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Abstract

The lack of SNP arrays for less traditional livestock species has led researchers to explore the possibility of applying the use of commercial SNPs already detected in other related livestock species. For example, the medium density bovine and ovine SNP chips have been successfully used to detect polymorphic SNPs in several other species. The dromedary camel belongs to the *Camelidae* family and represents an important source for production of milk, transportation and meat in several countries. Until now, no SNP chip has been developed for the dromedary and genome information is still incomplete when compared to other livestock species. In this study, 8 to 17 dromedary samples were genotyped using 3 different ruminant species SNP chips: the Bovine 777K SNP BeadChip, the Ovine 600K SNP BeadChip, and the Caprine 50K SNP BeadChip. The quality of hybridization was assessed considering the average value between the 10%GC and the 50%GC (averageGC) scores and the call rate. Only SNPs with averageGC >0.70 and call rate=1.0 were considered.

A total of 29,900 Bovine and 14,179 Ovine SNPs were considered successfully genotyped while none of the Caprine SNPs were qualified for further analysis. Among these SNPs, only 27,585 for the bovine and only 88 for the ovine were polymorphic in our dromedary dataset. These SNPs could represent the first step towards the development of a SNP chip useful for this camelid species after appropriate validations.

Keywords

Camelus dromedarius, gene call, ruminants

Disciplines

Agriculture | Animal Sciences | Genetics | Genomics

Comments

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Evaluation of the application of bovine, ovine and caprine SNP chips to dromedary genotyping

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Abstract

The lack of SNP arrays for less traditional livestock species has led researchers to explore the possibility of applying the use of commercial SNPs already detected in other related livestock species. For example, the medium density bovine and ovine SNP chips have been successfully used to detect polymorphic SNPs in several other species. The dromedary camel belongs to the *Camelidae* family and represents an important source for production of milk, transportation and meat in several countries. Until now, no SNP chip has been developed for the dromedary and genome information is still incomplete when compared to other livestock species. In this study, 8 to 17 dromedary samples were genotyped using 3 different ruminant species SNP chips: the Bovine 777K SNP BeadChip, the Ovine 600K SNP BeadChip, and the Caprine 50K SNP BeadChip. The quality of hybridization was assessed considering the average value between the 10%GC and the 50%GC (averageGC) scores and the call rate. Only SNPs with averageGC >0.70 and call rate=1.0 were considered.

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Introduction

Over the past several years, the increase in genomic technologies and molecular information has provided the possibility of developing useful tools for genome wide analyses in livestock. The implementation of high-density SNP panels for farm animals has provided the opportunity of performing genome-wide studies to scan in parallel a massive number of loci across the genome and identify associations with commercial production traits and/or diseases, providing more comprehensive pictures of the genetic control of important economic traits. Since 2008, beginning with cattle (Matukumalli et al 2009), a series of Single Nucleotide Polymorphism (SNP) chips of medium and high density have been developed and assessed for the major livestock species such as cattle, pig, sheep, horse and goat (reviewed by Nicolazzi et al 2015).

The single-humped Dromedary camel (*Camelus dromedarius*) belongs to the old world *Camelidae* subfamily and represents almost 90% of the genus *Camelus*. It is distributed in the hot arid areas of the Middle East and Africa (Dorman 1986). Dromedaries are multipurpose animals with females used primarily as milk producers, the males for transport or draught and both sexes provide meat as a tertiary product (Bulliet 1990; Grigson 2012). The dromedary camel represents a good source of meat especially in arid areas

because of its peculiar physiological characteristics that make it extremely tolerant to high temperatures, solar radiation, water and food scarcity. (Kadim et al 2008). So far, genomic resources tailored for dromedary studies are lacking. Recent research has provided first preliminary assemblies of two different female dromedaries (Wu et al 2014; Fitak et al 2016). Microsatellite variability (e.g. Evdotchenko et al 2003; Ahmed et al 2010; Mahmoud et al 2016) has been investigated mainly for population genetics purposes. Additionally, SNPs have been determined by resequencing gene regions to analyze genetic diversity and to identify markers associated with milk and meat traits (e.g. Pauciullo et al 2014; Khabiri et al 2014). The SRA (Short Read Nucleotide) database contains available NGS (Next Generation Sequencing) reads of a very limited number of dromedaries even if the number is increasing.

The lack of SNP arrays for less traditional livestock species, e.g. *Camelidae*, as well as the limited knowledge about their genomes, has led researchers to consider exploring the possibility of applying these existing commercial SNPs to other species and to assess the quality of hybridization and the possibility of their use in genotyping analysis. For example, the medium density bovine SNP chip has been used with several degrees of success to detect polymorphic SNPs in European and American bison (Pertoldi et al 2010), in two antelope species (Ogden et al 2012), in two reindeer species of the *Cervidae* family (Haynes and Latch 2012) and in Russian reindeers (Kharzinova et al 2015). In the same work, Kharzinova et al (2015) also analyzed and detected polymorphic SNPs genotyping the same Russian reindeer with the ovine SNP chip. The success of these approaches has varied between species, for number of SNPs detected, applications and the possibility of false positive rate.

In this study, we investigated the results of dromedary samples genotyped with three SNP chips belonging to three different species (Bovine, Ovine, and Caprine) to evaluate the quality of signals and to find SNPs that could be useful for the dromedary camel. The three SNP chips were chosen because of they were all similar species and to maximize the number of available SNPs that would allow finding the highest possible number of high quality SNPs for the dromedary.

Material and methods

Blood samples were randomly collected from 17 dromedaries of unknown sex located in Egypt (Southern Egypt Dromedaries, originated from Sudan and the Maghrabi breed). All animal care was provided by in country providers and approved locally. Blood was collected on FTA papers, and sent to the United States where they were processed. The animals were genotyped using the Bovine 777K-SNP BeadChip (8 dromedaries= DROM-BOV), the Ovine 600K-SNP BeadChip (17 dromedaries= DROM-OV) and the Caprine 50K SNP BeadChip (12 dromedaries= DROM-CAP), all provided by Illumina (Illumina, San Diego, CA).

The Genome Studio software (Illumina, San Diego, CA) was used to analyze the raw signals and to convert into genotype calls for individual animals, loci genotyping and quality scores. Because the probe hybridization can affect the genotyping results and therefore increase the number of false positives, the reliability of the genome calls was estimated using several steps. First, performance at each locus was evaluated calculating an averageGC (GeneCall) score, which was obtained considering the average between 10%GC and 50%GC scores (the 10th percentile and the 50th percentile of the GC scores respectively). Following the manufacturer information of Genome Studio, the GC score is means by which to rank and filter out failed genotypes, DNAs, and/or loci (Oliphant et al 2002). The threshold was assessed to capture the highest number of SNPs with the highest value for averageGC score that ranged from 0 (bad performance) to 1 (good performance). To select a quality threshold and perform the SNP selection, 70 cattle samples genotyped with the bovine 50K SNP chip (Kim and Rothschild 2014), 42 sheep samples genotyped with the ovine SNP chip (Zhao et al 2011) and approximately 80 goat samples genotyped with the goat SNP chip (Kim et al 2015) were inspected with the same parameters (Figure 1).

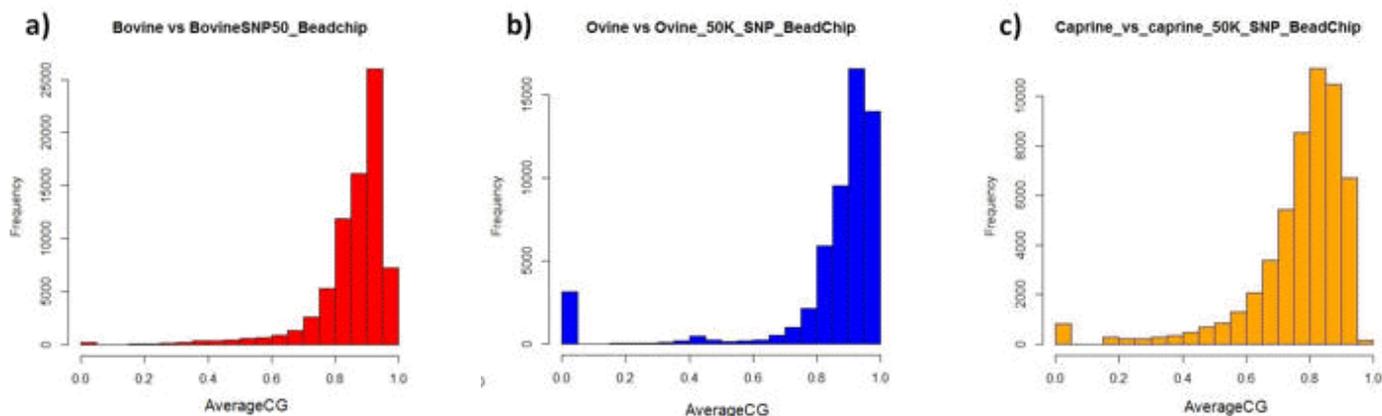


Figure 1. Average GC value frequency distribution of each locus using medium-density SNP chip and genotyping (a) Cattle with bovine SNP chip b) Sheep samples with the ovine SNP chip, and c) Goat samples vs caprine SNP chip.

In a same-species genotyping (bovine vs bovine, ovine vs ovine and caprine vs caprine) the averageGC value of 0.7 was set as a common threshold that allowed us to retain 93.2%, 90.4% and 79.2% of bovine, ovine and goat SNPs, respectively. Then, each SNP of the three different dromedary genotyping combinations was screened considering the averageGC and only the loci with an average value ≥ 0.70 were retained.

The software Plink 1.07 (Purcell et al 2007) was used to calculate the call rate of the SNPs selected during the averageGC filtering step. Only SNPs with call rate=1.0 were considered. For the final selected SNPs, the same software was used to calculate MAF (Minor Allele Frequency) to investigate the presence of polymorphic SNPs.

Results and discussion

The application of the averageGC threshold procedures to the dataset allowed the retention of 130,828 SNPs (~16.82%) for DROM-BOV, 18,871 SNPs (~3.11%) for DROM-OV and 8,009 SNPs (~15.01%) for DROM-CAP (Figure 2). The filtered SNPs showed a wide distribution based on the call rate in all of the SNP chip combinations (Figure 3).

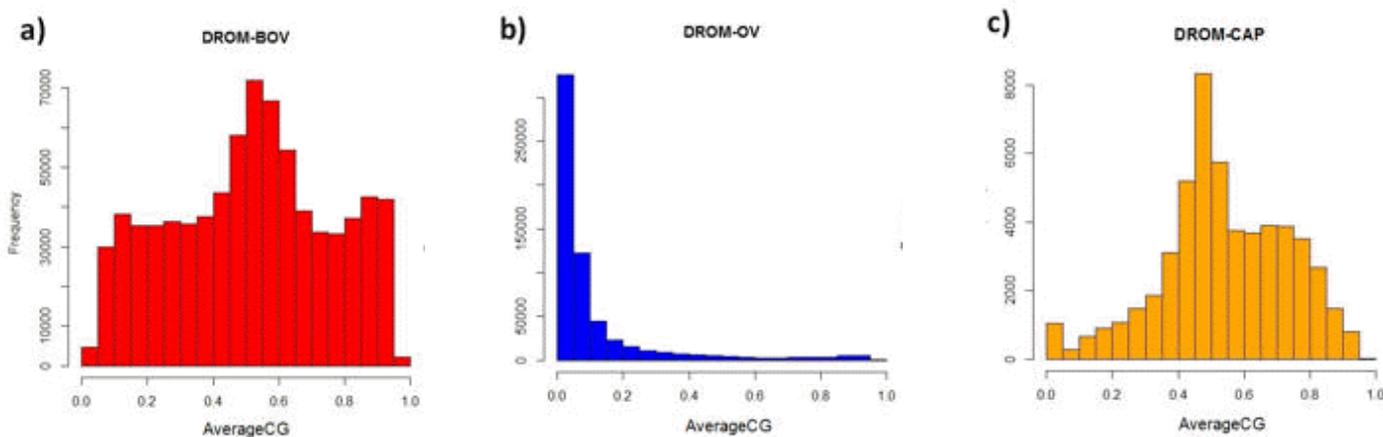


Figure 2. AverageGC values frequency distribution of each locus using cattle 777K, Ovine 600K and Caprine 50K SNP chips for dromedary (a) DROM-BOV b) DROM-OV c) DROM-CAP.

DROM-BOV: Dromedary samples genotyped with the Bovine 777K-SNP BeadChip.

DROM-OV: Dromedary samples genotyped with the Ovine 600K-SNP BeadChip.

DROM-CAP: Dromedary samples genotyped with the Caprine 50K SNP BeadChip.

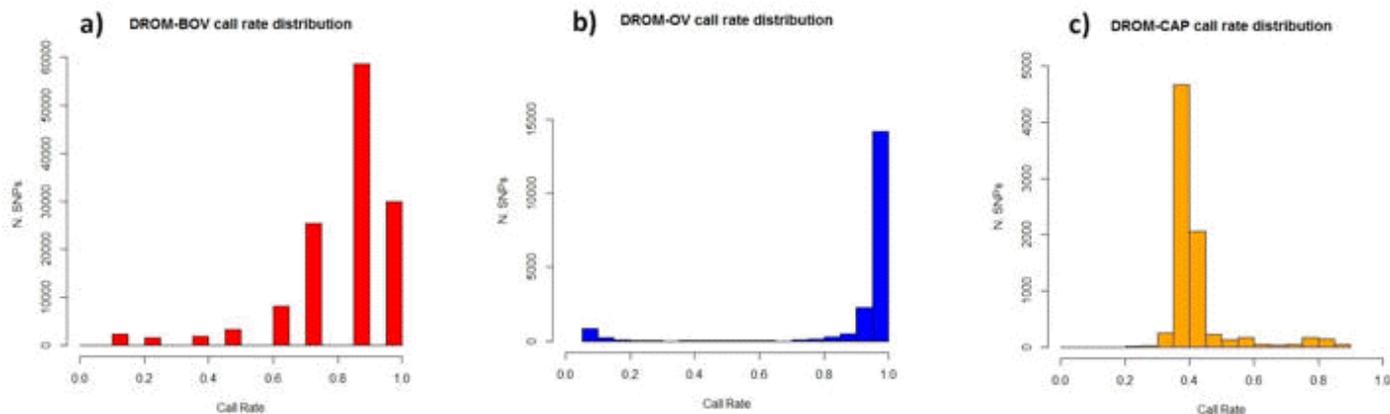


Figure 3. Call rate distribution of the SNPs with averageGC > 0.7 across the different SNP chips.

DROM-BOV: Dromedary samples genotyped with the Bovine 777K-SNP BeadChip.

DROM-OV: Dromedary samples genotyped with the Ovine 600K-SNP BeadChip.

DROM-CAP: Dromedary samples genotyped with the Caprine 50K SNP BeadChip.

Among the initial filtering a total of 29,900 SNPs for DROM-BOV and 14,179 for DROM-OV had Call Rate= 1.0 and were considered for the subsequent analysis. No SNPs with call rate=1 were detected for DROM-CAP. In Figure 4 is shown the distribution of the SNPs across the chromosomes based on cattle, sheep and goat genome. Generally, almost every chromosome is represented in the selection, but the number of SNPs varied depending on the chromosome. In DROM-BOV the lowest number of 338 SNPs was located on chromosome 25 and the highest number of 2,130 SNPs was on chromosome 1. In DROM-OV the lowest number of 152 SNPs was located on chromosome 26 and the highest of 1,761 SNPs was still on chromosome 1. The different number of SNPs detected along the different chromosomes was probably due to the initial number of SNPs available on each chromosome in each SNP chip. During the SNP selection process, SNPs located on sex chromosomes and non-annotated SNPs were included to maximize the number of SNPs detected. Since chromosomes are of different sizes and therefore have a different number of mapped SNPs on the chips, the general observation is that for each comparison larger chromosomes contributed with a higher number of SNPs than smaller chromosomes. However, this distribution cannot be applied directly to the dromedary genome without a good comparative genomic map between the dromedary genome and the bovine-ovine genomes.

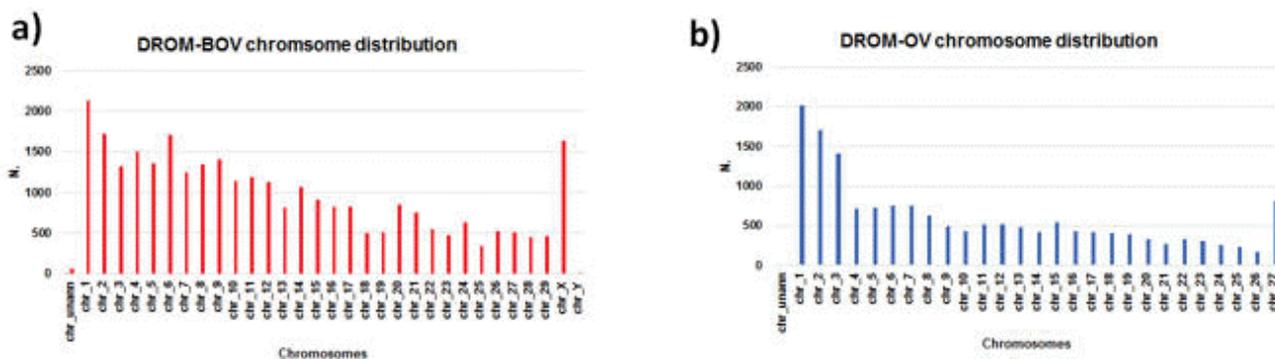


Figure 4. Chromosome distribution of the SNPs with call rate=1.0 for a) DROM-BOV considering the 777K bovine SNP chip and the UMD3.1 based map b) DROM-OV considering the 600K ovine SNP chip and the Oar 3.1 based map.

Due to the limited number of animals, each SNP with MAF > 0 was considered as polymorphic for the purpose of this study. Considering only the SNPs with Call Rate= 1.0 previously discussed, the majority of SNPs were polymorphic in the Bovine SNP chip, with 27,585 polymorphic SNPs for DROM-BOV comparison and 88 polymorphic SNPs for DROM-OV comparison.

Comparing the results with those obtained in other species, we note that the percentage of polymorphic SNPs for DROM-BOV is unexpectedly higher than those found in deer and bison genotyped with the Bovine 50K SNP chip (Haynes et al 2012, Pertoldi et al 2010) or in wild sheep genotyped with the Ovine 50K SNP chip

(Miller et al 2011). The high level of polymorphism could result from the high heterogeneity of the samples analyzed or we hypothesize that such results may be partially determined by false positives for DROM-BOV. It is known that the *Camelidae* and *Bovidae* families diverged around 42.7 Mya (Wu et al 2014) and *Bovidae* is considered now the closest family to the *Camelidae*. The divergence of the *Caprinae* subfamily (sheep and goat) from the *Bovidae* occurred around 19 Mya ago (Jiang et al 2014). The results obtained by our analysis suggest that the *Bovidae* subfamily, and *Bos Taurus* in particular, maintain the highest similarity across the *Camelidae* family at least based on our dromedary study which is confirmed by the high synteny of the two genomes (Wu et al 2014).

In conclusion, we detected thousands of high quality SNPs that were polymorphic in dromedary using SNP chips that belong to three different ruminant species. Several of these SNPs are within coding regions, reducing the variability around the SNPs for which the probes are built. These SNPs could be considered for future development of dromedary commercial SNP chips. More effort should be devoted to improving the genome assembly of the dromedary reference genome and increasing the number of whole sequenced genomes using single animals and pooled DNA, not only for the dromedary but also for the whole *Camelidae* family. This will allow the opportunity to build a *Camelidae* SNP chip and provide developing countries a comprehensive low-cost genomic tool that will help to understand the genomic bases of productive traits in this peculiar family and therefore allow for breeding plans to improve sustainability and resistance for these species.

Conclusions

- We evaluated the general performances obtained by genotyping dromedary samples with ruminant SNP chips.
- While overall performance is low, part of these SNPs could be potentially used and be part of a dromedary SNP chip.
- Taking advantage of the increasing information on high throughput genomic data, some prior SNP validation will be necessary to verify results found in this study.

Conflict of interest

Authors declare no conflict of interest.

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References

Ahmed M O, Salem F B, Bedhiaf S, Rekik B and Djemali M 2010 Genetic diversity in Tunisian dromedary (*Camelus*

dromedarius) populations using microsatellite markers. *Livestock Science* 132: 182-185.

Bulliet R W 1990 The camel and the wheel. Columbia University Press

Dorman A E 1986 Aspects of the husbandry and management of the genus *Camelus*. In A Higgins (Ed), *The camel in health and disease* (pp 3–20). London: Balliere Tindal.

Evdotchenko D, Han Y, Bartenschlager H, Preuss S and Geldermann H 2003 New polymorphic microsatellite loci for different camel species. *Molecular Ecology Notes* 3: 431-434.

Fitak R R, Mohandesan E, Corander J and Burger P A 2016 The de novo genome assembly and annotation of a female domestic dromedary of North African origin. *Molecular Ecology Resources* 16: 314.

Grigson, C 2012 Camels, copper and donkeys in the early iron age of the southern levant: timna revisited. *Levant* 44.1: 82-100.

Haynes G D and Latch E K 2012 Identification of novel single nucleotide polymorphisms (SNPs) in deer (*Odocoileus* spp) using the BovineSNP50 BeadChip *PLoS One* 7: e36536.

Kadim I T, Mahgoub O and Purchas R W 2008 A review of the growth, and of the carcass and meat quality characteristics of the one-humped camel (*Camelus dromedaries*) *Meat Science* 80: 555-569.

Khabiri A A, Tahmoorespur M, Nassiri M R and Hadi M 2014 Sequencing and bioinformatics analysis of the partial promoter region of κ -casein (CSN3) gene in Iranian Bacterianus and Dromedaries camels. *International journal of Advanced Biological and Biomedical Research* 2: 1719-1725.

Kharzinova V R, Sermyagin A A, Gladyr E A, Okhlopkov I M, Brem G and Zinovieva N A 2015 A study of applicability of SNP chips developed for bovine and ovine Species to whole-genome analysis of reindeer rangifer tarandus. *Journal of Heredity* 106: 758-761.

Kim E S, Elbeltagy A R, Aboul-Naga A M, Rischkowsky B, Sayre B, Mwacharo J M and Rothschild M F 2016 Multiple genomic signatures of selection in goat and sheep indigenous to a hot arid environment. *Heredity* 116: 255-264.

Kim E S and Rothschild M F 2014 Genomic adaptation of admixed dairy cattle in East Africa. *Frontiers in Genetics* 5: 1-9.

Jiang Y, Xie M, Chen W, Talbot R, Maddox JF, Faraut T, Wu C, Muzny D M, Li Y, Zhang W, Stanton J A, Brauning R, Barris W C, Hourlier T, Aken B L, Searle S M, Adelson D L, Bian C, Cam G R, Chen Y, Cheng S, DeSilva U, Dixen K, Dong Y, Fan G, Franklin I R, Fu S, Fuentes-Utrilla P, Guan R, Highland M A, Holder M E, Huang G, Ingham A B, Jhangiani S N, Kalra D, Kovar C L, Lee S L, Liu W, Liu X, Lu C, Lv T, Mathew T, McWilliam S, Menzies M, Pan S, Robelin D, Servin B, Townley D, Wang W, Wei B, White S N, Yang X, Ye C, Yue Y, Zeng P, Zhou Q, Hansen J B, Kristiansen K, Gibbs R A, Flicek P, Warkup C C, Jones H E, Oddy V H, Nicholas F W, McEwan J C, Kijas J W, Wang J, Worley K C, Archibald A L, Cockett N, Xu X, Wang W and Dalrymple B P 2014 The sheep genome illuminates biology of the rumen and lipid metabolism. *Science* 344: 1168-1173

Mahmoud A H, Alshaikh M A, Aljumaah R S and Mohammed O B 2016 Genetic variability of camel (*Camelus dromedarius*) populations in Saudi Arabia based on microsatellites analysis. *African Journal of Biotechnology* 11: 11173-11180.

Matukumalli L K, Lawley C T, Schnabel R D, Taylor J F, Allan M F, Heaton M P, O'Connell J, Moore S S, Smith T P, Sonstegard T S and Van Tassell C P 2009 Development and characterization of a high density SNP genotyping assay for cattle. *PLoS One* 4: e5350.

Miller J M, Poissant J, Kijas J W, Coltman D W and International Sheep Genomics Consortium 2011 A genome-wide set of SNPs detects population substructure and long range linkage disequilibrium in wild sheep. *Molecular Ecology Resources* 11: 314-322.

Nicolazzi E L, Biffani S, Biscarini F, Orozco Ter Wengel P, Caprera A, Nazzicari N and Stella A 2015 Software solutions for the livestock genomics SNP array revolution *Animal Genetics* 46: 343-353.

Ogden R, Baird J, Senn H and McEwing R 2012 The use of cross-species genome wide arrays to discover SNP markers for conservation genetics: a case study from Arabian and scimitar-horned oryx. *Conservation Genetics Resources* 4: 471–473.

Oliphant A, Barker D L, Stuelpnagel J R and Chee M S 2002 BeadArray™ Technology: Enabling an Accurate, Cost-Effective Approach to High-Throughput Genotyping SNPs; *Discovery of Markers for Disease Biotechniques* 32:S56-S61.

Paucullo A, Giambra I J, Iannuzzi L and Erhardt G 2014 The β -casein in camels: molecular characterization of the CSN2 gene, promoter analysis and genetic variability. *Gene* 15: 159-168.

Pertoldi C, Wójcik J M, Tokarska M, Kawalko A, Kristensen T N, Loeschcke V, Gregersen V R, Coltman D, Wilson G A, Randi E, Henryon M and Bendixen C 2010 Genome variability in European and American bison detected using BovineSNP50 BeadChip. *Conservation Genetics* 11: 627–634.

Purcell S, Neale B, Todd-Brown K, Thomas L, Ferreira M A R, Bender D, Maller J, Sklar P, de Bakker P I W, Daly M J, Sham P C 2007 PLINK: a toolset for whole-genome association and population-based linkage analysis. *American Journal of Human Genetics* 81: 559-57.

Wu H, Guang X, Al-Fageeh M B, Cao J, Pan S, Zhou H, Zhang L, Abutarboush MH, Xing Y, Xie Z, Alshaqeeti A S, Zhang Y, Yao Q, Al-Shomrani B M, Zhang D, Li J, Manee M M, Yang Z, Yang L, Liu Y, Zhang J, Altammami M A, Wang S, Yu L, Zhang W, Liu S, Ba L, Liu C, Yang X, Meng F, Wang S, Li L, Li E, Li X, Wu K, Zhang S, Wang J, Yin Y, Yang H, Al-Swailem A M and Wang J 2014 Camelid genomes reveal evolution and adaptation to desert environments. *Nature Communication* 5: 5188.

Zhao X, Dittmer K E, Blair H T, Thompson K G, Rothschild M F and Garrick D J 2011 A novel nonsense mutation in the DMP1 gene identified by a genome-wide association study is responsible for inherited rickets in Corriedale sheep. *PLoS One* 6: e21739.

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