ISAG CONFERENCE 2014, Xi'an, China

Pigeons, regional farmed animals and wild animals

STANDING COMMITTEES / WORKSHOPS Information will be posted online

Organised by a standing committee no

Date and meeting time: Tuesday, July 29th, 12:00-14:00 hr

Chair, name and contact email: Rainer Schubbert, RainerSchubbert@eurofins.com

Agenda / programme attached

1. *A new standard marker set for pigeons*
   Part 1: Results of the first comparison test - Leanne van de Goor
   Part 2: Agreement on the pigeon panel and interpretation rules for parentage verification - Leanne van de Goor
2. Is there a need for other standard marker sets for "less popular species" ? - Henriette van der Zwaan
3. Next steps / Reporting - Rainer Schubbert
4. Forming a Committee, election of members - Rainer Schubbert

Number of participants at meeting: 20

Summary of the meeting

5. ***A new standard marker set for pigeons***

Part 1: Results of the first comparison test - Leanne van de Goor

Routine parentage verification in pigeons is an emerging market. Racing pigeons are sold for prices ranging from $10k – $500k. International exchange of profiles not possible → need for international standard.

Two separate marker panels exist:

- PIGN markers developed by ABI
- CliμT / CliμD marker published by Traxler et al., 2000

The new marker panel with 16 STR markers combines ABI and Traxler markers.

The panel has to be tested in an ISAG CT, CT sample kit comprised of 16 samples + 1 positive control with known STR profile, Reaction mixture (primers + buffers) for 50 rxn, Kit-specific Taq polymerase.

Participants were asked to report the profiles back, using an Excel file. In total 10 labs agreed to participate in the CT: one from Canada, The Netherlands, France, Poland, South-Africa, Ireland, Spain and 3 from Germany. Until date of the workshop 7 labs already reported back:

16 samples x 16 markers 256 genotypes could be reported back.

- Of a total of 1792 genotypes, 1789 genotypes were reported back (99,83%).
- If four or more labs report the same genotype the genotype is scored as "concordant"
- Four genotypes had to be excluded from further calculations (PIGN12, sample B9 +B13 and CliμD16, sample B4 + B5).
- Majority of the genotypes has been scored equally.
- 1764 genotypes remain for calculations.
- 127 non-concordant genotypes (7,2%).
- From 127 non-concordant genotypes, 91 are found in only 4 markers: PIGN26: 22, PIGN10: 24, PIGN12: 26, PIGN15: 27.
- Furthermore one marker CliμT43 with 15 non-concordant genotypes, but all from one lab.
- All remaining markers 6 or less non-concordant genotypes.
- If the four markers with the most non-concordant genotypes (PIGN26, PIGN10, PIGN12, PIGN15) are excluded from the calculations, only 46 non-concordant genotypes remain (3,5%).

Part 2: Agreement on the pigeon panel and interpretation rules for parentage verification - Leanne van de Goor

It was decided by the representatives of the institutional members in the workshop that the following 12 STR markers are the ISAG Recommended Pigeon panel:
CliμT17, CliμT02, PIGN4, CliμD17, CliμD19, PIGN57, CliμD11, CliμT13, CliμD35, CliμD01, CliμD16, CliμT43

It was decided by the representatives of the institutional members in the workshop that the following 4 STR markers are the Back-up ISAG Pigeon panel:
PIGN26, PIGN10, PIGN12, PIGN15

Remaining goals:
- Determine allele frequencies of each marker,
- Determine PIC/PE/HO/HE,
- Publish findings in an article,
- Determine if 12 ISAG recommended markers are enough.

The following interpretation rules for parentage verification were accepted by the representatives of the institutional members in the workshop
- Minimum number of STRs available in parentage verification: 12
- If mismatches occur in a supposed parentage, the general rule is to first retest the samples involved or request new samples to confirm the determined genotypes. If the genotypes are confirmed the following guidelines should be followed:
    - 0 mismatches: Parentage qualifies
    - 1 mismatch: Parentage doubtful. If not all 16 STRs are complete, first complete the profiles to 16 STRs. If 1 mismatch remains, ask customer for other possible parents. If there are no other possible parents, then qualify the parentage
    - 2 or more mismatches: Parentage excluded

6. **Is there a need for other standard marker sets for "less popular species" ? - Henriette van der Zwaan**

Henriette gave a presentation about the situation in South Africa, no decision was made.

7. **Next steps / Reporting / Comparison tests - Rainer Schubbert**

It was discussed to form a Committee together with the Camelid Genetics and Genomics Committee. " Other species of economical interest" was proposed as name for this committee. The topic should be discussed in the workshop on Friday 01st of August.

The pigeon comparison test should be done in the future on regular bases.

COMPARISON TEST (2013-2014) **yes**

If yes: Number of enquiries – requests for consignment forms: 10
Number of participants receiving samples: 10
Number of samples: 16
Number of participants reporting results: 7

Duty laboratory Dr. van Haeringen Laboratorium, The Netherlands, Maarten de Groot, mgr@vhladmin.nl

Computing Laboratory Dr. van Haeringen Laboratorium, The Netherlands, Maarten de Groot, mgr@vhladmin.nl

List of recommended markers with primer information

Marker	Primer	Sequence	Size (bp)	Dye
CliμD11	F	CCAATCCCAAAGAGGATTAT	77 - 99	6-FAM
	R	ACTGTCCTATGGCTGAAGTG		
PIGN15	F	TTTCCTTTCATTTGCTGTGG	126 - 150	6-FAM
	R	AACCAGGCATTGGAGTCTTT		
CliμT43	F1	GGGAAAGGAAATTTGACACTG	191 - 223	6-FAM
	R	ACTGTCGATGCCATTAAGAC		
PIGN10	F1	TTCCACTGAATGGGTCTCAG	271 - 325	6-FAM
	R1	CTGCCAGAAGGTAAATGACAC		
CliμD01	F	GATTTCTCAAGCTGTAGGACT	75 - 113	VIC
	R	GTTTGATTTGGTTGGGCCATC		
PIGN57	F	CTCTTGTATGTCCATCTGAAC	155 - 187	VIC
	R	ACCCATTTACCACTCTCTAA		
CliμT13	F1	CTGTCGAGCAGTAACAGTCC	198 - 230	VIC
	R	GTTTGCAAGCCCTGGTTATCTCA		
PIGN26	F1	TCACTGTATTCACCAAAGTCTG	358 - 469	VIC
	R1	CAATGTGGGGGCGTCTATG		
CliμD16	F	GCAGTGATAAAGTTCTGGAACA	76 - 172	NED
	R	GTTTGCCTCACCGTGACATCA		
CliμD19	F1	CTGCCCGTTTCTTCTAATGCAC	193 - 199	NED
	R	GTTTGGATTTCTGGGAGTGTATG		
PIGN12	F1	CAGATCCAGCAGTCTTGAAG	232 - 348	NED
	R1	CCCATCTAATGCGATAAATCC		
CliμT02	F	AGTTTTAATGAAGGCACCTCT	92 - 104	PET

	R	TGTAGCATGTCAGAAATTGG		
CliμD17	F	TCTTACACACTCTCGACAAG	116 - 130	PET
	R	GTTTCCACCCAAATGAGCAAG		
CliμD35	F	GGGAGCTTAAGGGATTATTG	173 - 187	PET
	R	ATTCCTTGCATGCCTACTTA		
CliμT17	F	ATGGGTTTGGAGATGTTTTG	209 - 249	PET
	R	GTTTGATGGAGTTGCTATTTTGCT		
PIGN04	F	GGTTTTTCTGTTTCCTCACG	273 - 327	PET
	R	GGGATTCTGGGATTATTTTTTC		

Duty laboratory for the next comparison test : Dr. van Haeringen Laboratorium, The Netherlands, Maarten de Groot, mgr@vhladmin.nl

Computing laboratory for the next comparison test: Dr. van Haeringen Laboratorium, The Netherlands, Maarten de Groot, mgr@vhladmin.nl

SIGNATURES

Chair **Duty laboratory** **Computing laboratory**