAGTATGCAATTAGGAG	CCACTCCTCCTGAGAATATAACATG
McM527	GTCCATTGCCTCAAATCAATTC	AAACCACTTGACTACTCCCCAA
OarFCB20	GGAAAACCCCCATATATACCTATAC	AAATGTGTTTAAGATTCCATACATGTG
SRCRSP23	TGAACGGGTAAAGATGTG	TGTTTTTAATGGCTGAGTAG
SRCRSP5	GGACTCTACCAACTGAGCTACAAG	TGAAATGAAGCTAAAGCAATGC
SRCRSP8	TGCGGTCTGGTTCTGATTTCAC	CCTGCATGAGAAAGTCGATGCTTAG

Duty laboratory for the next comparison tests
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

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Duty laboratory Sheep

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Duty laboratory Goat

Contact person: Clementine Rodellar

| Affiliation: Laboratorio de Genética Bioquímica (LAGENBIO). University of Zaragoza, Spain |
| E-mail address: rodellar@unizar.es |

SIGNATURES

Chair	**Duty laboratory**