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LETTER TO THE EDITOR

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Covid-19 and thymoquinone: Connecting the dots

At present, the medical approaches to cope with Covid-19 infection caused by the respiratory syndrome coronavirus 2 (SARS-CoV-2) are mainly supportive. In the absence of specific anti-viral therapies or vaccines, the medical care is complemented with different combinations of broad-spectrum antiviral agents, antibiotics, hydroxychloroquine, and convalescent plasma transfusion (Jin et al., 2020).

Thymoquinone, the main constituent of *Nigella sativa*, has demonstrated anti-inflammatory, anti-oxidant, anti-tumor, and antimicrobial activities (Banerjee et al., 2009; Chaieb, Kouidhi, Jrah, Mahdouani, & Bakhrouf, 2011). Thymoquinone was also effective and tolerable in children with intractable epilepsy in a randomized controlled clinical trial at a dose of 1 mg/kg/day orally (Akhondian et al., 2011). Interestingly, thymoquinone and *Nigella sativa* extract were found to be effective against avian influenza virus (H9N2 AIV) and a murine cytomegalovirus infection model (Salem & Hossain, 2000; Umar et al., 2016). Ulasli and co-workers reported that the treatment of cells with *Nigella sativa* extract prior to infection with coronavirus decreases the replication of the virus (Ulasli et al., 2014). Moreover, gene expression analysis of the transient receptor potential proteins (TRPs) showed a reduction in virus loads upon extract treatments, which can decrease coronavirus survival inside cells. It should be noted, however, that these studies on the herbal extracts may not have been carried out according to the more recent scientific qualitative standards for plant-derived products (Heinrich et al., 2020). Therefore, there is the possibility that high concentrations in vitro or doses in vivo, which are of no translational value have been used.

Thymoquinone as a compound (purity >99%) has unveiled a remarkable anti-sepsis and immunomodulatory activities at specific doses (Alkharfy, Ahmad, Jan, & Raish, 2018; Alkharfy, Ahmad, Raish, & Vanhoutte, 2015; Alkharfy, Al-Daghri, Al-Attas, & Alokail, 2011). More specifically, thymoquinone modulates the production of nitric oxide (NO) and reactive oxygen species (ROS), and protects against multiple organ dysfunction syndrome (MODS). ROS including superoxide, hydrogen peroxide, and hydroxyl radicals are produced, among others, by xanthine oxidase and NADH/NADPH oxidases (Galley, 2011; Ichinose et al., 2007). The NADPH oxidases, uncoupled NO synthase (iNOS), and mitochondria are considered important mediators of ROS in sepsis and cardiovascular dysfunction (Kirkeboen & Strand, 1999; Munzel, Gori, Bruno, & Taddei, 2010; Tsolaki, Makris, Mantzarlis, & Zakynthinos, 2017). In fact, sepsis is characterized by the enhanced release of NO, which correlates with systemic dysfunction and tissue injury in humans and animal models (Rabuel et al., 2010; Tsolaki et al., 2017). NO can interact with the absorption of calcium in the myocytes and, therefore, can impede contractile activity (Forstermann & Sessa, 2012). In addition, NO plays a key role in the systemic inflammation of sepsis including vasodilatation, altered vascular permeability and extravasation, leukocyte migration, and activation (Ince et al., 2016). Notably, inflammatory cytokines such as TNF-α, IL-1α, IL-2, IL-6, and IL-10 also enhance NO production via iNOS (Green et al., 1994). Thymoquinone has been shown to downregulate inflammatory cytokines, reduce NO levels, and improve organ functions and survival of sepsis in an animal model (Alkharfy et al., 2015). This perhaps through a redox mechanism, which decreases the systemic oxidative stress and inflammatory response. Consequently, thymoquinone decreases the levels of early-stage sepsis biomarkers (e.g., ESM-1, CRP, and VEGF) by ∼30–50% (Alkharfy et al., 2018). Interestingly, thymoquinone has also been found to have a protective effect against lung fibrosis and collagen deposition by modulating the nuclear factor Kappa-B (NF-κB) and the antioxidant enzyme nuclear factor 2 heme oxygenase-1 (Nrf2/HO-1) signaling pathway (Ahmad et al., 2020).

Virus-induced phagocyte activation is correlated with oxidative stress, not just because ROS is produced, but also because activated phagocytes also produce inflammatory cytokines by the activation of NF-κB (S. F. Liu & Malik, 2006; Schwarz, 1996). Actually, many genes that are regulated by NF-κB, including inflammatory cytokines, COX-2, and iNOS, contribute to a rise in sepsis inflammatory responses (Ghosh, May, & Kopp, 1998; Schneider-Stock, Fakhoury, Zaki, El-Baba, & Gali-Muhtasib, 2014). Thus, NF-κB inhibition can suppress inflammatory genes, impede the cytokine storm, and reduce immune cells infiltration and activation, and, therefore, protecting against tissue and organ damage (T. Liu, Zhang, Joo, & Sun, 2017). While inhibition of NF-κB activation has been suggested as a therapeutic strategy for sepsis, it should be noted that NF-κB is an important component of normal immune defenses and that excessive blockade of NF-κB regulatory activities can be strong immunosuppressive (Coldewey, Rogazzo, Collino, Patel, & Thiemermann, 2013). Therefore, a more selective modulation of NF-κB activity is probably needed. Overall, existing evidence indicates that thymoquinone can favorably modulate NF-κB expression during sepsis (Alkharfy et al., 2015). Consequently, thymoquinone can be a strong candidate to avert MODS and mortality of sepsis (Figure 1). Recently, molecular docking studies have also proposed that thymoquinone may inhibit SARS-CoV-2 and interfere with its binding to ACE2 receptors. This can prevent virus entry and replication inside the host cell (Bouchentouf & Missoum, 2020; Sekiou, Ismail, Zihad, & Abdelhak, 2020). Furthermore, SARS-CoV-2 spikes can bind to a cell surface heat shock protein (HSPA5), which is upregulated during viral infections. Molecular dynamics simulations showed that thymoquinone can interfere with the attachment of SARS-CoV-2 to

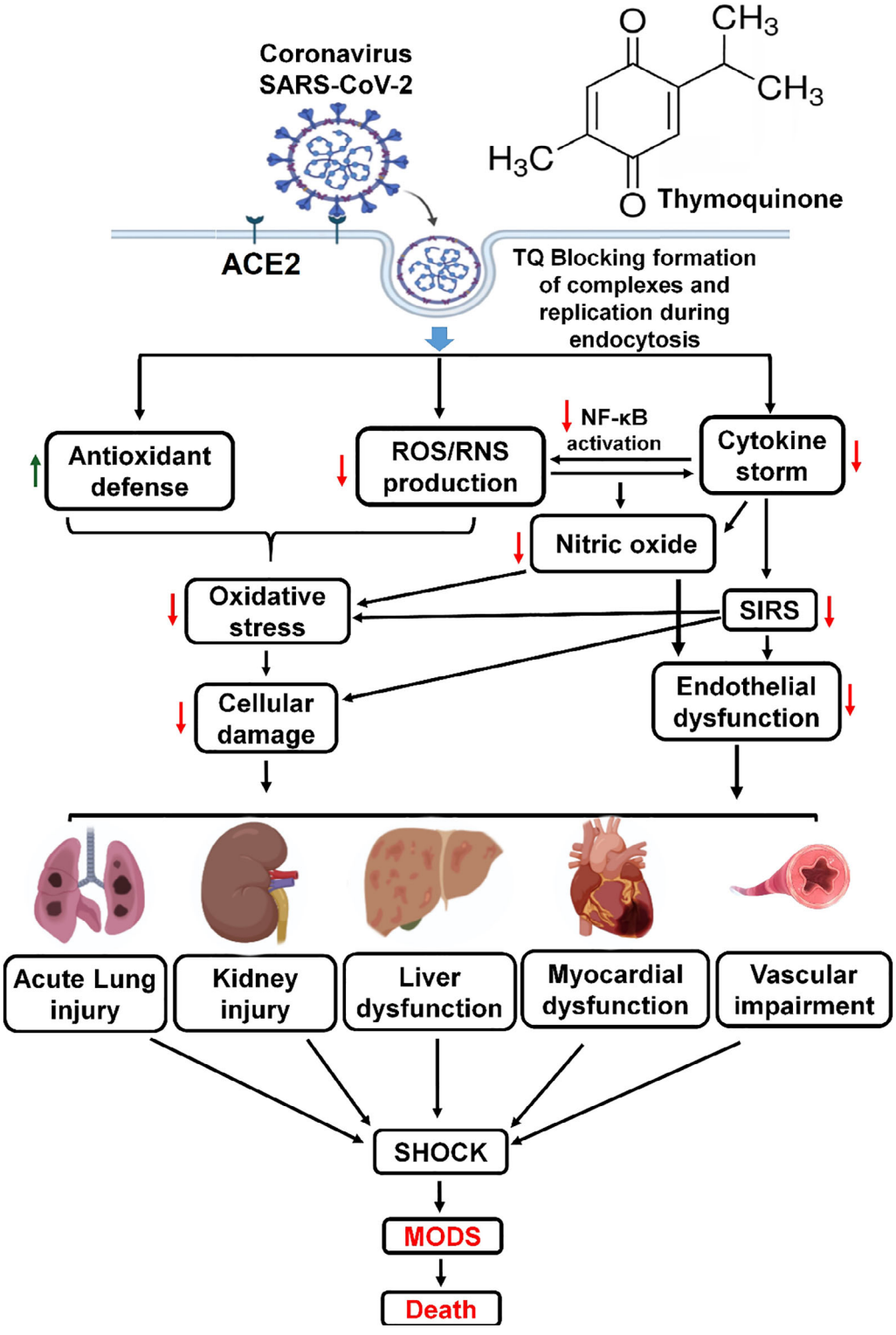

FIGURE 1 Thymoquinone potential protective mechanism against Covid-19 infection. Progression from risk factors for severe SARS-CoV-2 infection mediated by oxidative stress and cytokine storm-inducing multiple organ dysfunction syndrome (MODS). Thymoquinone inhibitory effects on viral infection and amelioration of MODS complications by restoration of the redox and immune balances. SARS-CoV-2 (Severe acute respiratory syndrome coronavirus 2); ACE (Angiotensin Converting Enzyme); ROS (reactive oxygen species); RNS (reactive nitrogen species); NF-κB (nuclear factor kappa-B); SIRS (systemic inflammatory response syndrome); green arrow indicates upregulation and red arrow indicates downregulation mediated by thymoquinone [Colour figure can be viewed at wileyonlinelibrary.com]

the HSPA5 substrate-binding domain b (SBDb) on the stressed cells, and thus may reduce the risk of infection (Elfiky, 2020). Therefore, the time is probably appropriate to move thymoquinone from experimentation on the bench to clinical testing for the Covid-19 pandemic.

CONFLICT OF INTEREST

None.

AUTHOR CONTRIBUTIONS

Ajaz Ahmad and Khalid M. Alkharfy: Conceptualization. Muneeb U. Rehman and Parvaiz Ahmad: Resources. Ajaz Ahmad and Muneeb U. Rehman: Writing. Parvaiz Ahmad and Khalid M. Alkharfy: Review and Editing. Ajaz Ahmad and Khalid M. Alkharfy: Supervision.

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