Chinese hamster ovary (CHO) cells have the capacity to correctly fold, assemble and modify proteins post-translationally, and consequently is commonly used expression systems for recombinant therapeutic proteins. In recent years, a thorough understanding of process parameters of individual CHO cell lines have been achieved, but comprehending the genomic or pathway-specific distinction of various CHO cell lines at transcriptome level still remains a challenge. To address this challenge i.e. to gain cell line specific understanding of modulation in the pathways and gene sets, an RNA-seq study of CHOS, CHOK1 and DG44 cell lines grown in batch culture was performed using an in-house developed pipeline. An R-based CHO gene expression visualization application was developed specifically for CHO dataset to further visualize expression values across different cell lines. Further, distinction between various CHO cell lines were identified by performing differential expression analysis on some selected pathways related to metabolic and cellular processes. Consequently, most efficient cell lines were picked on the basis of process and pathway specific gene networks. Furthermore, two main conditions i.e. p-value < 0.05 and log fold change of 1 was applied to perform differential expression (DE) analysis to study gene network across the cell lines. Among the identified up- and down-regulated genes, unique and common genes across cell lines were identified. Additionally, specific pathways were found to be similarly regulated and some to be transversely regulated across various cell lines. We have thereby mapped the cell line-specific genetic regulation. This can be implemented in picking desired characters, across various CHO cell lines and in determining the structure of super CHO cell lines having the capability to combat most of the deficiencies exiting till today.