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TRANSCRIPTIONAL RESPONSE OF THE MULTIDRUG RESISTANT *Staphylococcus aureus* FOLLOWING FS-1 EXPOSURE

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ABSTRACT

The aim of this study was to evaluate transcriptional regulation in a model multidrug resistant *Staphylococcus aureus* (MRSA) culture, *S. aureus* ATCC® BAA-39™, after exposure to a new antibacterial drug, FS-1. Growth media were supplemented with FS-1 at the end of the lag phase and in the middle of the exponential growth phase. To investigate the immediate effects of the iodine-containing antibacterial complex FS-1, 24 RNA samples were obtained 5 min after exposure to the drug.RNA sequencing revealed substantial changes in gene expression in the lag phase and, to a lesser degree, in the exponential growth phase. Analysis of the regulated genes suggested that FS-1 primarily targeted microbial cell wall proteins. The effect of FS-1 on *S. aureus* gene expression was similar, but not identical, to differential gene regulation profiles caused by several cell wall-disrupting antibiotics including bacitracin, D-cycloserine, and oxacillin. However, the gene regulation profiles were not completely congruent, suggesting that FS-1 displays a significant level of antimicrobial activity specificity. Therefore, it is possible that combining FS-1 with other anti-staphylococcal antibiotics will result in a cumulative synergetic effect.

Keywords: *S. aureus*, antibiotic resistance, FS-1, RNA, gene expression

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INTRODUCTION

Incidences of nosocomial infections caused by multidrug resistant *Staphylococcus aureus* have steadily increased in the recent years worldwide. Notable among them are the methicillin-resistant strains, which also can be resistant to other main classes of the modern antibiotics. Frequency of methicillin-resistant *S. aureus* (MRSA) isolates varies from 6-10% to 50%in different hospitals depending on the used culture isolation procedures and utilized antibiotic regiments [1].

S. aureus is a classic example of purulent post-surgical nosocomial infection agents, which threaten the public health system worldwide mainly because of the emergence of MRSA. Use of antibiotics, which is often irrational in many hospitals, causes outbreaks and fast distribution of the MRSA infection outwards the clinics to the community. Numerous cases of so-called community-acquired or personally acquired MRSA have been reported [2]. Currently, the methicillin-resistant staphylococci occupy a leading position among the most dangerous hospital and community based infectious outbreaks, which are difficult to control by conventional sanitary measures. MRSA became a common problem of intensive care, neonatal and cardiac surgery units even in highly reputable hospitals of developed countries. With an expansion of the spectrum of used antibiotics, bacteria have broadened their resistance patterns by acquiring additional genetic determinants of drug resistance. Nowadays, vancomycin resistant *S. aureus* (VRSA) are terribly frequent now among clinical isolates [1]. The extended resistance of staphylococci to antibiotics makes the generally accepted therapeutic regiments ineffective that increases the death toll of the nosocomial infections. The mortality rate from MRSA is 2.5 fold higher than that caused by susceptible strains. Also, it was reported that the death likelihood of patients infected with MRSA is 64% higher than in the case of the drug-susceptible forms of staphylococcal infections. The damage caused by MRSA infections to the patients' health and the growing costs of the treatment of these infections in clinics threaten the country's stable economic development and effectiveness of the healthcare system. Every year this problem is becoming more acute all over the world. Expansion of the staphylococcal infections requires development and introduction to the medical practice of principally new approaches to tackle the problem of the extended drug resistance [3-4].

The problem of the rapid emergence of pathogenic microorganisms resistant to multiple antibiotics is aggravated enormously by the crisis in the development of new drugs. For example, 18 new antibiotics had been introduced in total in the world in 1980-1984; but in the same time span of four years in 2010-2014,only one new antibiotic has been developed [5]. Passing through all the stages of development of new drugs from laboratory experiments to the market is a time consuming and costly process. Only the laboratory experiments may take a decade or longer and passing of a new antibiotic from the laboratory to the market takes in average more 12 years and costs from 50 million to 1 billion USA dollars to pharmacological companies. These numbers have an upward drift due to an increasing complexity of new antibiotics and shortening of the time of applicability of the antibiotics in clinics due to the rapid emergence of drug resistant pathogens [5].

In this work, we present an innovative iodine-containing medicinal complex FS-1. Iodine is a unique pharmaceutical substance characterized by a strong antimicrobial activity and versatile effects on biological systems. Because of these properties, iodine is widely used for a preparation of various dosage forms.All pathogens are sensitive to iodine.No acquired resistance to iodine-containing drugs has been ever reported. Having penetrated into cells, iodine interacts with the amino groups of proteins, thereby suppressing vital enzyme systems [6,7].Therefore, iodine is considered as a leading antisepticexerting antimicrobial, antiviral, and antimycotic activities through a local resorptive action.

Applicability of the iodine is generally limited due to chemical irritation of the skin and mucous membranes by this compound. Consuming of a single dose of 2-3 g of iodine is lethal to humans [6]. However, it was reported that in combination with polysaccharides and other biopolymers, the toxicity of iodine is significantly reduced, while the antiseptic propertiesare getting prolonged [7, 8]. An elaborated framework of polysaccharides, proteins and other polydentate ligands serve in the drug FS-1 as a vehicle to deliver iodine molecules to inflammation sites [9]. This drug has been developed in Kazakhstan by the JSC Scientific Center for Anti-Infectious Drugs (SCAID) for the treatment of drug-resistant forms of bacterial pathogens. Clinical trials have demonstrated a significant clinical efficacy of the drug, in particular for the treatment of tuberculosis with a broad spectrum of drug resistance [10].However, mechanisms of the therapeutic activity of FS-1 remain unclear. To address this issue, innovative technologies of massive parallel pyrosequencing of RNA samples were used in this work to discover an immediate effect of FS-1 in first 5 min of exposure on a model organism – collection MRSA strain *S. aureus* ATCC® BAA-39™. The experiment was carried out in different growth phases.

MATERIALS AND METHODS

Culture cultivation and determination of the minimum inhibitory concentration (MIC) of the drug FS-1

Staphylococcus aureus ATCC® BAA-39™was obtained from the American Type Culture Collection (ATCC) and stored at minus 80°C. Before the use in the experiments, the strain has been twice passaged on Müller-Hinton medium (MHB) (Himedia). Evaluation of the antibacterial activity of FS-1 against this bacterial culture was carried out in microarray plates by using the serial dilution method in MHB medium (Himedia) in the range of concentrations of FS-1 from 2000 μg to 100 μg of the active substance per 1 ml of the solution [11-13]. Overnight culture growth was 100 times diluted before inoculation. Inoculated plates were incubated at 37°C for 24 hours. The minimum inhibitory concentration (MIC) was defined as the drug concentration completely suppressing the growth of the microorganism. The absence of the growth was checked visually and then by plating of aliquots of the solutions to count colony forming units. Sterility of the initial solutions of FS-1 has been controlled.

Determination of *S. aureus* growth rate

Diluted overnight culture (1:100) of *S. aureus* was inoculated in MHB medium and then cultivated at 37°C on a shaker at 50-60 rpm. Recording of the optical density (OD) of the suspension at an optical light wave of 600nm was carried out every 5 min for 24 hours by Multiscan Ascent spectrophotometer (Thermo, Finland). The growth curve was estimated based on the obtained series of OD values by Baranyi and Roberts model implemented in an interactive DMF it application [14].

Cultivation of *S. aureus* with FS-1

FS-1 was injected into test tubes with *S. aureus* inoculants incubated for2.5 and 9 hours at 37°C (the end of the lag phase and the mid of the exponential growth phase, respectively) to achieve the final concentration of 450 μg/ml that corresponded to the 1/2 of the drug MIC. After incubation of the cultures supplemented with FS-1for more 5min, all metabolic processes in the cells were stopped by the killing buffer (2.0 ml of 1 MTris-HCl, pH 7.5; 0.5 ml of 1 M $MgCl_2$, 1.3 g of NaN_3; 997.5 ml of water) added in the ratio of 1:1 [12,15]. For a negative control, the same volume of physiological saline was applied to the *S.aureus* cultures. All experiments were performed in six replicates.

Isolation and purification of RNA

Isolation of the total RNA was performed by the RiboPure Bacteria Kit (Ambion, Lithuania) according to the developer's guidelines. The quality and quantity of the resulted RNA were determined using the NanoDrop 2000s spectrophotometer (Thermo Scientific, USA) at optical wavelengths of 260 and 280 nm. Purification of the total RNA from ribosomal RNA was carried out using the MICROBExpress Bacterial mRNA Purification Kit (Ambion, Lithuania), as recommended by the developer. The efficacy of the template RNA purification was determined on the Bioanalyzer 2100 (Agilent, Germany) by the RNA 6000 Nano LabChip Kit (Agilent Technologies, Lithuania). Quality of the samples was considered acceptable if no 23S and 16S ribosomal RNA peaks were observed.

RNA library preparation and sequencing

Library of RNA fragment was prepared by an enzymatic fragmentation using the Ion Total RNA Seq Kit V2 (Life Technologies, USA). Barcoding of the constructed library was carried out by the Ion Xpress RNA-Seq Barcode 01-16 Kit (Life Technologies, USA), according to the manufacturer's instructions. RNA sequencing was performed using the Ion Torrent PGM sequencer (Life Technologies, USA) with the Ion 318 Chip Kit V2.

Analysis of obtained RNA reads

Alignment of obtained RNA fragments (reads) was performed against the reference genome using the QIAGEN CLC Workbench 7.0.3 software package. RNA sequencing data was used for a quantitative analysis of the gene expression by calculating RPKM values (Reads per kilobase per million) [16]. The role of protein-coding genes in the metabolism of *S. aureus* was investigated by using the Pathway Tools 19.0 software package and the Biocyc database [17].

RESULTS

In previous experiments, the antimicrobial activity of the drug FS-1 was demonstrated on 114 clinical isolates of *S. aureus.* Among them, there were 58 methicillin-resistant (MRSA) and 56 methicillin-susceptible (MSSA) strains. Suppression of the growth of both resistant and susceptible *S.aureus* isolates by FS-1 was recorded [18]. MIC values varied from 620 to 2540 μg/ml from strain to strain. The differences in sensitivity between MSSA and MRSA were statistically unreliable.

Within the framework of the presented study, the gene expression levels of were determined after 5min incubation of the model *S. aureus* strain with FS-1 in different phases of the culture growth. The study was conducted to identify possible mechanisms of the antibacterial action of FS-1 on *S. aureus*.

The minimum inhibitory concentration (MIC) of the drug FS-1 against the tested strain was about 900μg/ml. The working concentration of FS-1 in this experiment was 450 μg/ml.

Growth phases of the cultured *S. aureus* were determined by using an experimentally estimated growth curve on the Mueller-Hinton broth by the Baranyi and Roberts model [14]. The correlation parameter R^2 of the established model was 0.962. Then the late lag phase and the middle of the exponential growth phase of *S. aureus* strain BAA-39 were estimated by the developed model as 2.5 and 9 hours, respectively. Cultures for inoculation were prepared by three successive passages of *S. aureus* for 20-hour of cultivation at 37°C until achievement of the stationary phase.

The time span of 5min of the exposure of bacterial cultures to FS-1 was chosen to determine an immediate effect of the drug on the bacterial metabolism. After 5min of incubation, all metabolic processes in bacterial cells were stopped by the killing buffer and then the total RNA was extracted from the samples. Obtained RNA molecules were purified from rRNAand then sequencing by the Ion Torrent PGM machine. A total of 24 RNA samples was obtained including the control samples, in which the same volume of the physiological saline was added to the medium instead of FS-1.

Quality of the resulted RNA sequence fragments(reads)was controlled using the CLC Genomics Workbench 7.0.3 software by the following parameters: range of the read lengths; nucleotide composition of the reads (GC-content); statistical confidence of base calling on average for each read and for every individual base position. Trimming of the reads was carried out to ensure the confidence level of base calling above 95%.Then the obtained reads were aligned against coding sequences of *S. aureus* ATCC® BAA-39™ used as the reference genome sequence. Coding sequences included all the protein coding genes and also as the genes of known short regulatory non-coding RNA (ncRNA) [19].

Alignment of the reads and comparison of obtained gene expression profiles were carried out using algorithms implemented in the CLC Genomics Workbench 7.0.3 software. To visualize the results of comparison of the gene expression profiles, an in-house script written on Python 2.5 was used. This script processed the outputs from CLC Genomics Workbench 7.0.3 to build volcano plots of the differential genes expression under the experimental conditions versus the control conditions. Additionally, the script summarized the gene expression data against the metabolic functions of the regulated genes. The latter data were obtained for *S. aureus* BAA-39 from the Biocyc database by using the Pathway Tools 19.0 software package [17]. Positive and negative regulation of several general metabolic pathways under the influence of FS-1 was presented in a form of histograms.

To evaluate the accuracy of the obtained gene expression profiles, a relative level of expression was estimated for every gene as a ratio of observed over expected numbers of reads aligned to the sequences of each transcribed gene:

[observed number of reads] / [expected number of reads]

Expected number of reads was calculated from an assumption of a uniform level of expression of all the transcribed genes taking into account differences in their lengths. Calculated values of the relative gene expression were above 1 for highly expressed genes and in the range from 0 to 1 for the genes with low or moderate levels of gene expression. Then they obtained patterns of the relative expression values were compared to the literature data on genes of *Staphylococcus* with a constant gene expression level and to the data on the gene expression regulation at different stress conditions [20-22].If the sequencing depth is sufficient, it was assumed that reads aligned against the permanently expressed genes must be present in all experiments independently on the experiment conditions.

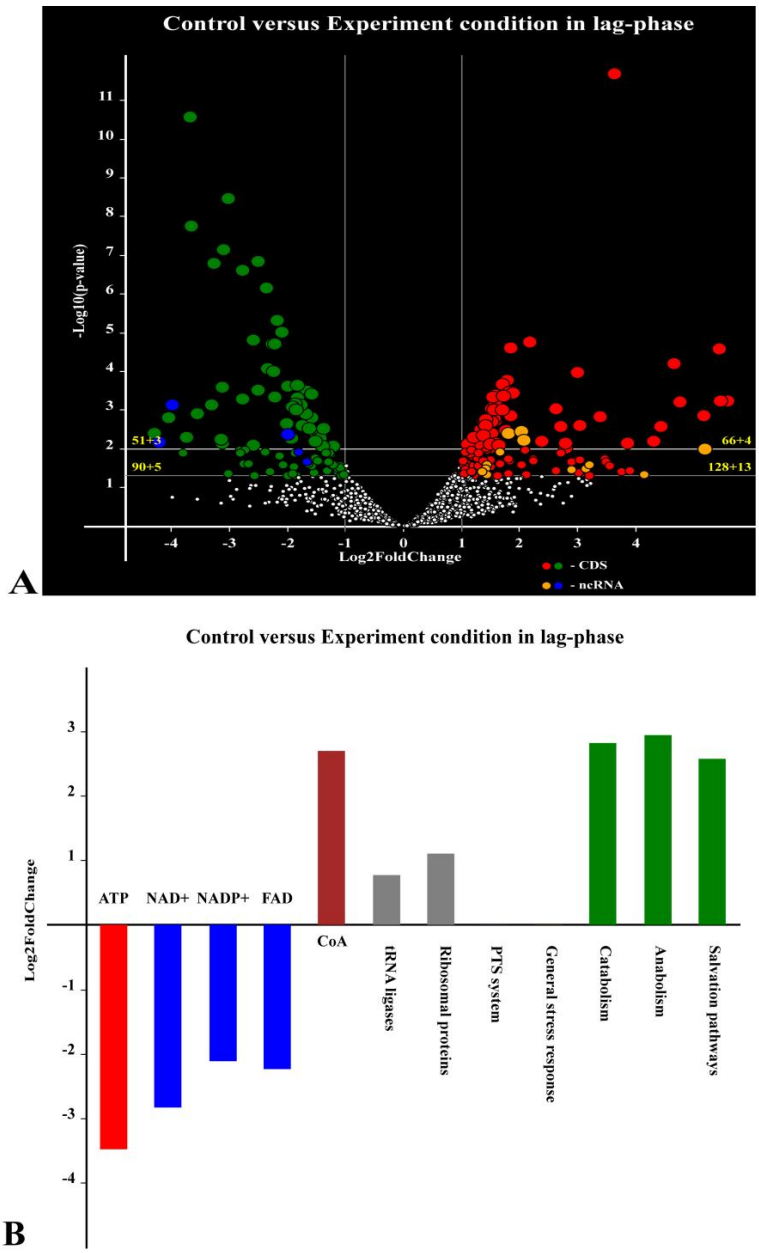
The highest expression level was characteristic for the genes encoding for enzymes of the citric acid cycle (*pdhABC* gene operon) and for DNA polymerase that is in agreement with the literature data. Reads aligned against the constantly expressed genes *dnaB* and *gyrA* (replication), *rho* (transcription termination factor), *fabD* (fatty acid biosynthesis) and *glmM* (glycolysis) were detected in all the experiments. It was concluded that the sequencing depth in this experiment was sufficient for an unbiased evaluation of the gene expression regulation. Reliability of the observed difference in the overall gene expression under the influence of FS-1 compared to the control condition was confirmed by the Fisher's criterion.

Analysis of changes in *S. aureus* gene expression profiles under the influence of FS-1 at different growth phases

Gene expression regulation by FS-1 was studied at the end of the lag phase and in the mid of the exponential growth phase. In the lag phase, the gene expression profile was affected by FS-1 to a greater level than during the exponential growth. We hypothesized that at the beginning of the culture growth the natural gene expression level was still low and the 5 min incubation of the culture with FS-1 could impact the gene expression stronger than during the exponential growth when many genes are over-expressed.

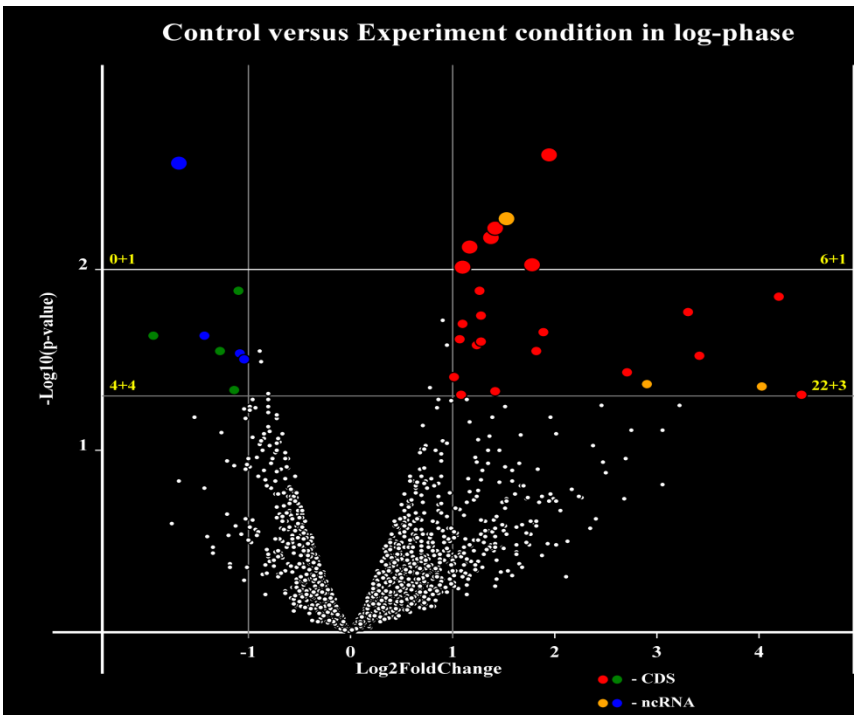
Genes with the altered expression in the lag phase are depicted in figure 1.In total, 128 protein-coding genes and 13 ncRNAs were induced by FS-1; while 90 genes and 5 ncRNAs were inhibited at this condition. The bacteria responded to the FS-1 addition by an activation of the transcription of ribosomal proteins, activation of tRNA synthesis, synthesis of cell wall fatty acids and by an increased ATP consumption. The balance of redox-controlling cofactors was shifted towards the synthesis of reduced forms of NADH, NADPH2, and FADH2, probably in order to withstand an oxidative stress. This assumption was also confirmed by anobserved activation of superoxide radical reducing enzymes. Glycolysis and the citric acid cycle were partially suppressed. Purine salvage pathways were activated that indicated an increased nucleotide catabolism.

Gene expression regulation by FS-1 in the exponential growth phase is summarized in figure 2. Statistically significant changes in gene expression were detected only for 26 protein-coding genes and 7 regulatory ncRNAs. However, the overall trends of the gene expression regulation was congruent to that in the lag phase.



A) Volcano plot of differentially regulated genes. Axis X depicts Log2 values of the fold change of gene expression under the experimental conditions versus the control conditions. Axis Y depicts the negative logarithm of the estimated *p*-value of the observed difference in gene expression. Differentially expressed genes represented on the plot by colored circles are depicted by a color code explained in the legend. The confidence of the differential expression of genes above the top horizontal line was ≥99%, and those in between horizontal lines was ≥95%. Numbers of differentially regulated protein coding genes + ncRNA genes are shown above the lines. B) Histogram of activation and suppression of major metabolic pathways.

Fig. 1. Gene and metabolic pathway regulation in the lag phase of growth of *S. aureus* in 5 min after injection of 450 μg/ml of FS-1



Meanings of the axes and the color code were the same as in figure 1A.

Fig. 2. Volcano plot of the gene regulation in the exponential growth phase of *S. aureus* in 5 min after injection of 450 μg/ml of FS-1

The analysis of functions of the regulated genes suggested that bacteria responded primarily to an increased oxidative stress damaging bacterial cell walls, cytoplasmic proteins and, to some extent, the DNA. Eight genes encoding chaperone proteins and several DNA repair proteins were activated. Dealing with the oxidative stress involves an activation of energy (ATP) consuming processes in the cells. To save the energy, the synthesis of cofactors and secondary metabolites was inhibited. Resulted shortage of the cofactors and purines was compensated by an induction of the salvage pathways. Pathways of synthesis of amino acids, ribosomal proteins, and tRNA ligases also were induced by FS-1that indicated an increased synthesis of protein such as the chaperones, but also to replace the damaged proteins. Genes encoding for several cell wall proteins such as antigens and pathogenicity factors of *S. aureus*, in particular – hemolysin, were inhibited by FS-1. The meaning of this inhibition may consist in a reduction of accessory cell wall proteins serving as substrates for the halogenation by iodine molecules released from FS-1.

Additionally, an activation of some plasmid and phage related genes in the bacterial genome was observed. This may indicate unfavorable changes in the cell homeostasis, which usually leads to an induction of the mobile genetic elements looking for an escape.

An activation of the nucleotide synthesis was also observed in the experimental gene expression profiles. However, this activation was not associated with the replication or the active growth, since the proteins controlling the cell growth and division were suppressed by the drug. Increased demand for nucleotides was possibly associated with the generally increased level of the RNA turnover and, probably, with the DNA reparation processes.

Gene expression in living organisms is controlled by transcriptional regulators and also by induction and/or inhibition of regulatory ncRNAs. Recent publication introduced a large number of ncRNAs in the *S. aureus* genome, although the role of these ncRNAs in the gene regulation of *S. aureus* remains unclear [19]. Application of FS-1 induced the transcription of 13ncRNAs (Sau7006, Sau73, sprG1, sRNA118, sRNA126, sRNA130, sRNA209, sRNA217, sRNA263, Teg25, Teg36as, Teg66, and Teg77), whereas 4 ncRNAs were suppressed by the drug in both experiments (4.5S, Sau6648, sRNA117 and sRNA342). This study has demonstrated that the ncRNAs can play an important role in controlling the response of *S. aureus* to the abiotic stresses and antibiotics.

DISCUSSION

Regulation of the gene expression in *S. aureus* in response to different stressors including several antibiotics has been reported in a number of publications [23, 24]. These studies examined an impact of high and low temperatures of cultivation, nutrient deficiency in the medium, SOS response to DNA ruptures and the effect of certain antibiotics. The profile of the gene expression alterations caused by FS-1 was rather specific and only slightly overlapped with the transcription profiles induced by the other stress factors. In particular, no one gene of the SOS-response activated in *S. aureus* by the DNA damaging antibiotic mitomycin C [23] was induced by FS-1. Moreover, FS-1 supplementation has inhibited the master SOS response regulators such as the *recA*, *lexA*, and *uvrB*. The observed induction of several DNA repair genes may indicate a DNA damaging by FS-1due to oxidation or halogenation of nucleotides. However, this damaging did not go to the level of the SOS response activation.

No overlap in the gene regulation by FS-1 and by the low temperature stress was observed. However, four FS-1activated genes were also induced by the heat shock. Among them, there were *bglA*6-phospho-beta-glucosidase, mercury-dependent reductase, *hla* alpha-hemolysin precursor and a conserved protein encoding gene. On the otherh and, four other genes induced by the heat shock, –*clcP* ATP-dependent protease, *tetR* tetracycline resistance protein, an extracellular protein transporter, and a conserved protein gene,– were suppressed by FS-1.

More commonalities were observed in the regulation profiles induced by FS-1 and by the stringent response in *S. aureus*. The stringent response in bacteria is triggered by an accumulation in the cytosol of uncharged tRNA molecules due to a shortage of nutrients (i.e. amino acids), or because of a sharp increase of protein synthesis, or under an influence of several antibiotics inhibiting aminoacyl-tRNA synthetases [25]. A total of 8 common genes were found, which were induced by both the FS-1 drug and in an experimentally induced amino acid starvation [23]. Among these genes, there were: *leuC* isopropyl malate dehydratase, *ilvD*dihydroxy-acid dehydratase, ABC phosphonic acid transporter, mercury-dependent reductase and several other genes of unknown functions. However, five conserved genes of unknown functions were suppressed by FS-1 but activated during the stringent response.

The biggest number of co-regulated genes was observed in the patterns of gene regulation induced by FS-1 and by the cell wall degrading antibiotics bacitracin, D-cycloserine and oxacillin. A total of 11 genes induced by these antibiotics and by the drug FS-1 were recorded: *topA* DNA topoisomerase, *copA* copper-transporting ATPase, *czrB* cation efflux protein, *hsp3*3 heat shock protein,*asp23* protein shock alkalization protein and several other genes of unknown functions. Four antibiotic-induced genes were suppressed by FS-1: *atl* autolysin, *but A* acetoin reductase, and two other genes encoding transmembrane proteins of unknown functions.

The performed transcriptional analysis suggested that the drug FS-1 affected primarily the bacterial cell wall integrity and also it could cause a chemical damage to cytoplasmic proteins and, to a lesser extent, to the bacterial chromosomal DNA. To repair the damaged cell wall and proteins, bacteria activated the synthesis of amino acids and molecules necessitated for replacement of the damaged proteins. Particularly, the genes for ribosomal proteins and tRNA ligases were strongly activated. Accumulation of uncharged tRNA molecules due to an active consumption of acylated tRNA in the protein synthesis caused the stringent response in bacteria. Energy deficiency has slowed down the growth and division of the bacterial cells and also it inhibited the synthesis of important cofactors and purines. The latter deficiency has been partially compensated by the activation of the salvage pathways. Bacteria also have reduced synthesis of several accessory protein constituents of the cell wall including important antigens and pathogenicity factors.

CONCLUSION

Analysis of the transcriptional regulation in *S. aureus* exposed to an immediate impact of the drug FS-1 confirmed that the molecular iodine and iodine-containing drugs primarily damage the microbial cell wall and cellular proteins [6,7]. In 5 min after the injection of the drug to the medium, bacterial cells responded by the activation of protein synthesis to replace the damaged molecules and to synthesize chaperones necessitated to maintain the protein integrity under stress conditions. Metabolic processes were regulated to promote the synthesis of the reduced forms of the cofactors NADH, NADPH2 and FADH2 to cope with the oxidative stress. Influence of the drug FS-1 on the metabolism of *Staphylococcus* resembled some extent the effect of the cell wall damaging antibiotics bacitracin, D-cycloserine and oxacillin [24].However, the patterns of the gene regulation under these conditions were not absolutely congruent. It suggested that the effect of the drug FS-1 on bacterial cells was rather specific. This specificity makes it possible to combine this drug with other antibiotics to achieve a cumulative antimicrobial effect.

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ФС-1 ПРЕПАРАТЫМЕН ӨҢДЕУ ЖАУАБЫНА, МУЛЬТИРЕЗИСТЕНТТІ *Staphylococcus aureus* ТРАНСКРИПЦИЯЛЫҚ ТАЛДАУ ЖАУАБЫ

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ТҮЙІН

ФС-1 бактерияға қарсы препарат әсерімен *S.aureus* өсіндісінің гендер экспрессиясының деңгейін бағалау - зерттеудің мақсаты болып табылады. ФС-1 препаратын ортағақосқаннан кейін 5 минуттан сон өсінді өсімінің экспоненциальдық сатысы ортасында лаг-фаза соңында алынған РНҚ үлгілерінің 24 үлгісі секвенирленді. Бактериалдық өсіндіні лаг-фаза өсімінде ФС-1 препаратымен 5 минут бойы өңдеу, гендер экспрессиясының саласын айтарлықтай өзгертті. Бұл уақытта 5нк РНКмен 90 гендер осы жағдайда басылып қалса, препараттың қосылуы мен 13 нкРНКжәне 128 кодталған-белок жекешелей көтерілді. ФС-1 препаратын қосу кезінде гендер экспрессиясы деңгейі экспоненциалды өсу фазасында шектеулі болды. 26 кодталған - белок гендері мен 7 реттеушін кРНКөзгерістербайқалды. Гендер экспрессиясы саласындағы талдау өзгерістері, молекулалы иод пен йодқұрамдас дәрілік заттар ең алдымен микроорганизмдердегі жасуша клеткаларының қабырғасының белоктарын зақымдайтынын көрсетті. ФС-1 препаратының әрекеті *S. aureus* антибиотиктер бацитрацин, D-циклосерин мен оксациллин әсерін еске түсіреді, бірақ бұл ұқсастық толық емес, яғни ФС-1 препаратының микробқа қарсы ерекшелігінің жоғары екеніне көз жеткізеді. Мұндай ерекшелік нақты препараттың басқа антибиотиктермен микробқа қарсы әсерінің куммулятивтілігіне үйлесімді екенін көрсетеді.

Негізгі сөздер: *S. aureus*, анбиотиккетұрақтылық, ФС-1 препараты, РНҚ, секвенирлеу, гендер экспрессиясы.