Bioinformatics Study on the Response of Human Endothelial Cells to Different Strains of Staphylococcus Aureus

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Research

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Abstract

Background

Staphylococcus aureus-induced bacteremia has an impact on human health due to its high mortality rate of 20–30%. To better study the invasion process of *staphylococcus aureus*, we conducted a study in human endothelial cells to try to find a link between the infection process and bacteremia at the molecular level.

Methods

In this study, the datasets GSE13736, GSE82036 were analyzed using R software to identify differentially expressed genes. Only the infection samples of four different strains had differential gene expression compared to the control samples. Then the GO analysis and KEGG analysis were conducted to construct a protein-protein interaction (PPI) network which shows the interaction and influence relationship between these differential genes. Finally, the central gene of the selected CytoHubba plug-in was verified using GraphPad Prism 8.

Results

There were 421 differential genes in the Strain 6850, including 64 up-regulated and 357 down-regulated; There were 319 differential genes in the Strain 8325-4, including 14 up-regulated and 305 down-regulated. There were 90 differential genes in the Strain K70058396, including 12 up-regulated and 78 down-regulated. There were 876 differential genes in the Strain K1801/10, accompanied by 261 up-regulated and 615 down-regulated. An analysis of GO and KEGG revealed that these differentially expressed genes were significantly enriched in pathways associated with immune response and cytokines; Verification of the hub gene can provide a molecular basis for studying the relationship between invasive endothelial infection and bacteremia.

Conclusions

We found specific gene expression patterns in endothelial cells in response to infection with Strain K70058396, and these central genes and their expression products (RSAD2, DDX58, IFIT3, and IFIH1) play a key role in this process of infection.

Introduction

Staphylococcus aureus, characterized by staphylococcus or clusters, is considered one of the most common and infectious pathogens^[1]. *Staphylococcus aureus* can colonize the human body and mucosa with a variety of pathogenic factors^[2], including immune escape surface factors, enzymes, α -toxin, and even causes invasive infection^[3,4].

Humans suffer from a series of diseases caused by *Staphylococcus aureus*, such as skin and soft tissue infections, infective endocarditis, pneumonia and *S. aureus* bacteremia (SAB), and so on^[2, 5, 6]. Among these infections, SAB, causing 10–30 cases per 100000 people per year with the mortality rate is as high as 25%^[7], draws a great amount of medical workers' attention^[6]. Patients with SAB exhibit a range of diseases and outcomes of varying severity. Studies have shown that some patients have eliminated pathogens in first-line treatment, while others have not resolved the infection problem. Persistent bacteremia leads to an immune response imbalance in the host and is associated with a mortality rate of 20–30% after the illness^[7]. In terms of treatment, the wild spread of antibiotic-resistant strains aggravates the disease and makes treatment more difficult.

Saureus bacteremia can be summarized as a systemic bacteria spreading disease, which may lead to systemic infection^[8]. So, it's reasonable to consider that entering and crossing through the endothelium is the vital link of *S. aureus* bacteremia^[4, 8]. Previous studies show that *Staphylococcus aureus* can influence endothelial barrier function through inducing an inflammatory response, apoptosis, microtubule destabilization, and so on^[8, 9]. However, it is well known that there are many different strains of *Staphylococcus aureus* causing staphylococcal bacteremia. Further studies, using proteomic methods^[5] and bacteriological methods^[10] to demonstrate the existence of differences in the interaction between *S. aureus* and the endothelium among different strains may facilitate an in-depth study of the molecular mechanisms underlying the infection process of the highly infectious strains.

This study intended to analyze the differences in gene expression between *S. aureus* strains with different endothelial invasion abilities. To understand the difference in gene expression between different strains of *S. aureus*, it is necessary to further understand how it infects the endothelial barrier function, to find new multi-targets to block this progress, which may provide some molecular basis for the research on antibiotic resistance. In addition, the discovery of marker gene symbols may help determine the risk of *S. aureus* bacteremia in the event of local infection, which is of auxiliary significance for clinical diagnosis and treatment.

Materials And Methods

Microarray data

We searched the GEO database (<https://www.ncbi.nlm.nih.gov/gds/>) for the appropriate data set for our study, using the keywords “*staphylococcus aureus*” and “endothelial cells”. Gene expression profiles of the GSE13736 and GSE82036 were downloaded from the GEO database for our study. Dataset GSE13736, based on the GPL570 platform [HG-U133_Plus_2] Affymetrix Human Genome U133 Plus 2.0 Array, contains samples of human umbilical vein endothelial cells (HUVEC) infected by *S. aureus* isolates from septic patients and five isolates from healthy males. As for GSE82036, it was processed on the GPL10558

platform Illumina HumanHT-12 V4.0 expression beadchip and contained 24 samples of HUVEC infected by different strains of *S. aureus*. Among the 24 samples, we selected 21 for our study (mock infection groups were excluded). The detailed information is listed in Table 1.

Identification of differentially expressed genes (DEGs)

We downloaded, calibrated and standardized the relative data files and then processed them with R package (R Foundation for Statistical Computing). The Fold Change (FC), P-Value and false discovery rate were calculated to screen DEGs between HUVEC infected by different strains of *S. aureus* and HUVEC went through mock infection. Finally, the cutoff point of DEGs was $|\log_2 \text{fold change (FC)}| > 1$ and $P\text{-value} < 0.05$. Besides, hierarchical cluster analysis was employed to show the heat map (Fig. 1) and volcano of DEGs (Fig. 2) identified above.

Functional enrichment analysis of DEGs

Gene ontology (GO) databases can help annotate genes and thus learn about relevant gene function. It contains three sections: biological process (BP), cellular component (CC) and molecular function (MF). Kyoto Encyclopedia of Genes and Genomes (KEGG) is another commonly used database that helps users learn about gene functions. In our study, we make use of the GO and KEGG databases to further investigate the function of DEGs. We performed GO analysis and KEGG analysis on strains 6850, 8325-4, K70058396, and K1801/10, respectively, and the GO analysis chart and KEGG analysis chart for each strain were listed as one chart (Fig. 3, Fig. 4, Fig. 5, Fig. 6).

PPI network construction and key module identification

The PPI network of these DEGs was constructed using the Search Tool for the Retrieval of Interacting Genes/Proteins (STRING), an online database that helps assess and integrate protein-protein interaction (PPI) information. Besides, interaction is admitted when the interaction score is > 0.4 . The protein interaction network was also adjusted and visualized by Cytoscape. Subsequently, the CytoHubba plug-in was also utilized to filter hub genes and the top 10 hub. The genes with the highest degree of connectivity within the PPI network were figured out and shown in the interaction network (Fig. 7).

Validation of gene expression levels

We found and used the data set GSE65088 in the geodatabase, and selected three samples from the control group and three samples from the infection group to verify the central genes screened by the CytoHubba plug-in, and used GraphPad Prism 8 software to produce verification maps related to the infection of four groups of *S. aureus* strains (Fig. 8).

Results

Identification of DEGs

Two datasets (GSE13736, GSE82036) were obtained from the GEO database and analyzed in the R language. A total of 7 groups of DEGs analysis were permitted, and only 4 groups showed DEGs with the stated cutoff including $|\log_2 \text{fold change (FC)}| > 1$ and $P\text{-value} < 0.05$. Those four groups are Strain 6850, Strain 8325-4, Strain K70058396 and Strain K1801/10 and they contained 421, 319, 90, 876 DEGs [respectively](#), among which 64, 14, 12, 261 were up-regulated and 357, 305, 78, 615 were down-regulated (Table 2). Volcano plot and heat map of each group of DEGs were also generated [respectively](#) (Figure 1–Figure 2).

GO functional enrichment analysis and KEGG pathway analysis of DEGs

With the help of the GO and KEGG database, we analyzed the enrichment of each group of DEGs respectively. The most significant biological processes for each group of DEGs enrichment are response and defense response to the virus, response to interferon- γ and they are shown in Figure 3,4,5,6. As for cellular components (CC), organelle outer membrane, outer membrane, proteasome core complex, beta-subunit complex are thought to be important in Strain 6850. Cytoplasmic ribonucleoprotein granule, ribonucleoprotein granule and proteasome core complex, beta-subunit complex are clearly enriched in strain Strain 8325-4. The aggregation of cellular components (CC) of Strain K70058396 is embodied in particles, such as cytoplasmic ribonucleoprotein granule, specific granule and ribonucleoprotein granule. The cell component enrichment of the Strain K1801/10 is mainly reflected in membrane raft, membrane microdomain and membrane region (Figure. 6). In addition, in terms of molecular function, for strain 6850, DEGs is mainly enriched and expressed in terms of receptor-ligand activity, signaling receptor activator activity and cytokine receptor binding; For Strain 8325-4, DEGs was mainly and significantly expressed in cytokine receptor binding, cytokine activity, and double-stranded RNA binding. For Strain K70058396, the differential genes were mainly enriched in receptor-ligand activity, signal receptor activation activity and cytokine activity. Finally, for Strain K1801/10, DEGs were significantly enriched in DNA binding transcription activator activity, RNA polymerase II specific, and DNA binding transcription activator activity. The last but not the least, the KEGG pathway analysis showed that the overlapping DEGs of each group were mainly enriched in Influenza A, NOD-like receptor signaling pathway, TNF signaling pathway, and Epstein-Barr virus infection, respectively. Through these functions enrich the analysis, we further study endothelial cells reactions toward different strains of *S. aureus*.

PPI network construction and Hub Gene analysis

PPI networks of each group was constructed and shown in Figure 7. There are 142 nodes and 698 edges in Strain 6850's PPI network, 113 nodes and 657 edges in Strain 8325-4' PPI network, 30 nodes and 168 edges in strain K70058396' PPI network and 254 nodes and 949 edges in Strain K1801/10's PPI network. What's more, we calculated the Degree using CytoHubba plug-in and found out 10 Hub genes in each PPI network and showed their interaction.

Validation of gene expression levels

We performed validation experiments on the genes screened by the CytoHubba plug-in and found that Strain K1801/10 and Strain K70058396 were significantly expressed in *S. aureus* strains that equip themselves with invasiveness to HUVEC compared to strains with no invasiveness. These significantly

expressed genes may play an important role in the invasion of endothelial cells by *S. aureus*. In addition, the central genes screened after the differential expression of *S. aureus* strain invading endothelial cells did not exactly match the hub genes that were significantly differentially expressed in the validation data set, which may further suggest that the bacteremia caused by the endothelial invasion and whole blood infection was not identical.

Discussion

Staphylococcus aureus, as the most common pathogen in human pyogenic infection, can cause local pyogenic infection or severe systemic infection. In this paper, we analyzed the gene expression of infections caused by the invasion of different strains of *Staphylococcus aureus* in human endothelium from the perspective of bioinformatics, which opened up a new idea for the molecular mechanism of disease infection and the early diagnosis and treatment in clinical practice.

In this study, we analyzed two datasets in the GEO database: GSE13736 and GSE82036. Samples with differential expression were selected for R analysis, and GO analysis and KEGG analysis were performed. We found that the cellular components, molecular functions and biological processes significantly expressed in GO analysis were related to the expression of KEGG pathways. At the same time, the central genes screened from PPI networks were significantly enriched in the related pathways and affected the expression of molecular functions and biological processes.

Strain K70058396: The different genes of Strain K70058396 mainly included RSAD2, DDX58, IFIT3, IFIH1 and so on. Among them, RSAD2 (eristostatin) is an evolutionally-conserved and interferon-induced protein, participating in the innate immune responses of cells to a variety of viruses in a variety of ways, such as regulating cellular signaling process. Especially, Epstein-Barr virus infection was significantly enriched in this study. The proteins get encoded by the Epstein-Barr virus, involved in viral replication and expression of virus particles packing and adjust the immune response process of the host cell. This may be related to the pathway through which *S. aureus* invades the endothelium to cause significant expression and functional changes of cell particles^[11]. Additionally, DDX58 is a defense reactive protein that is associated with constitutive upregulation of type I interferon^[12, 13]. Studies have shown that type I interferon increases the expression of functional tumor necrosis factor-associated apoptotic ligand (TRAIL), which may be related to the viral defense response.

IFIT3 is an IFN-induced antiviral protein, and the PPI network shows its significant expression after the invasion of human endothelial tissues^[14]. At the same time, the Influenza A, Epstein-Barr virus infection pathways enriched in KEGG may be related to the significant expression of viral genes caused by the interaction between cytokines. The enrichment of ribonucleoprotein granule in the cell fraction may be related to the cosmid of IFIT3 and ribosomes from lysates of infected cells^[15].

In addition, IFIH1 heightens antiviral response in vitro and enhanced virus control in vivo^[16]. The enrichment pathway obtained by KEGG analysis confirmed that *S. aureus* could be the main pathogen causing the spread of the influenza virus, with a possible pro-inflammatory effect^[17].

Strain 6850, Strain 8325-4 and Strain K1801/10: The major central genes expressed by Strain 6850, Strain 8325-4 and Strain K1801/10 are IFIT3 and ISG15. According to GO analysis, after *S. aureus* invades human endothelial cells, the body will initiate the biological response process to the virus and carry out self-defense. In addition, the enrichment of the tumor necrosis factor signaling pathway in the KEGG assay showed that it was active as a common response to bacterial infection, with significant upregulation of both the protein-encoding genes, IFIT3 and ISG15.

Studies have shown that IFIT2, belonging to the same family as IFIT3, can enhance the expression of factors that selectively target and inhibit viral mRNA transcription cells to invade proteins and nucleic acids into pathogens. Therefore, we speculate that the RNA bound by IFIT3 is important to enhance viral gene expression^[15].

As an interferon-induced protein, ISG15 inhibits virus replication, regulates host injury, repair response, host immunity and other signal pathways. ISG15 can be expressed under the stimulation of numerous interferons, thus encoding anti-viral mediators and establishing a cellular anti-viral state in which there is a process of signal receptor activation and interaction between cytokines. The expression is confirmed in the molecular function of GO analysis^[18].

In Strain K1801/10, MX1 is an intracellular antiviral protein that activates the type I and type III interferon signals and can inhibit viral replication by blocking the transcription of viral RNA^[19]. In viral replication, the MX1 protein interacts with viral nucleoproteins (NP) and PB2 to affect polymerase activity and provide interspecific limitations^[20]. In addition, the high expression of MX1 protein is also associated with tumor invasion^[21].

However, this study has certain limitations: for example, the highly infectious strain described in this study, Strain K70058396, was obtained by cell experiment and does not represent the strain that actually cause *Staphylococcus aureus* bacteremia in clinical practice. The sample size of each strain is small; No further wet experimental verification was performed. Central gene recognition facilitates the selection of biomarkers and targets genes for *S. aureus* invasion of endothelial cells, which may provide new ideas and directions for the early detection, control and treatment of infection in patients clinically.

Based on the above analysis, we speculate that there may be overlapping pathways between *S. aureus* and EB virus and the influenza virus in the process of infection that invade endothelial cells. Their co-infection aggravates the damage to endothelial cells, which may be related to the fact that highly invasive strains are more likely to cause bacteremia after infecting the endothelium.

Conclusion

In this study, we found that endothelial cells have differential expression in response to strains with different invasion abilities. In response to the Strain K70058396 strain infection, the differential gene expression has a certain specificity. At the same time, RSAD2, DDX58, IFIT3, and IFIH1 play a key role in the

endothelial cell response. Further studies on this basis may be beneficial for early recognition of *Staphylococcus aureus* infections and selection of action targets for treatment.

Declarations

Editorial policies

Not applicable.

Consent for publication

All of the authors have read and approved the content, and agree to submit the whole article in your journal.

Availability of data and materials

The GSE13736 and GSE82036 datasets used during the current study are available from GEO database(<https://www.ncbi.nlm.nih.gov/gds/>).

Competing interests

All the authors declare that there are no competing interests associated with the manuscript.

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Authors' contributions

Xu-Guang Guo provided the original idea and constructed the article outline. Tian-Ao Xie, Ke-Ying Fang, Wen-Chao Cao, Jie Lv, Jia-Xin Chen, Xun-Jie Cao, Heng Zhang, Yu-Wei Zhang were responsible for data analysis, table making and manuscript constructing. All the authors read and approve the final version of the manuscript.

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Footnotes

Not applicable.

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Tables

Table 1. Details of the data sources for this study.

Gene expression profile	Sample collection	Sample genetic data included	Platform	Reference
GSE13736	Human umbilical vein endothelial cells	GSM345268	GPL570 [HG-U133_Plus_2] Affymetrix Human Genome U133 Plus 2.0 Array	(Stark, Matussek et al. 2009)
		GSM345269		
		GSM345270		
		GSM345271		
		GSM345272		
		GSM345273		
		GSM345274		
		GSM345275		
		GSM345276		
		GSM345277		
GSE82036	Human umbilical vein endothelial cells	GSM345278	GPL10558 Illumina HumanHT-12 V4.0 expression beadchip	-
		GSM345279		
		GSM2181637		
		GSM2181639		
		GSM2181640		
		GSM2181641		
		GSM2181642		
		GSM2181643		
		GSM2181644		
		GSM2181645		
		GSM2181647		
		GSM2181648		
		GSM2181649		
		GSM2181650		

Table 2. Up-regulated genes and down-regulated genes that meet the screening criteria.

a) Strain 6850

DEGs	Gene terms	Count
Up-regulated	ARL4A, ARL4A, HMGCS1, SC4MOL, C14orf132, SC4MOL, C7orf49, ACAT2, ZNF792, FDFT1, DIXDC1, MARVELD2, FAM84B, NUA1, SC5DL, LSS, ZBTB16, GRRP1, PIK3IP1, FDFT1, SLC40A1, MVK, SQLE, EML1, NSDHL, SQLE, TMEM154, HMGCR, C18orf1, IDI1, ZNF395, TMCC3, GPR126, NUA1, EML1, ZBTB16, LIMCH1, DHCR7, MVD, ITLN2, IDH1, ST6GAL1, LOC158160, LOC647859, FLJ14603, RGS7BP, DIXDC1, ART4, ARMC7, TP53INP1, SPI1, PCYT2, IDH1, ACSS2, PHOSPHO2, PALMD, MEIS2, C3orf54, TMEM170B, SLC40A1, TMOD2, ST6GAL1, EFCAB4A, PCMTD2	64
Down-regulated	CXCL10, OASL, IFIT1, IFNB1, RSAD2, FOSB, CX3CL1, IFIT2, FAM46A, OASL, IFIT3, CCL5, IFIT1, IFIT3, ATF3, CCL8, EPSTI1, IFIT3, ANGPTL4, FOS, PMAIP1, KLF4, LINCR, IL6, ADAMTS1, LOC100128274, OAS2, CCL5, DDX58, BIRC3, OAS1, CXCL2, OAS1, KLF4, C12orf48, NFKBIZ, ISG15, OAS2, ISG20, HERC5, IFIH1, LOC387763, BATF2, INDO, TNFAIP3, ZC3HAV1, GBP1, IL8, PMAIP1, USP18, RTP4, IDO1, IRF1, ZC3HAV1, PTGS2, GBP1, NR4A2, PRIC285, CCL2, IL8, OAS2, IFI44L, FST, GCH1, MX2, VIP, RN7SK, CCNA1, IRF7, CMPK2, RIPK2, IFI44, CXCL1, SAMD9, CCL20, ICAM1, CD83, TAP1, C8orf4, KITLG, PTGS2, NR4A2, STX11, IRF7, RARRES3, OAS1, MSX1, CXCL9, RNU1-3, RNU1-5, RNU1G2, SLC15A3, CEBPD, BIRC3, LOC400759, GCH1, KITLG, IFIT5, TNFSF13B, DUSP19, ANGPTL4, CCRN4L, MIR155HG, ZFP36, CD83, ZC3HAV1, KIAA1683, OAS3, CREB5, SOD2, HERC6, SP110, PARP9, USP41, SP110, SP110, EGR2, TNFSF13B, CCNF1, BIRC3, SOD2, HIST1H2BK, SKIL, TRIM69, C3orf52, LOC25A28, RNF19B, CEACAM1, PARP9, EGR3, SOCS3, APOL2, GBP5, SLC7A5, CEACAM1, OAS2, FJX1, TRIB1, IFI35, TIPARP, TNFSF10, CD274, TGFB3, PMCH, HK2, PSMB9, XAF1, OAS1, DTX3L, STC2, AXUD1, HIST2H2AAA, DHDH, ZNFX1, VIP, ZC3H12C, PLAUR, KITLG, NFKBIA, SAMD9L, CNKSR3, SOCS1, DHX58, BCL6, TRIM21, RND3, PLEKHA4, DDX60L, CEACAM1, SLC25A24, C10orf54, CH25H, JUNB, MAP3K8, NUA2, ERFF1, TNFAIP8, LTA, NPTX1, LOC100131718, GJC2, ELL2, FAP, TMEM158, SP110, XAF1, LOC727895, PLEKHF1, CCL4L1, SDC4, IRAK2, PLAUR, BAMBI, JUN, IL10, NFIL3, APOL6, IL7R, BATF3, PPAP2B, RGS17, RGMB, LOC387841, OGFR, PHLDA1, TMEM217, FOXF1, DDX60, C19orf66, EIF2AK2, PHLDA1, TLR3, PPM2C, PPAP2B, GCH1, FGF2, PARP10, DUSP1, MLKL, DUSP5, CCL7, ARID5A, PARP12, NKX3-1, NAV3, PLSCR1, MCL1, IL15RA, HIST1H2BK, MLKL, PTP4A2, APOL3, PPM1K, MCL1, TRIM69, AADA1, HAS2, OAS3, RGMB, TMEM31, ID2, LOC100132564, NUA2, B3GNT5, USP36, SPAG9, RNU1A3, PPARG, ADAMTS18, RNF149, RNU11, ARAP2, C1orf107, EGR1, RELB, IRF9, SOCS3, CCNTA1, ICAM4, RNF148, ART3, RNF114, CRY1, LRFN5, LOC650832, MLKL, UBE2L6, RNU6-1, RPPH1, PATE2, SP100, FRMD3, AADA1, RNF149, CRY1, ELF1, UBA52, ZC3H12A, DLC1, MAP2K3, DUSP16, PPP1R15A, CASP7, PDZD2, LOC653158, CYP4B1, CBX4, JAK2, TSC22D2, AKAP2, DCP1A, MYD88, KCNJ2, PMP22, TRIM26, RNF213, PNPT1, LOC645993, CD68, SLC35E1, FRMD5, PSMB8, PRR16, NRIP3, FBXO6, CEP27, RNF114, HIST2H2AA3, LOC100130053, TAC3, KLHL6, NLF2, LOC643031, MIR302C, PAPD5, KCNN2, TSC22D2, LOC346887, FGF2, LRRN3, TDRD7, RNS59, LOC732371, CASP7, KLF2, NFKBIB, KIAA1666, ACTA2, FRMD5, RNU1F1, RNU6-15, EID3, C10orf2, IRF2, CDK6, LOC728835, C21orf63, LOC100129502, FLNC, CHN2, FAM133B, IFI30, LRRFIP2, CRCP, ZNF223, BCL3, ABL2, LBA1, PSMB8, PPM1K, ZNF786, PPARG, PANX1, PMP22, PHF15, PTPLAD2, NOV	357

b) Strain 8325-4

DEGs	Gene terms	Count
Up-regulated	C7orf49, CDK5R1, ITLN2, PLD6, TSHZ1, TBC1D4, DDIT4, GRRP1, EPS8L1, MN1, PIK3IP1, RASSF9, MEX3B, DCAF6	14
Down-regulated	CXCL10, OASL, RSAD2, IFIT1, INDO, CCL8, IFIT3, IFIT3, IDO1, FAM46A, OASL, EPSTI1, IFNB1, IFIT2, CX3CL1, MX2, OAS1, OAS1, CCL5, OAS2, DDX58, CXCL9, BATF2, IFI44L, TNFSF13B, RARRES3, GBP1, IFIH1, ATF3, OAS2, RTP4, IFIT3, USP18, ISG20, ISG15, OAS1, TNFSF13B, GBP1, SLC15A3, CMPK2, PLA1A, LOC400759, PRIC285, HERC5, IFIT1, OAS2, IRF7, IFI35, OAS3, SAMD9, IRF7, IRF1, SAMD9L, DDX60L, TAP1, SP110, IL18BP, IFITM1, GCH1, CCL5, ZC3HAV1, APOL2, PARP9, SP110, PSMB9, SP110, PMAIP1, XAF1, GBP5, HERC6, TNFSF10, IFI44, MSX1, TLR3, USP41, DHX58, ZC3HAV1, SP110, PARP9, IFIT5, DDX60, XAF1, PARP10, OAS1, SOCS1, TLR3, DTX3L, GCH1, DUSP19, ART3, APOL3, CCL2, SLC25A28, IL15, ICAM1, SP100, NFKBIZ, ANGPTL4, TRIM21, C19orf66, IFI30, PARP14, PPM1K, RNF19B, OAS2, CEBPD, RIPK2, APOBEC3G, ZNFX1, LAMP3, PARP12, APOL6, CCRN4L, GBP4, MLKL, PLSCR1, CBR3, OAS3, PMAIP1, FBXO6, IL15RA, CD274, MLKL, ZC3HAV1, UBE2L6, JAK2, KIAA1751, LBA1, EIF2AK2, STAT2, MOBK2C, APOL1, MLKL, TRAFD1, LOC389386, PSMB8, OGFR, STAT1, HESX1, CASP1, MYD88, LOC732371, APOBEC3G, CASP1, PHF15, PSMB8, TRIM22, LAP3, FAM46C, UNC93B1, PARP10, SP100, TAP2, CD68, TDRD7, RNF114, CEACAM1, BCL6, LOC389386, LOC643384, RBM43, ASPHD2, PARP10, IRF9, CEACAM1, BATF3, SOCS3, PLEKHF1, STAT1, STX11, TRIM25, GBP2, CEACAM1, PSMB8, IRF8, TRIM38, C10orf140, APOBEC3F, PHF11, LOC728216, CD7, RNF114, PNPT1, TIPARP, IRF2, LY6E, CASP7, GMPR, CD47, LOC100131718, CASP7, LOC650799, PI4K2B, FAS, ANKFY1, LGALS9, TRIM56, CBX4, TMEM62, ZBTB42, PHF11, MASTL, MOV10, STAT1, IRF2, RGS20, LGALS9, NLRC5, TTC39B, FTSJD2, RAB24, TRIM26, RIPK1, PCGF5, CD47, BTN3A1, ADAR, IFI16, RAB24, NUB1, LRCH2, SLC35E1, PPM1K, XRN1, GTF2B, HOXD8, GCNT1, PI4K2B, PML, UBE2L6, DCP1A, ADPRHL2, LOC730417, AKAP7, ADAR, FGF2, AIM2, TESK2, MCL1, FAM26F, UGCG, APOBEC3F, ELF1, CYP2J2, PDZD2, GSDMD, MCL1, CXorf38, USP42, LGALS8, NCOA7, LGALS8, SUOX, IL12A, LOC154761, NEDD1, TRIM78P, GPR180, SCYL3, PRR16, HES4, SQRDL, N4BP1, PPM2C, ZFYVE26, SOCS3, PARP11, MT1M, HK2, SP100, FHL3, PSMB10, SLFN12, PCNX, PATL1, MASTL, SUOX, B3GALNT1, WHAMM, RBCK1, TMEM16A, LYSMD2, GTPBP2, TNFSF18, RBCK1, RNF149, NT5C3, MAA, TEAD4, OTUD4, CCDC117, GPBP1, NT5C3, TRIM5, FLJ39827, RELB, NBN, CCND3, GPBP1, RNF149, HOXB4, ANKIB1, GOLM1, LOC654346, TLE4	305

c) Strain K70058396

DEGs	Gene terms	Count
Up-regulated,	ARL4A, ARL4A, REEP1, ITLN2, LIMCH1, KIAA0182, MARVELD2, ST6GAL1, YPEL1, CRISPLD1, CSGALNACT1, PCMTD2	12
Down-regulated	IFIT1, CXCL10, OASL, FOSB, IFIT2, IFIT3, ANGPTL4, RSAD2, CX3CL1, CXCL1, IFIT3, FAM46A, OASL, CCL2, ISG15, OAS2, OAS1, OAS1, CCL5, DDX58, PRIC285, LOC387763, IFIT1, IFIT3, ANGPTL4, PTX3, PMAIP1, LOC100128274, IRF1, IRF7, USP18, BATF2, IFIH1, NFKBIZ, RTP4, OAS1, ZC3HAV1, KIAA1683, IFI44, IRF7, TAP1, VIP, STX11, ZC3HAV1, SAMD9, SLC7A5, CCRN4L, C3orf52, JUN, DKK1, MSX1, PARP9, GCH1, RND3, TRIB1, SP110, LOC201175, NFKBIA, DLC1, PLAUR, PHLDA1, FRMD3, PARP9, KITLG, CMPK2, XAF1, DTX3L, SOCS1, SLC25A28, TRIM21, MAP2K3, MAFF, GJC2, LOC650832, CCL5, GCH1, LTA, SLC30A1	78

d) Strain K1801/10

DEGs	Gene terms	Count
Up-regulated	<p>ARL4A, C14orf132, SLC40A1, ARL4A, C7orf49, DIXDC1, RAB11FIP1, NAP1L3, MARVELD2, ARHGAP28, TSPYL3, C18orf1, SFRS7, KBTBD7, ARHGAP28, LARGE, FLRT2, GNAI1, TERF1, CSGALNACT1, SPIN4, AGGF1, REEP1, HSPA2, LOC401720, SLC40A1, C1orf59, ZNF135, NUAKE1, DIXDC1, TSHZ1, FAM78A, KLHL12, LOC728661, ATP2B1, TRIM2, GNAI1, PIGM, TP53INP1, VPS36, PABPC4L, GUCY1A3, RBM12, TMEM19, RGS7BP, ARL4A, ZNF512, ZNF828, GPAM, ZNF696, HOXA9, SLC35E3, TP53INP1, TMOD2, C3orf54, MAN1C1, CBR4, KIAA0232, TMOD2, GRRP1, C20orf177, KIAA0430, ZNF252, IVNS1ABP, NUAKE1, C17orf97, GUCY1A3, ZNF518A, ABHD6, TRIM13, C17orf58, KCTD18, PDIK1L, LOC400657, ST6GAL1, MKL2, N6AMT1, RNF170, C16orf53, ZNF30, CBX2, FAM13B, PRMT6, EHHADH, TNFRSF1B, FLJ41603, LOC643888, LOC644860, KLHL9, SLC24A1, ZNF792, INSIG2, DACH1, HSPA2, IVNS1ABP, DOLK, PLD6, B3GALT6, TTPAL, PAFAH1B1, C17orf58, C17orf58, CCDC121, KLHL12, ZNF167, ZNF302, CRISPLD1, SLC39A11, C5orf54, LOC647346, PKD2, KBTBD6, GPAM, BRWD1, EML1, LRRC33, PCMTD2, C2orf64, ZFP3, C16orf30, SNORA12, RCBTB2, ITLN2, SLC39A10, A2LD1, PEX11B, BBS10, ZNF788, LOC728661, CLDN12, RNF141, ALDH5A1, THNSL1, C14orf167, SLC2A10, COL3A1, C5orf51, PPAPDC2, ZNF285A, FAR1, LOC728006, FBXL2, AP3M2, PRKAA1, C5orf5, MAPK1, MBLAC2, PRRC1, ZNF680, MEIS2, RIPK5, ARHGEF6, NLRX1, LOC642342, SRL, USP13, LIMCH1, CRISPLD1, EEF2K, ZFP112, SORBS1, GSPT2, CSTF2T, TSPYL5, KLRG1, LPIN2, BEND5, THNSL1, C10orf6, ZNF69, HNRNPA0, C4orf18, DICER1, MAGEE1, C6orf85, PTPRE, FAM84B, LOC644799, APBB3, ANKRD46, HOXA10, LOC151162, ST3GAL2, BUDT18, TMCC3, ZNF395, GPRASP2, OSBPL2, CCDC126, BTBD12, FLJ20444, RBM4B, PIGW, AKAP11, BIVM, CYB561D1, C5orf51, AHCTF1, MEGF9, ZNF260, DOPEY1, FNBP1L, CPT2, PRRT3, FAM50B, USP47, PDS5B, SLC29A3, FAM173B, ZNF33B, ANGEL2, C22orf29, SYNMM, WWP1, ZNF706, KBTBD7, PPM1H, LRRC20, SRBD1, PPCS, VAMP2, TMEM55A, PHOSPHO2, PDPR, PFAAP5, AR, BMP2K, SRI, RAB22A, SMARCA1, MDP1, C9orf40, SLC35B4, DICER1, GMCL1, C20orf11, KIAA1370, GMCL1, MAPK6, ARMC7, TUBA1A, FAM120B, TGFB3, CENPBD1, ATP9A, C14orf28, ASTE1, EPM2AIP1, ZNF706, MANSC1, POLR3B, ZNF658, PAQR8, WDR37, ATP7B, GCNT2, CBY1, SRI, ZFP30, RP5-1022P6.2</p>	261
Down-regulated	<p>OASL, CXCL10, IFNB1, OASL, RSAD2, IFIT1, CCL5, FOSB, FAM46A, LOC100128274, ATF3, IFIT3, CX3CL1, IFIT2, FOS, LINCR, IFIT3, KLF4, CXCL2, CCL8, IL6, IFIT1, IFIT3, ANGPTL4, RN7SK, CCL5, INDO, PMAIP1, TNFAIP3, KLF4, OAS1, EPSTI1, OAS1, IDO1, BIRC3, SELE, OAS2, CXCL1, ISG15, NFKBIZ, RND1, DDX58, BATF2, IL8, C8orf4, GEM, HERC5, MX2, CXCL9, LOC387763, ISG20, ZFP36, PTGS2, RNU1G2, RTP4, RNU1-3, IFIH1, MIR1974, GEM, RNU1-5, IRF7, OAS2, PMAIP1, CCL20, IRF7, USP18, GBP1, RN7SK, MX1, ZC3HAV1, ICAM1, IL8, IFI44L, IRF1, CCL2, GBP1, RARRES3, OAS1, NR4A2, CMPK2, ADAMTS1, ZC3HAV1, JUNB, PTGS2, AXUD1, RIPK2, MIR155HG, PRIC285, GBP5, ANGPTL4, TRIM69, CH25H, BIRC3, DUSP19, CD83, CCNA1, CCRN4L, SOCS3, NR4A2, TAP1, SNORD3D, STX11, NUAKE2, LOC400759, LOC100008588, KIAA1683, SOD2, MSX1, BCL6, GCH1, GCH1, IFI44, SAMD9, DHDH, SLC15A3, IFITM1, USP41, BIRC3, CD274, CEBPD, OAS3, DDX60L, FAP, EGR2, SKIL, OAS2, KIAA1751, JUN, CD83, GADD45B, HERC6, SP110, OAS1, HIST1H2BD, MAP3K8, RNF19B, TNFSF10, LOC653610, TNFSF13B, IFI35, PARP9, PLAUR, PDZD2, DTXL3, BAMBI, KIAA1666, ICAM4, SP110, NFKBIA, EGR3, LOC727895, PLEKHA4, ZC3HAV1, F3, VIP, APOL2, SOCS1, F3, CEACAM1, TRIB1, TRIM69, CEACAM1, RNU6-1, PHLDA1, SLC25A24, TNFSF13B, SDC4, C14orf4, LOC100131718, SP110, CEACAM1, HIST2H2AA3, FXJ1, RRAD, IFIT5, IRF8, PHLDA1, IL10, SP110, DHX58, RNU6-15, XAF1, ART3, SLC25A28, NUAKE2, ARID5A, APOL2, IFIT1L, SAMD9L, BATF3, HIST2H2AA4, IRAK2, MIR1267, PTP4A2, CXCR7, PSMB9, RNU1A3, PPP1R15A, NEDD9, TIPARP, RNU11, SP100, HIST2H2AC, EGR1, PARP9, ERRF1, FOXF1, ID2, INSIG1, TMEM217, NLF2, MTE, ID2, XAF1, RPPH1, DDX60, CALD1, SOCS3, RNU1F1, ZC3H12C, C19orf66, ZC3H12A, PLAUR, C3orf52, ZNFX1, TNFSF15, MAFF, FAM175A, PLSCR1, PPM1K, IL18BP, CREB5, PARP10, SIK1, SNORD3A, LOC653158, CCNL1, PROX1, DUSP1, OAS2, APOL6, CCNL1, CCL7, CNP, PTX3, TRIM13, TLR3, TRIM26, RN5S9, ANGPTL4, SNORD3C, PARP14, APOL3, GBP4, IL15RA, LTA, HAS2, SERPINE1, TRIM21, NCRNA00152, HIST1H2BG, TRIM69, SLC1A3, PTGER4, NFIL3, PNPT1, RRAD, AADACL1, EGR4, MCL1, USP36, LOC643031, WARS, RND3, PARP12, DUSP2, LOC401317, SLC35E1, HIST1H2BK, LOC100129502, LOC440900, LOC648852, PIM1, CEP27, PATE2, OGFR, EIF2AK2, CD69, BCL3, RELB, CLK1, NKX3-1, CBX4, LOC387841, LOC100132564, LOC100130053, LOC389765, ZNF223, CD68, SLC25A37, SERTAD1, LOC100132727, CNKSR3, CYP4B1, FAM80B, TSC22D2, MLKL, LOC100134322, KIAA1143, BHLHB2, LOC645993, RNF213, TNFAIP8, KCNF1, PPFBP1, DCLRE1C, OAS3, FLJ36131, VIP, TSC22D2, ANKRD37, C2CD4B, MLKL, RAD51, GCH1, ZNF682, BCOR, RIPK1, EID2B, WARS, ATF3, SPAG9, CD7, PPM1K, LOC100132391, GEM, LOC441763, RNF114, FGR, FOSL2, GDF15, IFI30, ZNF669, DTWD2, PPM2C, ASPHD2, LOC644250, MAFF, MCL1, LOC644852, DENR, HIVEP1, MASTL, SLC4A5, NLRP8, C21orf63, TNFSF14, KLF10, KCNE1L, SPOCD1, IER5L, TNFAIP2, CDKN2AIPNL, OVGP1, PAPD5, SMAD7, AKAP2, DCP1A, TLR3, PML, KCNJ2, MLKL, GJC2, LOC401098, DLC1, LOC648921, PLEKHF1, SIK1, ACTA2, FAM153B, SMAD7, ID1, C9orf80, MIR302C, LOC732371, SPAG9, FLJ40722, FOSL1, SGK1, IRF2, RAXL1, LOC100129362, TGFB3, CSF2RA, ZNF786, CYCSL1, C2CD4A, IL1A, FLJ35390, CCDC125, LOC729120, ZNF34, SP100, BCL10, TMEM217, RNF148, N4BP2, LOC653086, ARID5A, DUXAP3, PLAUR, HES1, HSPA6, C9orf130, YIPF4, FLJ25363, FAM40B, HK2, TRIM56, KITLG, LOC645452, ARC, AIRE, PTPLAD2, ANKRD10, NFKB2, PSMB8, RNF152, RUNDC2C, MOBKL2C, FAM133B, PFKFB3, NEDD1, CRCP, DNHD1, LOC100130598, KLF6, APOL1, ARAP2, CATSPER2, LGALS9, AADACL1, CPEB3, NME1, GSDMB, RNF149, IRF2, MAFF, LOC732450, KLF2, IL12A, LOC389386, TNFSF15, LOC154761, TTC39B, PSMB8, LEP, LBA1, MYD88, SLC2A3, ZNF738, TMEM158, C5orf28, USP49, LOC649841, DKFZp434K191, TRIM38, GCG, JAK2, CEBPB, ALPP, SGK, SGMS2, LOC728620, FBXO6, IER2, ELF4, HSCB, UBA52, MC1R, HIVEP2, LOC729603, C14orf43, FGF2, DUSP10, MAP4, ZNF14, DIRAS3, FLJ22536, KLF6, WDR43, TDRD7, SLC7A5, KLHL28, GNB4, LOC730993, COX19, SOX8, ZNF652, PHF15, LOC646996, ATF4, ABL2, BBC3, TICAM1, PPAP2B, SPHK1, PIP5K2B, GMPR, ANKRD30B, SGK1, LDHA, NXF1, ARHGAP20, TICAM1, RHBDL2, ZNF430, UGCG, OSGIN2, FOXC2, F2RL3, SDHALP1, ANKRD10, STAT1, UBE2L6, WTAP, IRF9, LOC728779, SHCBP1, LOC100130445, NXF1, SMOX, OTUD1, SMOX, HOXD8, ATF2, ENC1, SLC7A2, PML, ICA1, ERN1, CASP7, DUSP16, LOC731789, PCDHB9, PLA2G2D, LOC100133888, LOC100131544, CDAN1, NR2F1, TNFRSF12A, INSIG1, UTX, PPA2, LOC389517, HEY2, IPPK, LOC389386, FLJ27255, KLF10, KLHL6, TEAD4, C13orf33, LOC23117, SSTR2, RGM8, KLF11, SLC44A4, SULT1A1, MIDN, LAMP3, SNHG3-RCC1, PKX, ZNF486, TMEM106A, C11orf63, GTF2B, PNPT1, MCM8, TSC22D1, PFKFB3, NCOA7, LOC650832, NFKB2, OTUD4, LOC100133772, TRAFD1, NUBPL, DAPP1, UBD, CDKN1A, HLA-C, ETV3, RNF114, MASTL, UNC93B1, FBXL7, LOC728153, TDP1, SIAH2, PSME1, C16orf52, HIAT1, LOC729978, GPBP1, NAMPT, RASSF1, BTG3, LOC143666, LOC391019, ZNF697, NFKBIB, LOC440093, LOC100133950, IER5</p>	615

Table 3 : Strain 6850 : The collection of significantly enriched GO and KEGG pathways (top 5 according to P-value).

Section	ID	Description	Count	P-value	Gene ID
BP	GO:0009615	response to virus	46	1.84E-32	ACTA2/BIRC3/BCL3/CDK6/GBP1/IFIT2/IFIT1/IFIT3/IFNB1/IL6/CXCL10/IRF1/IRF2/IRF7/ISG20/MX2/MYD88/OAS1/OAS2
BP	GO:0051607	defense response to virus	39	5.97E-30	BIRC3/GBP1/IFIT2/IFIT1/IFIT3/IFNB1/IL6/CXCL10/IRF1/IRF2/IRF7/ISG20/CXCL9/MX2/MYD88/OAS1/OAS2
BP	GO:0060337	type I interferon signaling pathway	23	4.10E-22	EGR1/IFI35/IFIT2/IFIT1/IFIT3/IFNB1/IRF1/IRF2/IRF7/ISG20/MX2/MYD88/OAS1/OAS2
BP	GO:0071357	cellular response to type I interferon	23	4.10E-22	EGR1/IFI35/IFIT2/IFIT1/IFIT3/IFNB1/IRF1/IRF2/IRF7/ISG20/MX2/MYD88/OAS1/OAS2
BP	GO:0034340	response to type I interferon	23	1.14E-21	EGR1/IFI35/IFIT2/IFIT1/IFIT3/IFNB1/IRF1/IRF2/IRF7/ISG20/MX2/MYD88/OAS1/OAS2
MF	GO:0008009	chemokine activity	11	1.32E-10	CXCL1/CXCL2/CXCL10/CXCL9/CCL2/CCL5/CCL7/CCL8/CCL20/CX3CL1/CCL4L1
MF	GO:0005125	cytokine activity	20	1.48E-10	FGF2/CXCL1/CXCL2/IFNB1/IL6/IL10/CXCL10/LTA/KITLG/CXCL9/CCL2/CCL5/CCL7/CCL8/CCL20/CX3CL1/CCL4L1
MF	GO:0005126	cytokine receptor binding	22	4.40E-10	CXCL1/CXCL2/IFNB1/IL6/IL10/CXCL10/JAK2/LTA/KITLG/CXCL9/MYD88/CCL2/CCL5/CCL7/CCL8/CCL20/CX3CL1/CCL4L1
MF	GO:0042379	chemokine receptor binding	11	3.87E-09	CXCL1/CXCL2/CXCL10/CXCL9/CCL2/CCL5/CCL7/CCL8/CCL20/CX3CL1/CCL4L1
MF	GO:0003725	double-stranded RNA binding	10	1.81E-07	OAS1/OAS2/OAS3/EIF2AK2/TLR3/OASL/DDX58/DDX60/IFIH1/DHX58
KEGG	hsa00900	Terpenoid backbone biosynthesis	6	6.74E-07	ACAT2/HMGCR/HMGCS1/IDI1/MVD/MVK
KEGG	hsa00100	Steroid biosynthesis	5	9.99E-06	DHCR7/FDFT1/LSS/SQLE/NSDHL
KEGG	hsa04010	MAPK signaling pathway	13	0.000425892	MAP3K8/DUSP1/DUSP5/FGF2/FLNC/FOS/JUN/KITLG/MYD88/MAP2K3/RELB/TGFB

Table 4 : Strain 8325-4: The collection of significantly enriched GO pathways (top 5 according to P-value)

Section	ID	Description	Count	P-value	Gene ID
BP	GO:0009615	response to virus	59	1.56E-54	ADAR/GBP1/IFI16/IFIT2/IFIT1/IFIT3/IFNB1/IL12A/IL15/CXCL10/IRF1/IRF2/IRF7
BP	GO:0051607	defense response to virus	52	3.46E-52	ADAR/GBP1/IFI16/IFIT2/IFIT1/IFIT3/IFNB1/IL15/CXCL10/IRF1/IRF2/IRF7/ISG20
BP	GO:0034341	response to interferon-gamma	41	7.00E-40	CASP1/CD47/GBP1/GBP2/GCH1/ICAM1/IRF8/IRF1/IRF2/IRF7/JAK2/LGALS9/OAS1
BP	GO:0071346	cellular response to interferon-gamma	38	2.15E-37	CASP1/CD47/GBP1/GBP2/ICAM1/IRF8/IRF1/IRF2/IRF7/JAK2/LGALS9/OAS1/OAS2
BP	GO:0034340	response to type I interferon	30	7.71E-35	ADAR/GBP2/IRF8/IFI35/IFIT2/IFIT1/IFIT3/IFNB1/IRF1/IRF2/IRF7/ISG20/MX2/MX3
MF	GO:0003725	double-stranded RNA binding	11	1.63E-09	ADAR/OAS1/OAS2/OAS3/EIF2AK2/TLR3/OASL/DDX58/DDX60/IFIH1/DHX58
MF	GO:0003950	NAD+ ADP-ribosyltransferase activity	7	5.18E-09	ART3/TIPARP/PARP14/PARP11/PARP12/PARP9/PARP10
MF	GO:0005126	cytokine receptor binding	16	4.49E-07	IFNB1/IL12A/IL15/CXCL10/JAK2/CXCL9/MYD88/CCL2/CCL5/CCL8/CX3CL1/ST2
MF	GO:1990404	protein ADP-ribosylase activity	5	6.87E-07	TIPARP/PARP14/PARP11/PARP12/PARP10
MF	GO:0016763	transferase activity, transferring pentosyl groups	7	1.48E-06	ART3/TIPARP/PARP14/PARP11/PARP12/PARP9/PARP10
CC	GO:0000932	P-body	9	3.21E-07	MOV10/TRIM21/XRN1/DCP1A/APOBEC3G/MEX3B/TRIM5/APOBEC3F/PATL1
CC	GO:0036464	cytoplasmic ribonucleoprotein granule	12	3.68E-06	MOV10/TRIM21/SOCS1/TDRD7/XRN1/DCP1A/APOBEC3G/NFKBIZ/MEX3B/TRIM5
CC	GO:0035770	ribonucleoprotein granule	12	6.17E-06	MOV10/TRIM21/SOCS1/TDRD7/XRN1/DCP1A/APOBEC3G/NFKBIZ/MEX3B/TRIM5
CC	GO:0019774	proteasome core complex, beta-subunit complex	3	0.000195076	PSMB8/PSMB9/PSMB10
CC	GO:0061702	inflammasome complex	3	0.000420115	CASP1/AIM2/GSDMD

Table 5 : Strain K70058396 The collection of significantly enriched GO pathways (top 5 according to P-value).

Section	ID	Description	Count	P-value	Gene ID
BP	GO:0009615	response to virus	20	4.49E-20	IFIT2/IFIT1/IFIT3/CXCL10/IRF1/IRF7/OAS1/OAS2/PMAIP1/CCL5/OASL/ISG15/IFI44/DDX58/
BP	GO:0051607	defense response to virus	18	1.21E-19	IFIT2/IFIT1/IFIT3/CXCL10/IRF1/IRF7/OAS1/OAS2/PMAIP1/OASL/ISG15/DDX58/ZC3HAV1/R
BP	GO:0060337	type I interferon signaling pathway	12	4.74E-16	IFIT2/IFIT1/IFIT3/IRF1/IRF7/OAS1/OAS2/OASL/ISG15/USP18/XAF1/RSAD2
BP	GO:0071357	cellular response to type I interferon	12	4.74E-16	IFIT2/IFIT1/IFIT3/IRF1/IRF7/OAS1/OAS2/OASL/ISG15/USP18/XAF1/RSAD2
BP	GO:0034340	response to type I interferon	12	7.94E-16	IFIT2/IFIT1/IFIT3/IRF1/IRF7/OAS1/OAS2/OASL/ISG15/USP18/XAF1/RSAD2
MF	GO:0008009	chemokine activity	5	1.12E-06	CXCL1/CXCL10/CCL2/CCL5/CX3CL1
MF	GO:0042379	chemokine receptor binding	5	5.00E-06	CXCL1/CXCL10/CCL2/CCL5/CX3CL1
MF	GO:0003725	double-stranded RNA binding	5	9.40E-06	OAS1/OAS2/OASL/DDX58/IFIH1
MF	GO:0005125	cytokine activity	7	1.93E-05	CXCL1/CXCL10/LTA/KITLG/CCL2/CCL5/CX3CL1
MF	GO:0048018	receptor ligand activity	9	8.05E-05	CXCL1/CXCL10/LTA/KITLG/CCL2/CCL5/CX3CL1/VIP/DKK1

Table 6 : Strain K1801/10 The collection of significantly enriched GO and KEGG pathways (top 5 according to P-value).

Section	ID	Description	Count	P-value	Gene ID
BP	GO:0009615	response to virus	60	1.74E-31	ACTA2/BIRC3/BCL3/FGR/GBP1/IFIT2/IFIT1/IFIT3/IFNB1/IL6/IL12A/CXCL10/IRF1/IRF
BP	GO:0051607	defense response to virus	49	5.55E-28	BIRC3/GBP1/IFIT2/IFIT1/IFIT3/IFNB1/IL6/CXCL10/IRF1/IRF2/IRF7/ISG20/CXCL9/MX
BP	GO:0034340	response to type I interferon	29	1.30E-21	EGR1/HLA-C/IRF8/IFI35/IFIT2/IFIT1/IFIT3/IFNB1/IRF1/IRF2/IRF7/ISG20/MX1/MX2/M
BP	GO:0060337	type I interferon signaling pathway	28	5.43E-21	EGR1/HLA-C/IRF8/IFI35/IFIT2/IFIT1/IFIT3/IFNB1/IRF1/IRF2/IRF7/ISG20/MX1/MX2/M
BP	GO:0071357	cellular response to type I interferon	28	5.43E-21	EGR1/HLA-C/IRF8/IFI35/IFIT2/IFIT1/IFIT3/IFNB1/IRF1/IRF2/IRF7/ISG20/MX1/MX2/M
MF	GO:0005126	cytokine receptor binding	30	3.94E-09	CXCL1/CXCL2/IFNB1/IL1A/IL6/IL10/IL12A/CXCL10/JAK2/LTA/SMAD7/KITLG/CXCL9
MF	GO:0005125	cytokine activity	25	1.61E-08	FGF2/CXCL1/CXCL2/IFNB1/IL1A/IL6/IL10/IL12A/CXCL10/LTA/KITLG/CXCL9/CCL2/C
MF	GO:0001228	DNA-binding transcription activator activity, RNA polymerase II-specific	35	2.28E-07	AIRE/AR/ATF3/ATF4/CEBPB/CEBPD/KLF6/ATF2/EGR1/EGR2/EGR4/ELF4/FOXF1/FO
MF	GO:0001216	DNA-binding transcription activator activity	35	2.41E-07	AIRE/AR/ATF3/ATF4/CEBPB/CEBPD/KLF6/ATF2/EGR1/EGR2/EGR4/ELF4/FOXF1/FO
MF	GO:0008009	chemokine activity	10	1.78E-06	CXCL1/CXCL2/CXCL10/CXCL9/CCL2/CCL5/CCL7/CCL8/CCL20/CX3CL1
KEGG	hsa04010	MAPK signaling pathway	20	0.000182062	ATF4/MAP3K8/ATF2/DUSP1/DUSP2/FGF2/FOS/HSPA2/HSPA6/IL1A/JUN/KITLG/MY

Figures

Figure 1

Heat map of the DEGs in HUVEC infected by four *S. aureus* strains. The abscissa axis represents sample types and the ordinate axis represents gene names. Red indicates log2FC>0; green indicates log2FC<0; the color deepens as log2FC's absolute value enlarges. (a) Strain 6850; (b) Strain 8325-4; (c) Strain K70058396; (d) Strain K1801/10.

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Figure 2

Volcano plot of the DEGs in HUVEC infected by four *S. aureus* strains. Red indicates $\log_2FC > 1$ while blue indicates $\log_2FC < -1$, which means different gene expressions between two groups (red for up-regulated DEGs and blue for down-regulated DEGs). Gray indicates no difference. (a) Strain 6850; (b) Strain 8325-4; (c) Strain K70058396; (d) Strain K1801/10.

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Figure 3

Strain 6850's bubble diagram results of GO analysis KEGG pathway analysis. The abscissa axis represents Gene Ratio while the ordinate axis represents term names. The size of a single bubble represents the degree of enrichment; the color variety represents different q-value (those bubbles with red color are considered to be of significance). (a) Top 10 enriched GO terms in Biological Process (BP); (b) Top 10 enriched GO terms in Cellular Component (CC); (c) Top 10 enriched GO terms in the Molecular Function (MF); (d) Top 10 of enriched KEGG pathways.

Strain 6850

Figure 3

Strain 6850's bubble diagram results of GO analysis KEGG pathway analysis. The abscissa axis represents Gene Ratio while the ordinate axis represents term names. The size of a single bubble represents the degree of enrichment; the color variety represents different q-value (those bubbles with red color are considered to be of significance). (a) Top 10 enriched GO terms in Biological Process (BP); (b) Top 10 enriched GO terms in Cellular Component (CC); (c) Top 10 enriched GO terms in the Molecular Function (MF); (d) Top 10 of enriched KEGG pathways.

Strain 8325-4

Figure 4

Strain 8325-4's bubble diagram results of the GO analysis and KEGG pathway analysis. The abscissa axis represents Gene Ratio while the ordinate axis represents term names. The size of a single bubble represents the degree of enrichment; the color variety represents different q-value (those bubbles with red color are considered to be of significance). (a) Top 10 enriched GO terms in BP; (b) Top 10 enriched GO terms in CC; (c) Top 10 enriched GO terms in the MF; (d) Top 10 of enriched KEGG pathways.

Strain 8325-4

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Strain 8325-4's bubble diagram results of the GO analysis and KEGG pathway analysis. The abscissa axis represents Gene Ratio while the ordinate axis represents term names. The size of a single bubble represents the degree of enrichment; the color variety represents different q-value (those bubbles with red color are considered to be of significance). (a) Top 10 enriched GO terms in BP; (b) Top 10 enriched GO terms in CC; (c) Top 10 enriched GO terms in the MF; (d) Top 10 of enriched KEGG pathways.

Figure 5

Strain K70058396's bubble diagram results of GO analysis and KEGG pathway analysis. The abscissa axis represents Gene Ratio while the ordinate axis represents term names. The size of a single bubble represents the degree of enrichment; the color variety represents different q-value (those bubbles with red color are considered to be of significance). (a) Top 10 enriched GO terms in BP; (b) Top 10 enriched GO terms in CC; (c) Top 10 enriched GO terms in the MF; (d) Top 10 of enriched KEGG pathways.

Figure 5

Strain K70058396's bubble diagram results of GO analysis and KEGG pathway analysis. The abscissa axis represents Gene Ratio while the ordinate axis represents term names. The size of a single bubble represents the degree of enrichment; the color variety represents different q-value (those bubbles with red color are considered to be of significance). (a) Top 10 enriched GO terms in BP; (b) Top 10 enriched GO terms in CC; (c) Top 10 enriched GO terms in the MF; (d) Top 10 of enriched KEGG pathways.

Figure 6

Strain K1801/10's bubble diagram results of the GO analysis and KEGG pathway analysis. The abscissa axis represents Gene Ratio while the ordinate axis represents term names. The size of a single bubble represents the degree of enrichment; the color variety represents different q-values (those bubbles with red color are considered to be of significance). (a) Top 10 enriched GO terms in BP; (b) Top 10 enriched GO terms in CC; (c) Top 10 enriched GO terms in the MF; (d) Top 10 of enriched KEGG pathways.

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Strain K1801/10's bubble diagram results of the GO analysis and KEGG pathway analysis. The abscissa axis represents Gene Ratio while the ordinate axis represents term names. The size of a single bubble represents the degree of enrichment; the color variety represents different q-values (those bubbles with red color are considered to be of significance). (a) Top 10 enriched GO terms in BP; (b) Top 10 enriched GO terms in CC; (c) Top 10 enriched GO terms in the MF; (d) Top 10 of enriched KEGG pathways.

Figure 7
Protein-protein interactions (PPI) network and hub gene identification. Nodes represent genes; lines represent interactions between gene-encoded protein. (a) Strain 6850; (b) Strain 8325-4; (c) Strain K70058396; (d) Strain K1801/10.

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Figure 8

Validation of hub genes expression level. Genes are the top 10 selected from DEGs and PPI networks co-regulated by control and infected samples. (a) Strain K1801/10; (b) Strain 6850; (c) Strain 8325-4; (d) Strain K70058396.

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