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Computational Analysis of Physical and Chemical Properties of the GDF9 Protein of Goat

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Abstract

As essential growth factors and receptors in follicular development in the ovaries, different mutations in genes of the transforming growth factor β (TGF β) superfamily have been shown to affect ovulation rate in sheep and goat. This study aimed at investigating computationally, the molecular genetic variation of the GDF9 protein of goats. A total of 8 GDF9 protein of goats were retrieved from the GENBank. The GDF9 gene is one of the ovary derived transforming growth factor β (TGF β) super family, the genes code for proteins chemical parameters of the proteins was analyzed. The secondary structure of GDF9 protein was also predicted. Analysis of amino acid composition indicates leucine as having the highest percentage (11.0%) while selenocysteine and pyrrolysine have the lowest percentage (0.0%). Secondary structure indicates 16.56% extended strand, 30.0% alpha helix, 5.74% beta turn and 4.68% randomized coil. Study of the GDF9 gene would contribute to increase production efficiency and provide knowledge about fertility and genetic disorders associated with reproduction.

Keywords: GDF9, sequences, protein, goat

Introduction

Growth differentiating factor9 (GDF9) is part of the ovary-derived transforming growth factor- β (TGF β) superfamily. The genes code for proteins that are essential growth factors and receptors in follicular development in the ovaries. The genes have thus an important effect on ovulation rate and litter size (Pramod *et al.*, 2013). Ovulation rate is determined by a complex exchange of endocrine signals between the pituitary gland and the ovary. Three related oocyte- derived members of the transforming growth factor-9 (TGF-9) superfamily, namely growth differentiation factor 9 (GDF-9), bone morphogenic protein 15 (BMP15) and bone morphogenic protein-IB have been shown to be essential for ovulation rate and follicular growth (Galloway *et al.*, 2000). It is clear that GDF9 and BMP15 are essential for ovarian follicular development and normal ovulation and/ or corpus luteum formation in sheep (Hanrahan *et al.*, 2004).

The study of genes associated with fertility is important and has many applications both in the animal and human health sector. BMPs are essential for female fertility and knowledge of its function allows direct manipulation of ovulation rate and litter size in farm animals and can also provide useful information in treating infertile individuals. The study of fecundity genes is also important to gain knowledge about genetic disorders associated with reproduction (Pramod *et al.*, 2013).

The objective of this study is to investigate using in-silico method, the molecular characteristics of GDF9 protein of goats.

Materials and Methods

Sequences: A total of eight (8) sequences of GDF9 protein were retrieved from the GenBank. The GenBank accession numbers of the sequences, were, NM_001285708.1, JN601041.1, JN601040.1, XM_013965446.1, JN655693.1, JX513391.1, JX513390.1, JX483872.

Physical and chemical properties of the GDF9 gene using amino acid sequences of goats: ProtParam Tool was used for the computation of various physical and chemical properties of the GDF9 gene using amino acid sequences. The computational parameters included molecular weight, theoretical pI (isoelectric point), amino acid composition, atomic composition, extinction coefficient, estimated half-life, instability index, aliphatic index and grand average of hydropathicity (GRAVY) (Gasteiger, 2005). The amino acid sequences of GDF9 gene was further subjected to secondary structure prediction using ExPASy's SOPMA tool.

3D structure of GDF9 gene goats: The Phyre2 server was used to predict the 3D structure of GDF9 gene. The server predicts the three-dimensional structure of a protein sequence using the principles and techniques of homology modeling (Kelley & Sternberg, 2009).

Results and Discussion

The physico-chemical characteristics of proteins are predicted by ProtParam as shown in table 1. The percentage alpha helix, extended strand, beta turn and randomized coil are shown in table 2. The amino acid composition of protein sequences of GDF9 protein of goats is shown in table 3

Table 1: Physico-chemical characteristics of proteins of Sheep and goats predicted by ProtParam

No of AA	Mol. Wt (kDa)	pI	Q	Half Life	II	AI	GRAVY
453	51886.5	9.13	+ve	30hrs	55.21	76.87	-0.42

AA= amino acid; Mol. Wt= molecular weight; pI= isoelectric point; Q= net charge; II= Instability index; AI= aliphatic index; GRAVY= Grand average of hydropathicity

Table 2: Prediction of secondary structures of GDF9 proteins of sheep and goats

Alpha helix (%)	Extended strand (%)	Beta turn (%)	Random coil (%)
30.02	16.56	5.74	47.68

Table 3: Amino acid composition of the GDF9 proteins of sheep and goats

Amino acid (%)	Goats
Alanine	7.1
Arginine	6.4
Asparagine	4.0
Aspartate	4.2
Cysteine	2.9
Glutamine	3.3
Glutamate	5.7
Glycine	4.0
Histidine	3.8
Isoleucine	2.9
Leucine	11.0
Lysine	6.0
Methionine	1.5
Phenylalanine	5.3
Proline	8.4
Serine	8.8
Threonine	4.0
Trypsin	1.3
Tyrosine	4.0
Valine	5.5
Pyl	0.0
Sec	0.0

GDF9 belongs to transforming growth factor- β (TGF- β) superfamily. This oocyte-secreted paracrine factor plays a critical role during folliculogenesis (Juengel *et al.*, 2002). The GDF9 protein has been reported to influence ovulation rate in goat (Ran *et al.*, 2009). As a consequence of GDF9s ability to effect follicular growth, it follows that mutations in GDF9 might alter the reproductive potential of animals not only negatively, but also in a beneficial manner. Gene insertions and deletions have been widely acknowledged to play an essential role in the evolution of genes in vertebrates (Gemmell and Slate, 2006). Most mutations in a protein are associated with disease, only a small number of examples show the heterozygote advantage. The good example is *Bmp15* and *Gdf9* genes, which are identified to be related with female fecundity in several animals. *Bmp15* and *Gdf9* genes encode for bone morphogenic protein 15 (BMP15, also known as GDF9B) and growth differentiation factor 9 (GDF9), respectively. Mutations in *Bmp15* and *Gdf9* genes have been shown to increase ovulation rate and ultimately litter size in sheep (Hanrahan *et al.*, 2004). The amino acid sequences determine the higher structural levels of proteins (secondary, tertiary and quaternary) and specify their biological properties. (Branden and Tooze, 1991). Most genetic variation is considered neutral but single base changes in and around a gene can affect its expression or the function of its protein products Collins *et al.*, 1997)

Different amino acids play important roles in the formation of the third structure of protein, some amino acids such as alanine, glutamic acid and leucine tend to participate in α -helix and some others tend to take part in β -sheet. Variation in amino acid composition tends to change the structure of the obtained protein leading to the change in conformation of the third structure of the protein and its function. Many disease-related effects of polymorphisms involve effects on protein structure and/or function. Thus, accurate predictions of the effect of polymorphisms on the structure and function of protein gene products are essential (Wang and Moulton, 2001).

Conclusion

Analysis of amino acid composition indicates leucine as having the highest percentage (11.0%) while selenocysteine and pyrrolysine have the lowest percentage (0.0%). Secondary structure indicates 16.56% extended strand, 30.0% alpha helix, 5.74% beta turn and 4.68% randomized coil. Study of the GDF9 gene would contribute to increase production efficiency and provide knowledge about fertility and genetic disorders associated with reproduction

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