Review Article

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The capture of host cell's resources: The role of heat shock proteins and polyamines in SARS-COV-2 (COVID-19) pathway to viral infection

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Abstract: The exposure of organisms and cells to unfavorable conditions such as increased temperature, antibiotics, reactive oxygen species, and viruses could lead to protein misfolding and cell death. The increased production of proteins such as heat shock proteins (HSPs) and polyamines has been linked to protein misfolding sequestration, thus maintaining, enhancing, and regulating the cellular system. For example, heat shock protein 40 (Hsp40) works hand in hand with Hsp70 and Hsp90 to successfully assist the newly synthesized proteins in folding properly. On the other hand, polyamines such as putrescine, spermidine, and spermine have been widely studied and reported to keep cells viable under harsh conditions, which are also involved in cell proliferation, differentiation, and growth. Polyamines are found in all living organisms, including humans and viruses. Some organisms have developed a mechanism to hijack mammalian host cell machinery for their benefit like viruses need polyamines for infection. Therefore, the role of HSPs and polyamines in SARS-CoV-2 (COVID-19) viral infection, how these molecules could delay the effectiveness of the current treatment in the market, and how COVID-19 relies on the host molecules for its successful infection are reviewed.

Keywords: heat shock proteins, polyamines, SARS-CoV-2, drug targets, human hosts

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Introduction

The discovery of heat shock proteins (HSPs) dates back as many decades ago when it was observed that heat shock produced chromosomal puffs in the salivary glands of fruit flies (*Drosophila*) [1]. HSPs are ubiquitously found in almost all living organisms studied so far. The DNA sequence that makes up this family of genes is highly conserved across species. This family of genes originally was named because of their expression after exposure to heat. However, the genes are now known to be induced by a wide variety of environmental or metabolic stresses that include the following: anoxia, ischemia, heavy metal ions, ethanol, nicotine, surgical stress, and viral agents. The recent coronavirus outbreak has drawn a lot of attention in research as a matter of urgency to come up with effective vaccines or drugs. Molecules such as RNA, DNA, and proteins are a starting point in drug development and the therapeutic industry in general [2].

Therefore, understanding how HSPs and polyamines assist viruses in developing infection and in their differentiation and growth is vital in coming up with the effective treatment of Coronavirus disease 2019 (COVID-19). It has been widely reported that viruses compete with the host's molecules for their survival, growth, and successful infection. Most organisms, including viruses like coronavirus, require HSPs to protect the cellular system and balance its proteomic system. In short, HSPs act as housekeepers in the cellular system of the viruses for them to acclimatize to the host (new environment) [3]. COVID-19 has affected millions of people worldwide, and it continues to be a threat to many human beings, especially those with a compromised system [4,5]. Moreover, the emergence of new coronavirus strains adds more pressure to the science world, working hard to find a cure that can successfully curb the spread of coronavirus. Therefore, both HSPs and polyamines show molecular chaperone activities in the most studied organisms thus far. Briefly, polyamines are ubiquitous molecules known

for their involvement in cell differentiation, proliferation, and growth. Polyamines are present in various organisms and play a vital role in their survival. These positively charged molecules interact with numerous molecules of DNA, proteins, and RNAs, thus playing a part in protein synthesis. The precursors of polyamine biosynthesis are driven by both S-adenosylmethionine decarboxylase (AdoMetDC) and ornithine decarboxylase (ODC) to form molecules such as putrescine, spermidine, and spermine [6–10]. In plants, like any other organisms, polyamines act as molecular chaperones in the cellular system exposed to unfavorable conditions such as heat shock. Their role as thermotolerant has been widely studied in plants, especially for crop production, and has been reported to improve plant growth in both the biotic and abiotic environments [11]. In organisms such as Plasmodium, polyamine biosynthesis is controlled by a unique structure that links both AdoMetDC and ODC, but they function independently, whereas, in humans, these two molecules are not joined together. Other studies have suggested that polyamines are involved in or influence the synthesis of HSPs to strengthen molecular chaperone activities that are taking place inside the cellular system [12–15]. However, the mechanism behind this is not yet known. In this review, we outline the role of HSPs and how they cooperate as molecules of different sizes. We also look at the role of polyamines in the survival of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in humans. In addition, how these molecules can contribute to a vaccine or drug development against the COVID-19 pandemic.

COVID-19 is a disease triggered by a novel coronavirus called SARS-CoV-2 (formerly called 2019-nCoV). COVID-19 was first identified amid an outbreak of respiratory illness cases in Wuhan City, Hubei Province, China. It was initially reported to the World Health Organization (WHO) on December 31, 2019 [5]. On January 30, 2020, the WHO declared the COVID-19 outbreak a global health emergency. On March 11, 2020, the WHO declared COVID-19 a global pandemic, its first such designation since declaring the H1N1 influenza pandemic in 2009. Today, more than 179,686,071 confirmed cases with 3,899,172 deaths have been reported worldwide. This pandemic has led many companies, both big and small, to shut down. Consequently, thousands of people have lost their jobs and income due to coronavirus [4,5]. As well as being a human tragedy, the COVID-19 pandemic has been an unprecedented economic shock for the world economy. The International Monetary fund estimates global output to have fallen by 3.5% in 2020, and all countries – big and small, rich, and poor – have been hit. There is now an increasingly clear route to bring the immediate crisis to an end by relying on a mass vaccination program unprecedented in speed and scale. However, implementation remains complex and risky, while the long-term consequences of the pandemic – impacts on health, acquisition of skills, and the accelerated spread of technology – remain highly uncertain [5].

The role of human HSPs

HSPs are the main cluster of evolutionarily conserved molecular chaperones that play an important role as housekeepers of the cell. HSPs are grouped into two main families centered on their molecular weight. The first group is small ATP-independent HSPs that are chaperones with a molecular mass between 8 and 28 kDa. These chaperones include ubiquitin, α-crystallins, Hspb1 (also known as Hsp25 in mice or Hsp27 in rats and humans), and many others [16]. These small HSPs have received growing research attention in the past and present moment, primarily due to their possible unprotective approaches [16]. Second, the large, ATP-dependent HSPs are chaperones with a molecular mass between 40 and 110 kDa. These include the well-known chaperones of the 40, 70, and 90 kDa families. The 40 kDa group (Hsp40s or J-proteins) plays an important role in facilitating the interaction between the substrate and 70 kDa molecular chaperones (Figure 1). They interact with Hsp70 through their J domain and serve as regulatory co-chaperones. The 70 kDa group consists of the stress-inducible Hsp70

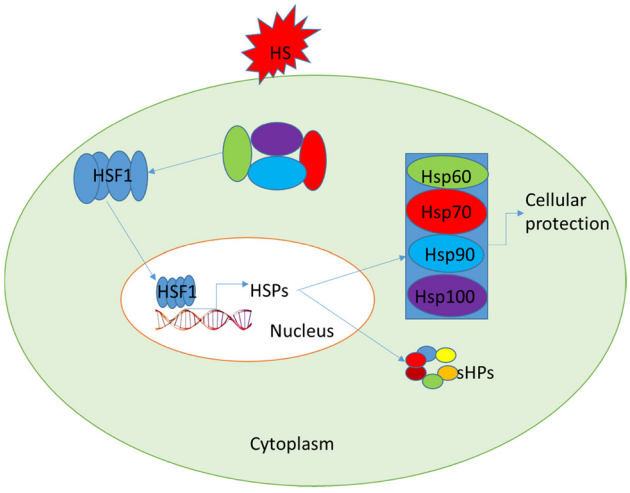

Figure 1: Expression of HSPs in mammalian cells. The regulation of HSPs by heat shock factor 1 (HSF1) both under normal and heat shock results in the expression of small HSPs, Hsp60, Hsp70, Hsp90, and Hsp100 as protective tools inside the cellular system.

and the constitutively expressed heat shock cognate 70 (Hsc70) [17,18].

Similarly, the 90 kDa group consists of two major isoforms, namely, the inducible Hsp90α and the constitutively expressed Hsp90β. However, because these isoforms are often challenging to isolate, many studies have resorted to studying co-purified aggregates containing both Hsp90α and Hsp90β, simply referring to the whole as Hsp90 [18]. In a host cell, a diverse network of chaperones is responsible for proteostasis, whereas viruses only depend on a subset of the host chaperones. Here, we review the role of some of these HSPs in SARS-Cov-2 since the advent of COVID-19.

40 kDa, heat shock protein 40 (Hsp40)

Hsp40, also known as DnaJ in *E. coli*, plays an essential role in coordinating the newly synthesized proteins to form a three-dimensional structure without forming part of the final product. The primary function of Hsp40 is to bring the substrate to other molecular chaperones such as Hsp70 for a proper folding process. There is evidence that Hsp40s take part in virus entry, virus replication, virus gene expression, protein maturation, immunity modulation, and cellular transformation in RNA and DNA viruses [19]. However, there is not enough information on the role of Hsp40 in the current SARS-CoV-2, which requires a detailed investigation to speed up the drive to come up with effective drugs or alternative treatments for the current pandemic. It is also important to mention that in our group we are busy working on getting clear answers on the role of this important molecule in SARS-CoV-2 [20].

60 kDa, Hsp60

Hsp60 are proteins composed of two rings that lie back to back, forming a cylinder-like structure with a central cavity in which protein folding occurs. Generally, Hsp60 is vital for expediting protein folding, transportation, and proteostasis in the mitochondria. Together with Hsp90, Hsp60 has been implicated in the development and progression of viruses, taking part in folding viral proteins and genome replication. According to Wan et al., the Hsp60 family of proteins is involved in virus replication, viral protein translocation, immunity modulation, apoptosis regulation, and genome integration in different viral infections. Recently, Hsp60 has been implicated in inappropriate inflammatory reactions that exacerbate the progression of COVID-19 [20,21].

70 kDa, Hsp70

Hsp70 is the most studied molecular chaperone and is found in almost all living organisms, from plants to humans. They are made of N-terminal domain (NTD), substrate-binding domain, and C-terminal domain with a special glutamic acid, glutamic acid, valine, aspartic acid motif that has been shown to interact with Hsp90 through the heat organizing protein in the presence of Hsp40 as a co-chaperone. Some members of the Hsp70 family, including Hsc70 and GRP78, have been implicated in a wide range of viral processes: virus entry, replication, virus gene expression, assembly, virus release, cellular transformation, immunity modulation, cell survival, and apoptosis [22,23]. Two of the Hsp70s in host cells have been implicated in the modulation of SARS-CoV-2. Hsp70A1L was reported to be epigenetically modulated by SARS-Cov-2, among other genes, including those from the Hsp70A1L family of proteins [24,25]. This massive overexpression of Hsp70 protein in COVID-19 patients signifies the importance of Hsp70s in all viral processes, that is, viral entry, replication, and viral exit from host cells, as has been reported with rabies and Zika viruses [26,27]. Similarly, this suggests that Hsp70s possibly promote SARS-CoV-2's replication and infectivity.

The second, Hsp70, has been implicated in host cell recognition of SARS-CoV-2 and viral entry. Since the dawn of COVID-19, initial studies have shown that GRP78 is overexpressed in the endoplasmic reticulum (ER) destined for the cell membrane. Both predictive and structural studies have shown that GRP78 assists in the host cell recognition of SARS-CoV-2 spikes. The binding of Grp78 to a C480-C488 (CNGVEGFNC) region on the C-terminus of S1 (Spike) is predicted to occur via hydrogen and hydrophobic interactions [28–30]. The CNGVEGFNC is mainly composed of polar (C and N) and hydrophobic (G, V, and F) residues. The binding is consistent with the Grp78's propensity for hydrophobic patches in proteins [31,32]. On the other hand, GRP78 being an ER chaperone has been reported to take part in protein folding in the ER lumen upon host-inflicted stress. This is so different in Covs as well as in cancer, and this function could be conserved in SARS-CoV-2 as it was observed in SARS-CoV [33]. Inhibiting GRP78 function could limit viral entry as well as the chaperoning of viral proteins. However, inhibition of Hsp70 usually elicits an HSP cascade, especially

the overexpression of Hsp90, to counteract the adverse effects of inhibition. Hence, it is also essential to review the role of Hsp90 in COVID-19.

90 kDa, Hsp90

Hsp90 is a molecular chaperone essential for the constancy and function of various conditionally activated and/or expressed signaling proteins and multiple mutated chimeric, or overexpressed signaling proteins, which promote cancer cell growth or survival or both. In viral infections, Hsp90 is essential for virus entry, replication, protein maturation, virus assembly, virus gene expression, immunomodulation, and cellular transformation [20]. Upon entry into the host, a myriad of viral proteins are rapidly synthesized. Hsp90 is responsible for the folding, stabilization, and maturation of a variety of viral structural proteins [23,34–37]. Li et al. [35] reported the stabilizing effect of Hsp90β on a MERS-CoV nucleoprotein (NP), while the maturation of the viral capsid proteins was reported in refs [36,37]. Recent studies on human coronaviruses, MERS-CoV, SARS-CoV, and SARS-CoV-2 in the study by Li et al. [35], demonstrated that Hsp90 is essential for viral replication. Their work showed the high dependence of viral proteins on Hsp90. When Hsp90β was genetically deleted in MERS-CoV, replication was observed to be suppressed and this was coupled with the cessation of viral spread. In the ER, a buildup of unfolded proteins is reported to elicit the transcription of yet another Hsp90, glucose-regulated protein 94 (GRP94), as part of the unfolded protein response. Grp94 together with other ER chaperones becomes protective and controls viral protein homeostasis [38]. In this way, the ER recovers from viral protein-induced ER stress to the benefit of the virus. This highlights the importance of Hsp90 in some of the various viral processes, and hence, it has been proposed as a drug target. Other researchers also demonstrated that a well-known Hsp90 inhibitor 17-AAG suppressed MERS-Cov replication. Interestingly, studies have suggested that another known Hsp90 inhibitor, Geldanamycin, and its derivatives can be useful in combating the progression of SARS-CoV-2 [39].

The small HSPs

Small HSPs are a highly coordinated group of proteins found across all species, including humans, and are vital for stress tolerance. Many sHsps show chaperone-like activity in inhibiting the aggregation of target proteins, keeping them in a folding-competent form, and refolding them by themselves or in concert with other ATP-dependent chaperones. In short, these chaperone proteins are ATP-independent as opposed to major chaperones such as Hsp70 for example. sHsps have subunit molecular masses of 12–43 kDa and are characterized by the presence of a highly conserved stretch of 80–100 amino acids in their C-terminal domain called the "α-crystallin domain" (ACD) that is flanked by the less conserved (except a few stretches) NTD and C-terminal extension. Some members of the sHsp family such as Hsp27, αA-, and αB-crystallin form large oligomeric species. sHsps are reported to take part in viral replication. In coronaviruses and other viruses, Hsp27 is reported to be rapidly upregulated, suggesting a role in early replication. Wan et al. (2020) discuss the role of Hsp27 in different viruses. However, not much is known about Hsp27 in SARS-CoV-2 save for speculation about its function in other coronaviruses. Hence, it has been proposed as a potential drug target in SARS-CoV-2 [40]

The synthesis of HSPs is regulated by the HSF

The transcriptional range of Hsps is regulated by HSF1 in invertebrates and HSFs 1–4 in vertebrates, with HSF1 being the most important. In response to proteotoxic stress conditions, heat shock factor 1 (HSF1) is subject to a multistep activation and attenuation cycle. Inactive HSF1 monomers are retained in the cytoplasm in a complex with regulatory proteins such as Hsps 40, 70, and 90, as well as the cytosolic chaperonin TCP1 ring complex (TRiC) [41,42]. Upon stress sensing, HSF1 is activated, causing the dissociation of inhibitory proteins, HSF1 oligomerization, and nuclear retention. HSF1 is modified by several activated posttranslational modifications (PTMs) that promote DNA binding and transcriptional activation of target genes in concert with cofactor recruitment [43,44]. HSF1 is then modified by different inhibitory PTMs and by p23, causing DNA dissociation (Figure 2). Taken together, viruses such as SARS-CoV-2 require HSP to maintain and keep their cellular proteins in good condition. Therefore, HSPs are important for viral infection. It is important to develop a strategy to block the virus from accessing these molecules (Table 1).

How the production of HSPs is triggered by some viruses

The exposure of cells to foreign substances triggers the expression of protective molecules to keep the cellular

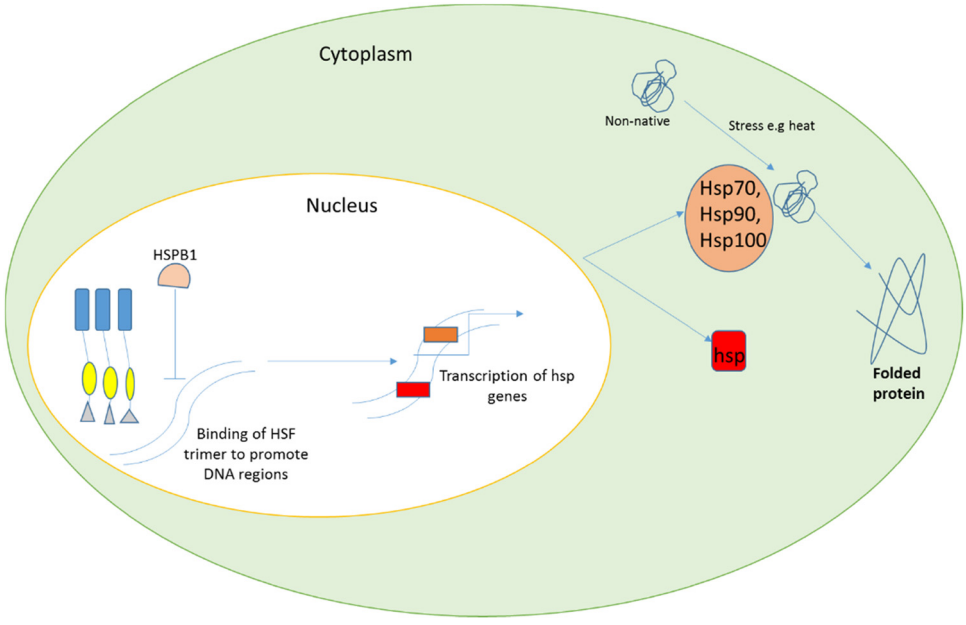

Figure 2: Transcriptional regulation of HSPs by HSF. The expression of Hsps 70, 90, and 100 helps in the stabilization of newly synthesized proteins. Hspb1 binds to HSF to promote the synthesis of HSPs.

system in good shape and viable. Among the molecules that act as cellular housekeepers are HSPs, whose role is to prevent proteins from forming aggregates that could otherwise become toxic to the entire cellular system and lead to cell death. Some of the substances that could trigger the excessive production of HSPs are bacteria and viruses. Figure 3 shows some of the viruses that could lead to the high expression of HSPs as a protective measure for the human host cellular system. The heat shock response is regulated at the transcriptional level by activities of the family of heat shock factor 1 (HSFs). The most studied HSF genes from humans are *HSF1*, *HSF2*, and *HSF4*, and these genes play important role in heat shock response [41,45]. HSF1 localizes at cytoplasmic level under normal conditions in mammalian cells in a negatively charge form and is associated with molecular chaperones such as HSP70 and 90 as well as co-chaperones [41]. Upon the appearance of unfolded proteins and release of interacting chaperones, HSF1 DNA-binding activity is de-repressed, and monomers oligomerize to a trimeric state, translocate to the nucleus, are inducibly phosphorylated and bind to specific sequence elements referred to as heat shock elements, located within the HSP gene promoters, activating transcription of heat shock genes and synthesis of HSP. Therefore, when foreign substances such as viruses enter the human system, they

Table 1: Summary of mammalian HSPs and their roles

Family name		Localization	Functional activities
Small HSPs	Aβ-crystalline	Cytoplasm	Cytoskeleton stabilization
	Hsp27	Cytoplasm/nucleus	Actin dynamics
	Heme oxygenase, Hsp32	Cytoplasm	Heme catabolism, antioxidant properties
Hsp60, chaperonins	Hsp60	Mitochondria	Assist newly synthesized proteins during folding
	TCP-1	Cytoplasm	ATP binding activity
Hsp70	Hsp70	Cytoplasm/nucleus	Folding, maintenance of newly produced proteins
	Hs70	Cytoplasm	Folding activity
	mtHsp70/Grp75	Mitochondria	HSF1 regulations
Hsp90	Hsp90 (α and β)	Cytoplasm	Prevents protein aggregation
	Grp94/gp96/Hsp100	ER	Maintenance of HSF1 activity
Hsp110	Hsp110	Cytoplasm/nucleus	Thermal tolerance
	Apg-1	Cytoplasm	Protein refolding
	Hsp105	Cytoplasm	

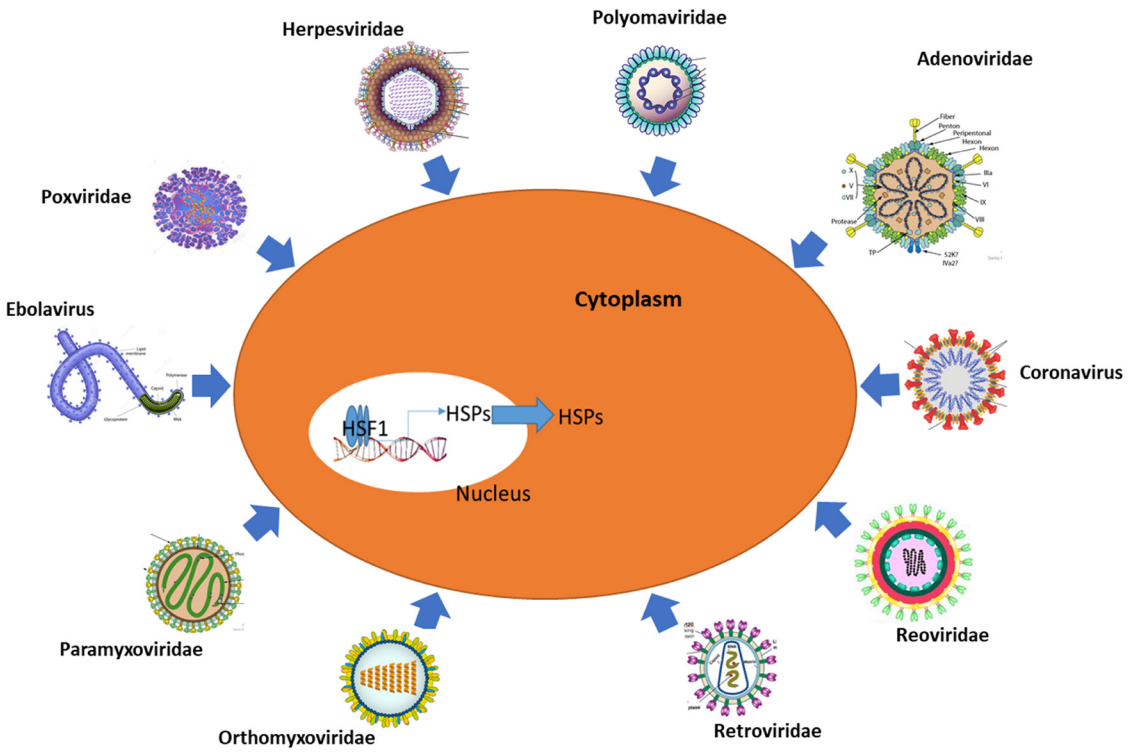

Figure 3: Some of the viruses that activate the expression of HSPs in human cells. HSF1 activates the expression of HSPs in response to foreign substances such as viruses, adapted from ref. [46].

hijack human host HSPs to help in the folding of the viral proteins during their synthesis inside the mammalian system, thus changing the normal functional properties of the host [46]. For example, when SARS-CoV-2 enter the human host, they cannot produce proteins on their own and therefore exploit the human host to produce proteins such as spike proteins. Viruses use the human host's ribosomes to produce their proteins, during the synthesis of proteins such as spike proteins, upon their exit from ribosomes like any other proteins they require the assistance of molecular chaperones also known as HSPs to fold properly. Hence, the human host's molecular chaperones are then hijacked by the SARS-CoV-2 for their benefit of the viral proteins to be folded properly. It is crucial to design a drug that would stop the virus from entering the human host system and use the chaperones from the human host. In our laboratory, work is currently underway to develop innovative strategies to block the SARS-CoV-2 from its dependency on the human host.

Polyamines from mammalian cells

Almost all living organisms have polyamines such as putrescine, spermidine, and spermine; however, their concentrations differ depending on the type of organism. At physiological pH, polyamines are positively charged and these biogenic molecules have a flexible carbon chain with amino acids. Organisms like bacteria and archaea possess various groups of polyamines, including spermidine, homospermidine, norspermidine, putrescine, cadaverine, and 1,3-diaminopropane in bacteria and agmatine, spermidine, homospermidine, norspermidine, and norspermine in archaea. The variety of polyamines present in each of these kingdoms varies among the different organisms [47,48]. The main polyamine synthesis pathway present in mammals is summarized in Figure 4. Polyamines' vital role is defined as being molecules involved in cell growth, differentiation, and proliferation. In a normal cellular system or healthy cell, polyamines are involved in various cellular processes such as protein synthesis, RNA folding and bending, membrane interactions, protein–RNA interactions, DNA structure, and gene expression. Polyamines bind both RNA and DNA, altering the conformation and function of nucleic acids. Polyamines alter DNA structure by facilitating the conformational transition from the B form to the Z form or by bending DNA. Furthermore, up to 80% of polyamines in the cell are directly associated with RNA and spermine has also been implicated in the stabilization of tRNA structure [47,49]. Additionally, these

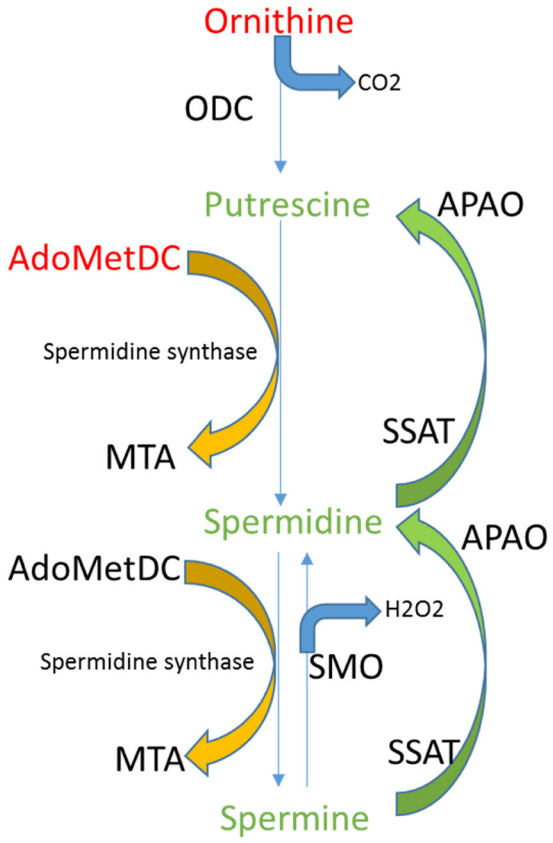

Figure 4: Biosynthesis of polyamines in humans. Both ornithine and S-adenosylmethionine are precursors of polyamine synthesis such as putrescine, spermidine, and spermine. Abbreviations: AdoMetDC, S-adenosylmethionine decarboxylase; ODC, ornithine decarboxylase; MTA, methylthioadenosine; SSAT, *Spermidine/spermine-N*(1)-acetyltransferase; SMO, spermine oxidase; and APAO, acetylpolyamine oxidase.

molecules are reported to exhibit molecular chaperone activity, therefore protecting cells exposed to harsh conditions such as temperature increases. Polyamines include putrescine, spermidine, and spermine, thus are synthesized through ornithine and adomet as precursors with the help of various enzymes. For example, ornithine is converted to putrescine with the help of ODC, while adomet is converted into spermidine and spermine with the help of adomet decarboxylase enzyme [50].

The role of polyamines in humans and viruses

The polycationic nature of polyamines allows them to be involved in various activities inside the cellular system. Besides their role in cell proliferation, growth, differentiation, and chaperone activities, polyamines in viruses are involved in packaging, transcription, translation, protease activity, and cell binding. However, in humans, polyamines play a pivotal role in nucleic acid metabolism, transcription, translation, chromatin structure, and membrane fluidity (Figure 5). In general, viruses cannot synthesize their proteins, but they depend on the host system. Therefore, viruses hijack the role of polyamines from the human host and manipulate them to synthesize and replicate their proteins such as spike proteins [48].

Viruses compete for molecule hosts

To initiate infection, a virus enters a host cell typically via receptor-dependent endocytosis. It then penetrates the subcellular membrane, reaching a destination that supports transcription, translation, and viral genome replication [51]. These steps lead to the assembly and morphogenesis of the new viral progeny. The mature virus

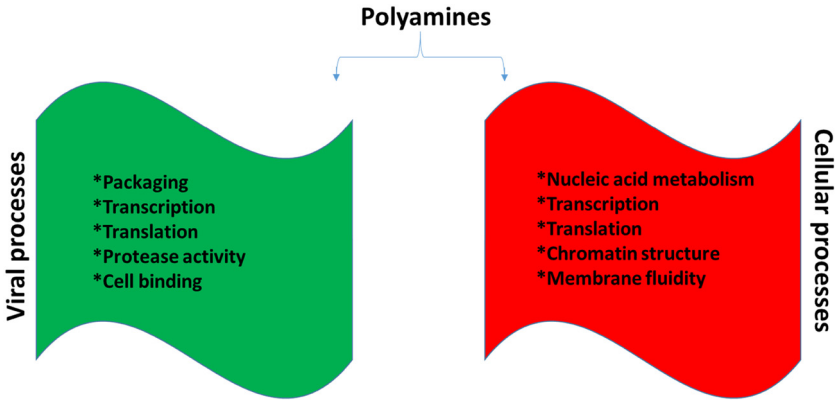

Figure 5: Summary of polyamines in cellular and viral processes.

finally exits the host cell to begin the next infection cycle. Strikingly, viruses hijack host molecular chaperones to accomplish these distinct entry steps. Viruses are acellular, parasitic entities that are not classified within any domain because they are not considered alive [52]. They have no plasma membrane, internal organelles, or metabolic processes, and they do not divide. Instead, they infect a host cell and use the host's replication processes to produce progeny virus particles. Viruses infect all forms of organisms including bacteria, archaea, fungi, plants, and animals. Other organisms grow, metabolize, and reproduce whereas viruses do not, but are assembled in their mature form. Viruses replicate, but to do so, they are entirely dependent on their host cells. More so, viruses are diverse. They vary in their structure, their replication methods, and their target hosts or even host cells.

Conclusion and future perspectives

To develop effective and sustainable drugs or treatments for COVID-19, it is essential to look at how viruses, in general, hijack molecules found in the human host. The point of reference could be the role of polyamines and HSPs in virus reproduction and viral infection [48]. A full understanding of how a virus-like SARS-CoV-2 utilizes the aforementioned molecules could lead to the innovative development of vaccines that could prevent the current virus from hijacking and altering the host system. As no host has no virus, preventing viruses to access the human host is essential in eliminating viruses. In our laboratory, studies are underway to elucidate the role of these molecules in SARS-CoV-2 and possibly come up with alternative therapeutics for SARS-CoV-2. It is important to mention that dual treatment has been proposed by various studies including our group, of which our current research is also looking at the current virus, COVID-19.

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References

[1] Ritossa F. A new puffing pattern induced by temperature shock and DNP in drosophila. Cell Mol Life Sci. 1962;18(12):571–3.
[2] Hughes JP, Rees S, Kalindjian SB, Philpott KL. Principles of early drug discovery. Br J Pharmacol. 2011;162(6):1239–49. doi: 10.1111/j.1476-5381.2010.01127.x.
[3] Ko SH, Huang LM, Tarn WY. The host heat shock protein MRJ/DNAJB6 modulates virus infection. Front Microbiol. 2019;10:2885. doi: 10.3389/fmicb.2019.02885.
[4] World Health Organization (WHO), 2019.
[5] World Health Organization (WHO), 2020.
[6] Chattopadhyay MK, Tabor CW, Tabor H. Polyamines protect *Escherichia coli* cells from the toxic effect of oxygen. PNAS. 2003;100(5):2261–5.
[7] Perdeh J, Berioso B, Love Q, LoGiudice N, Le TL, Harrelson JP, et al. Critical functions of the polyamine putrescine for proliferation and viability of *Leishmania donovani* parasites. Amino Acids. 2020;52:261–74.
[8] Vannier-Santosa MA, Suarez-Fontes AM. Role of polyamines in parasite cell architecture and function. Curr Pharm Des. 2017;23:3.
[9] Oh TJ, Kim IG. Polyamines protect against DNA strand breaks and aid cell survival against irradiation in *Escherichia coli*. Biotechnol Tech. 1998;12(10):755–8.
[10] Ivanov IP, Shin B, Loughran G, Cao C, Tzani I, Young-Baird SK, et al. Polyamine control of translation elongation regulates start site selection on antizyme inhibitor mRNA via ribosome queuing. Mol Cell. 2018;70:254–64.
[11] Kusano T, Berberich T, Tateda C, Takahashi Y. Polyamines: essential factors for growth and survival. Planta. 2008;228:367–81.
[12] Niemand J, Birkholtz L, Louw AI, Kirk K. Polyamine uptake in the malaria parasite, *Plasmodium falciparum*, is dependent on the parasite's membrane potential. Malar J. 2010;9(Suppl 2):O24.
[13] Miller-Fleming L, Olin-Sandoval V, Campbell K, Ralser M. Remaining mysteries of molecular biology: the role of polyamines in the cell. J Mol Biol. 2015;427:3389–406.
[14] Rhee HJ, Kim E-J, Lee JK. Physiological polyamines: simple primordial stress molecules. J Cell Mol Med. 2007;11:685–703.
[15] Carrillo C, Cejas S, González NS, Algranati ID. *Trypanosoma cruzi* epimastigotes lack ornithine decarboxylase but can express a foreign gene encoding this enzyme. FEBS Lett. 1999;454:192–6.
[16] Nagai Y, Fujikake NH, Popiel A, Wada K. Induction of Molecular chaperones as a therapeutic strategy for the polyglutamine diseases. Curr Pharm Biotechnol. 2010;2010(11):188–97.

[17] Kalia SK, Kalia LV, McLean PJ. Molecular chaperones as rational drug targets for Parkinson's disease therapeutics. CNS Neurol Disord Drug Targets. 2010;9(6):741–53.
[18] Lebepe CM, Matambanadzo PR, Makhoba XH, Achilonu I, Zininga T, Shonhai A. Comparative characterization of *Plasmodium falciparum* Hsp70-1 relative to *E. coli* DnaK reveals functional specificity of the parasite chaperone. Biomolecules. 2020;10:856. doi: 10.3390/biom10060856.
[19] Kolaj O, Spada S, Robin S, Wall JG. Use of folding modulators to improve heterologous protein production in *Escherichia coli*. Microb Cell Fact. 2009;8:9. doi: 10.1186/1475-2859-8-9.
[20] Wan Q, Song D, Li H, He ML. Stress proteins: the biological functions in virus infection, present and challenges for target-based antiviral drug development. Signal Transduct Target Ther. 2020;5:125.
[21] Jakovac H. COVID-19 and hypertension: is the HSP60 culprit for the severe course and worse outcome? Am J Physiol Heart Circ Physiol. 2020;319(4):H793–6. doi: 10.1152/ajpheart.00506.2020.
[22] Iyer K, Chand K, Mitra A, Trivedi J, Mitra D. Diversity in heat shock protein families: functional implications in virus infection with a comprehensive insight of their role in the HIV-1 life cycle. Cell Stress Chaperones. 2021;26:743–68.
[23] Lubkowska A, Pluta W, Stronska A, Lalko A. Role of heat shock proteins (HSP70 and HSP90) in viral infection. Int J Mol Sci. 2021;22:9366. doi: 10.3390/ijms22179366.
[24] Muhammad JS, Saheb Sharif-Askari N, Cui ZG, Hamad M, Halwani R. SARS-CoV-2 infection-induced promoter hypomethylation as an epigenetic modulator of heat shock protein A1L (HSPA1L) gene. Front Genet. 2021;12:622271. doi: 10.3389/fgene.2021.622271.
[25] Muhammad JS, Siddiqui R, Khan NA. COVID-19: does SARS-CoV-2 modulate acanthamoeba epigenetics to enhance survival and transmission in the environment? ACS Pharmacol Transl Sci. 2021;4(2):1021–3.
[26] Lahaye X, Vidy A, Fouquet B, Blondel D. Hsp70 protein positively regulates rabies virus infection. J Virol. 2012;86(9):4743–51. doi: 10.1128/jvi.06501-11.
[27] Taguwa S, Yeh M-T, Rainbolt TK, Nayak A, Shao H, Gestwicki JE, et al. Zika virus dependence on host Hsp70 provides a protective strategy against infection and disease. Cell Rep. 2019;26(4):906e903. doi: 10.1016/j.celrep.2018.12.095.
[28] Elfiky AA. Natural products may interfere with SARS-CoV-2 attachment to the host cell. J Biomol Struct. 2020;39(9):3194–203. doi: 10.1080/07391102.2020.1761881.
[29] Elfiky AA. SARS-CoV-2 spike-heat shock protein A5 (GRP78) recognition may be related to the immersed human coronaviruses. Front Pharmacol. 2020b;11. doi: 10.3389/fphar.2020.577467.
[30] Ibrahim IM, Abdelmalek DH, Elshahat ME, Elfiky AA. COVID-19 spike-host cell receptor GRP78 binding site prediction. J Infect. 2020;80(5):554–62. doi: 10.1016/j.jinf.2020.02.026.
[31] Tsai Y-L, Lee AS. Cell surface GRP78: anchoring and translocation mechanisms and therapeutic potential in cancer. In: Salvatore V. Pizzo, editors. Cell surface GRP78, a new paradigm in signal transduction biology. Academic Press; 2018. p. 41–62. doi: 10.1016/b978-0-12-812351-5.00003-9.
[32] Li J, Lee A. Stress induction of GRP78/BiP and its role in cancer. Curr Mol Med. 2006;6(1):45–54. doi: 10.2174/156652406775574523.
[33] Chan CP, Siu KL, Chin KT, Yuen KY, Zheng B, Jin DY. Modulation of the unfolded protein response by the severe acute respiratory syndrome coronavirus spike protein. J Virol. 2006;80(18):9279–87. doi: 10.1128/JVI.00659-06.
[34] Geller R, Vignuzzi M, Andino R, Frydman J. Evolutionary constraints on chaperone-mediated folding provide an antiviral approach refractory to development of drug resistance. Genes Dev. 2007;21(2):195–205. doi: 10.1101/gad.1505307.
[35] Li C, Chu H, Liu X, Chiu MC, Zhao X, Wang D, et al. Human coronavirus dependency on host heat shock protein 90 reveals an antiviral target. Emerg Microbes Infect. 2020;9(1):2663–72.
[36] Hung JJ, Chung CS, Chang W. Molecular chaperone Hsp90 is important for vaccinia virus growth in cells. J Virol. 2002;76(3):1379–90. doi: 10.1128/jvi.76.3.1379-1390.2002.
[37] Newman J, Asfor AS, Berryman S, Jackson T, Curry S, Tuthill TJ. The cellular chaperone heat shock protein 90 is required for foot-and-mouth disease virus capsid precursor processing and assembly of capsid pentamers. J Virol. 2018;92(5):1–14. doi: 10.1128/JVI.01415-17.
[38] Rainbolt TK, Saunders JM, Wiseman RL. Stress-responsive regulation of mitochondria through the ER unfolded protein response. Trends Endocrinol Metab. 2014;25(10):528–37. doi: 10.1016/j.tem.2014.06.007.
[39] Sultan I, Howard S, Tbakhi A. Drug repositioning suggests a role for the heat shock protein 90 inhibitor geldanamycin in treating COVID-19 infection. Res Sq. 2020. doi: 10.21203/rs.3.rs-18714/v1.
[40] Cao Z, Han Z, Shao Y, Liu X, Sun J, Yu D, et al. Proteomics analysis of differentially expressed proteins in chicken trachea and kidney after infection with the highly virulent and attenuated coronavirus infectious bronchitis virus in vivo. Proteome Sci. 2012;10(1):24. doi: 10.1186/1477-5956-10-24.
[41] Gomez-Pastor R, Burchfiel ET, Thiele DJ. Regulation of heat shock transcription factors and their roles in physiology and disease. Nat Rev Mol Cell Biol. 2018;19(1):4–19. doi: 10.1038/nrm.2017.73.
[42] Dayalan Naidu S, Dinkova-Kostova AT. Regulation of the mammalian heat shock factor 1. FEBS J. 2017;284(11):1606–27. doi: 10.1111/febs.13999.
[43] Xu YM, Huang DY, Chiu JF, Lau AT. Post-translational modification of human heat shock factors and their functions: a recent update by proteomic approach. J Proteome Res. 2012;11(5):2625–34. doi: 10.1021/pr201151a.
[44] Kmiecik SW, Mayer MP. Molecular mechanisms of heat shock factor 1 regulation. Trends Biochem Sci. 2022;47(3):218–34. doi: 10.1016/j.tibs.2021.10.004.
[45] Puustinen MC, Sistonen L. Molecular mechanisms of heat shock factors in cancer. Cells. 2020;9(5):1–20. doi: 10.3390/cells9051202.
[46] Santoro MG, Amici C, Rossi A. Role of Heat Shock Proteins in Viral Infection. In: Pockley A, Calderwood S, Santoro M editors. Prokaryotic and eukaryotic heat shock proteins in infectious disease. Heat Shock Proteins. vol 4. Dordrecht: Springer; 2009. doi: 10.1007/978-90-481-2976-8_3.
[47] Michael AJ. Polyamines in eukaryotes, bacteria, and archaea. J Biol Chem. 2016;291(29):14896–903. doi: 10.1074/jbc.R116.734780.
[48] Mounce BC, Olsen ME, Vignuzzi M, Connor JH. Polyamines and their role in virus infection. Microbiol Mol Biol Rev. 2017;81:e00029-17. doi: 10.1128/MMBR.00029-17.

[49] Lightfoot HL, Hall J. Endogenous polyamine function – the RNA perspective. Nucleic Acids Res. 2014;42(18):11275–90. doi: 10.1093/nar/gku837.
[50] Casero RA, Murray-Stewart T, Pegg AE. Polyamine metabolism and cancer: treatments, challenges, and opportunities. Nat Rev Cancer. 2018;18:681–95.
[51] Speckhart K, Williams JM, Tsai B. How DNA and RNA viruses exploit host chaperones to promote infection. Viruses. 2021;13(6):1–15. doi: 10.3390/v13060958.
[52] Molnar C, Gair J. Introduction to the immune system and disease. Chapter 12 in Concepts of biology – 1st Canadian edition, BC campus; 2015. ISBN: 978-1-989623-98-5.