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Sequence Characterization of Alpha 1 isoform (*ATPIA1*) of Na^+/K^+ -ATPase Gene and Expression Characteristics of its Major Isoforms across Tissues of Riverine Buffaloes (*Bubalus Bubalis*)

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Abstract: Na^+/K^+ -ATPase is an integral plasma membrane protein complex which maintains the active transport of Na^+ and K^+ ions across the plasma membrane in the animal. The alpha subunit harbours the binding and catalytic site for Na^+ , K^+ and ATP hydrolysis. The alpha (α) subunit has four known isoforms; $\alpha 1$, $\alpha 2$, $\alpha 3$ and $\alpha 4$ which are encoded by *ATPIA1*, *ATPIA2*, *ATPIA3*, and *ATPIA4* genes respectively. The role of Na^+/K^+ -ATPase variants has been associated with heat stress in dairy animals but the data with respect to molecular characterization and tissue expression kinetics of its alpha isoforms is lacking in riverine buffaloes (*Bubalus bubalis*) which is one of the major dairy species in India. Hence the present work is aimed to sequence characterize the bubaline $\alpha 1$ gene and also to evaluate the tissue expression pattern of all four different isoforms ($\alpha 1$ - $\alpha 4$). Our *ATPIA1* sequence showed a similar structural organization with other mammalian species. The predicted polypeptide has 1021 amino acids showing maximum identity with *Bos taurus* (99.71%) and least with *Gallus gallus* (92.75%). 32 variations were detected in the *ATPIA1* CDS of buffalo when compared with *B.taurus*. qPCR expression analysis of *ATPIA1*, *ATPIA2*, *ATPIA3*, and *ATPIA4* genes revealed tissue specific expression. *ATPIA1* and *ATPIA2* showed ubiquitous expression in all the tissues examined while *ATPIA3* and *ATPIA4* showed tissue-specific expression confined to nervous tissue and testis respectively. Hence the present study has enriched the bubaline data and also strongly points towards the critical role that these isoforms of Na^+/K^+ -ATPase gene might be playing in maintaining the ionic concentration in the cells and tissues in which they are expressed.

Keywords: Na^+/K^+ -ATPase, alpha isoform, *Bubalus bubalis*, characterization, expression profile.

1. Introduction

The sodium-potassium adenosine triphosphatase (Na^+/K^+ -ATPase) is an integral plasma membrane protein complex which helps in active transport of Na^+ and K^+ ions across the plasma membrane in the animal cells via coupling the transmembrane transport of three Na^+ outward and two K^+ inward utilizing energy derived from ATP hydrolysis (Mobasheri et al., 2000; Blom et al., 2011; Bensimon-Brito et al., 2012). The electrochemical gradient is

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