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REVIEW ARTICLE

**COVID-19 AND THE ENVIRONMENT – THE ROLE OF THE PUBLIC
HEALTH INSTITUTE**

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Abstract: The Croatian National Health Care Act defines the areas of activities of the public health institute, including the activities of the epidemiology of infectious diseases and chronic non-communicable diseases, public health, health promotion, environmental health, microbiology, school and adolescent medicine, mental health and addiction prevention at Zagreb City level. This paper reviews the highly variable activities in the Andrija Štampar Teaching Institute of Public Health with the aim of promoting a comprehensive approach to the COVID-19 pandemic. Human and analytical resources in the Institute, activities and rapid implementation of innovations testify to the high capacities for adaptation to emerging risks. In the Institute, it is possible to carry out a whole range of tests and to monitor the environmental factors with predominant impact on human health and safety of the Zagreb environment. The supply of safe water for human consumption in the Republic of Croatia during the current COVID-19 crisis has been uninterrupted and in accordance with applicable legislation. Also, our laboratories have been developing and introducing a method for wastewater testing for SARS-CoV-2 presence. The sludge from wastewater treatment plants is used in agriculture, and potential risks associated with the COVID-19 outbreak should be assessed prior to each application on the soil. Increased use of disinfectants during the epidemic may present a higher risk to the aquatic environment. Air quality monitoring indicates a positive impact on air quality as result of isolation measures.

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INTRODUCTION

Beginnings of the epidemic and the Institute's actions

Even before the first case of COVID-19 in Croatia, the epidemiology service monitored the situation daily. In order to delay the entry of the disease in our country, different preventive measures of control were taken over passengers from "risk" areas, and, at the same time, awareness and sensibilisation of the both the health system and the public was carried out through a series of guidelines.

The first case of infection by the virus called SARS-CoV-2 was reported in Zagreb on 25 February 2020. It initiated the creation of a detailed map of the patient and his contacts, including: the date of first symptoms, symptom strength, medical history, travels, lifestyle, private and business contacts, attendance of events with a higher number of people in a smaller area, healthcare system use, etc. The date of onset of the first symptoms of the disease in a positive patient is essential for determining contacts that are required to be in self-isolation and supervised by the healthcare system. Contact tracing is extremely important to alert the individuals with the highest risk of coronavirus exposure and prevent the negative consequences.¹

After the first few dozen imported cases, the onset of COVID-19-compatible symptoms started appearing in people who had not travelled or been, to their knowledge, in close contact with any affected person, which means that the so-called local transmission of the disease was identified. Local virus transmission raised another challenge for epidemiologists, as the infected

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person had to be identified as early as possible and self-isolation ordered for close contacts. A response followed by the Institute's clinical microbiologist who introduced SARS-CoV-2 diagnostics and accelerated the process of confirming individual infections thus speeding up the actions of epidemiologists.²

Preparations for the new SARS-CoV-2 coronavirus testing began in February 2020 with the procurement of a molecular test kit, and in early March the test was set up *in-house*. The test GeneFinder RT-PCR (CE-IVD) was imported from Korea, and it detects as many as three genes of the new coronavirus³ simultaneously. Following excellent results, the same test was introduced by clinical microbiology departments in other public health institutes. Also, in early April 2020, the Institute introduced a drive-in diagnostic of coronavirus in order to speed up diagnostics, increase the level of testing availability and provide a higher safety level of both the person tested and the health care professional, as people tested stay in their vehicle while swabbed.

DISCUSSION

Application of new technologies in epidemiological screening - 'Epidemicon'

The rapid rise of the number of infected people and the daily introduction of new guidelines have made the issue more complex. The Institute had to increase capacity to accept a large quantity of information from citizens and

to find a way to process these data, on the basis of which decisions are taken for further actions. This new situation also required the introduction of new IT tools to increase efficiency and for safer data management.

Through a donation by the Zagreb-based company INTIS, the Institute has digitized the demanding and highly dynamic process of epidemiological screening and set a good example for the wider use of the donated Epidemicon - the center for the coordination of epidemiologists, family physicians, testing laboratories and services for epidemic surveillance. In the further developmental phase of the process, citizens potentially infected with coronavirus were given the possibility to report data and symptoms via a web questionnaire <https://korona.prijavnicentar.hr>⁴ The web page was customized for access via mobile device or personal computer without the need to download additional mobile apps. The data provided are linked into an overall picture of the person's condition in the form of a digital Medical Record, which is required for the decision to discontinue or resume treatment, and the need for further supervision. All the decisions on the prescribed supervision are automatically sent to the central oversight system at the level of the Republic of Croatia and this reduces the time needed for reporting. The reporting center and the digitalization of the process of epidemiological screening and processing from report to discharge is colloquially called the 'epidemiological mill' (Figure 1), since through its coordinated action it effectively resolves - or 'mills' - the epidemic. By creating 'digital memory of the development of the epidemic', it allows better preparation and a higher level of response for future period.³

'EPIDEMIOLOGICAL MILL'

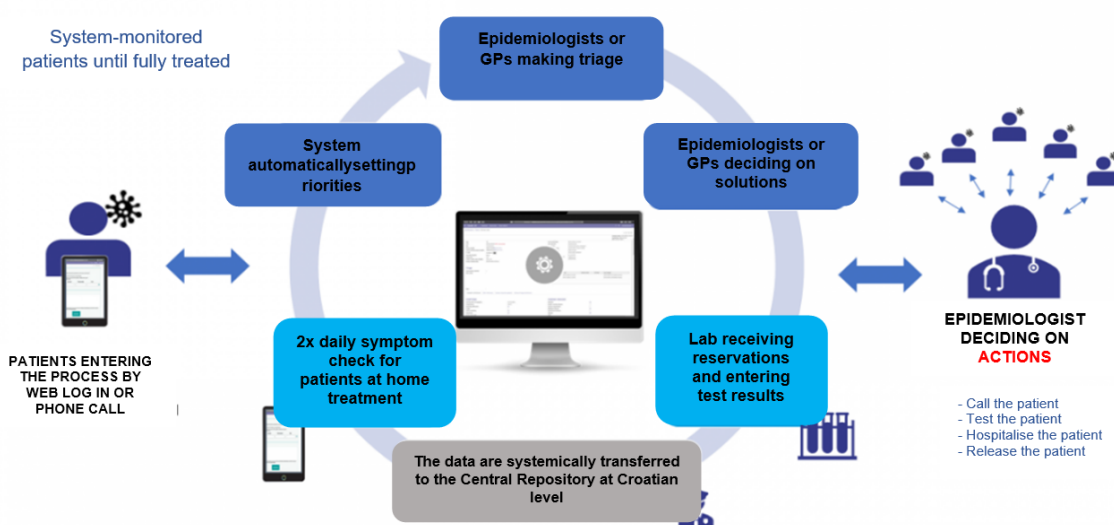


Figure 1. Schematic overview of the digitalization of the 'Epidemiological Mill' screening process

The task of our Institute is to carry out a whole range of tests and to monitor the environmental factors with a predominant impact on human health and safety of the Zagreb environment. Accordingly, the activities of drinking water and air quality monitoring continued daily throughout the coronavirus epidemic.

Implementation of anti-epidemic and preventive disinfection measures during the COVID-19 epidemic

Following evidence of the first case of infection with the new coronavirus on February 25, 2020 in Zagreb and the epidemiological assessment of the case, during which it was determined that the patient had previously been at work and at an event with a higher number of people in a small space, the epidemiological service - in preventing the spread of the epidemic - ordered urgent implementation of the measures of anti-epidemic disinfection. The measures were carried out the same night by the Division of Disinfection, Disinfestation and Pest Control (DDPC) of the Institute's Department of Epidemiology. Following the onset of new cases of infection during the COVID-19 epidemic, the Division continued, following orders by epidemiologists, to implement anti-epidemic measures in areas identified as requiring their implementation. In areas where numerous infected people resided, employees' protection became a challenge at each step - from carrying out the measures through handling protective equipment at the beginning of the process till the very end, including careful and safe disposal. Requests were raised during the epidemic for the regular implementation of measures in spatially demanding conditions within the prescribed time interval. In addition to the implementation of the measures, the DDPC Division has an advisory role for other institutions and organizations in assessing the need for and determining the manner of implementing preventive disinfection measures.

The implementation of disinfection measures is regulated by virtue of the Law on Protection of Population Against Infectious Diseases and is carried out during the COVID-19 epidemic also in accordance with applicable guidelines and recommendations.⁵

Water quality for human consumption during the COVID-19 epidemic

In order to ensure the functioning of society, and in the event of a further spread of the coronavirus epidemic, it is important to ensure continuous delivery of safe water for human consumption. Households, health care institutions, hospitals, and the food processing industry are very sensitive to water supply disruptions. Ensuring the safety of water for human consumption and ensuring hygienic conditions is essential to the protection of human health during the outbreak of infectious disease epidemics, including the COVID-19 epidemic. Due to

both the earthquake on 22nd March and the epidemic, and in line with the Croatian Institute for Public Health recommendations, it was emphasized and required to ensure continuous delivery of safe water for human consumption.

The most recent WHO document on water management and sanitation related to COVID-19 stated that no additional preventive and control measures are needed compared to what is already set out in the Guidelines of the World Health Guidelines on quality of water for human consumption, which are the basis for regulations and methods of water management for human consumption in Europe and also in the Republic of Croatia.⁶ The supply of safe water for human consumption in the Republic of Croatia during the current COVID-19 crisis has been uninterrupted and in accordance with applicable legislation. Tap water is safe for consumption and is safe from the risk of COVID-19 transmission, and there are no health-related reasons why consumers should be guided towards resorting to bottled water or other bottled beverages.

The monitoring of the safety of water for human consumption in the City of Zagreb area has been conducted under an Agreement concluded between the City Office for Health and the Dr. Andrija Štampar Teaching Institute for Public Health. The monitoring program fully covers the entire water supply system of the City of Zagreb and provides systematic monitoring of the safety of water for human consumption, identification of system sensitivities, and potential hazards. The program has been drafted in accordance with the applicable legislation and includes control of the safety of water from the central water supply system and local water supply system and local supply lines in the area of the city of Zagreb. The monitoring program has been conducted continuously, evenly, in a planned manner, throughout the year in all urban neighborhoods, and it includes hydrants of the distribution network, aggregated hydrants, water reservoirs, and public facility taps.

Based on the results of analyses obtained so far, we can conclude that the water for human consumption from the central public water supply system of the city of Zagreb is safe under the prescribed conditions of the Regulation, and these results indicate the importance of regular maintenance and control of the public water supply system for the purpose of detecting and preventing pollution.

Wastewater monitoring - a powerful tool to monitor viral exposure

Monitoring the transmission pathways of SARS-CoV-2 and its timely detection are further hampered by prolonged incubation time and the existence of asymptomatic cases of infection.⁷ As the testing of the entire population for SARS-CoV-2 for most countries still remains an impractical and economically inviable solution, there is a clear need for additional methods to

acquire an image, as complete as possible, of the course of the epidemic, the number of infected people, or the absence thereof.

According to the experience acquired until now, some SARS-CoV-2 genes have been successfully detected in stool samples^{8,9} and wastewater.^{10,11} Study data indicate that the SARS-CoV-2 virus can be detected in wastewater samples as early as a couple of weeks before confirmation of the first official positive cases.^{10, 12} Although the current assumptions by WHO leave little probability for wastewaters as an influential method of coronavirus transmission¹³, its increased incidence in the population is directly reflected also on its presence in wastewater, and it provides an indication of the relevance of the studies and monitoring of wastewater for the purpose of monitoring the circulation of the virus in the population.¹⁰

Systematic monitoring of biomarkers (biological and chemical) in wastewater has already been proven as a very useful method of monitoring population health^{11, 14, 15}, and the data obtained, among other things, prove to be very useful for monitoring both infectious and chronic non-infectious diseases.^{14, 15} Such monitoring represents great potential for temporo-spatial early warning system development.¹⁵

In June 2020, the City of Zagreb joined the pan-European SARS-CoV-2 monitoring project in the city sewage (EU-wide Wastewater Monitoring System for SARS-CoV-2 Surveillance). The project was led by the Joint Research Centre (JRC) and the European Commission's Directorate General for Environment (ENV). Participating in this pan-European project provides data on the presence or absence of SARS-CoV-2 in the waste waters of the City of Zagreb during the study period, with the aim of continuous monitoring of the presence of SARS-CoV-2 virus in Zagreb (and potentially in the rest of the Republic of Croatia). During the project, Andrija Štampar Teaching Institute for Public Health has been developing and introducing a method for wastewater testing for SARS-CoV-2 presence. Due to the absence of a standardized method, most laboratories conducting this type of tests have developed their own methods for the isolation and detection of SARS-CoV-2 in wastewater. Matrix complexity requires the selection and implementation of an adequate concentration technique and the use of RT-PCR for virus detection. Implementation of this method is an important step in monitoring wastewater and monitoring the COVID-19 epidemic as it contributes to an active monitoring of the course of epidemic, timely information on the decline or growth of the number of patients, and early implementation of anti-epidemic measures in case of a recurrence of the epidemic.

Sludge quality from wastewater treatment plants and COVID-19

More than 70% of sludge from wastewater treatment plants is used in agriculture to supply soil with organic

substances and nutrients such as nitrogen and phosphorus. Sludge analysis in Croatia has been conducted according to the applicable Regulation on the Management of Sludge from Wastewater Treatment Plants When Sludge is Used in Agriculture.¹⁶ For agricultural purposes, the only sludge that can be used is the one whose proportion of heavy metals and organic substances (polychlorinated biphenyls (PCB) and polychlorinated dibenzodioxin/dibenzofurans (PCDD/PCDF) may not exceed the values prescribed by the Regulation. Also, the sludge should be stabilized so that pathogens, potential causes of disease, have been destroyed.

Although the results from literature are still variable, ongoing research will upgrade the surveillance of COVID-19 in wastewater and sludge and compliment public health data.¹⁷ Wang et al¹⁸ highlighted that SARS-CoV existed even after three days in a patient's stool, and at a lower temperature (4°C) even after 17 days. SARS-CoV was detected in hospital wastewater, household sewage, and tap water after two days at 20°C and after up to 14 days at 4°C, indicating that temperature has a strong impact on virus persistence. There is evidence that higher temperatures have been associated with the rapid inactivation of enteric viruses, and temperature has been recognized as the most influential environmental factor for the survival of the virus in water and sludge due to increased protein denaturation and activity of extracellular enzymes.¹⁹ Water and sewage research have shown that infectious virus titer decreases faster at 25°C than at 4°C, which also confirmed that temperature is an important factor affecting the virus survival in water and thus in sludge.²⁰ Given the abovementioned, the potential risks associated with the COVID-19 outbreak should be assessed prior to each application of the sludge from the agricultural treatment plant. As virus transmission onsets through the airways, problems mainly relate to exposure to droplets and dust which can be released during sludge/slurry application. In wastewater treatment plants, the sludge intended for use in agriculture undergoes several treatments: it is thickened and dried, then often stabilized to block fermentation and limit odors. Finally, in some plants, it undergoes thermic processing, it is combined with lime or processed by composting to reduce the presence of microorganisms. Given the current incompleteness of the data on the inactivation of SARS-CoV-2 in sludge and wastewater, the analyses are based on knowledge of other viruses, such as enterovirus and animal coronavirus, in order to assess its resistance in sludge and against the treatment applied at the plant itself.

Since sludge generated during the epidemic underwent disinfection treatment, the French Agency for Food, Environmental Protection, Health and Occupational Safety (ANSES) considers that the risk of the presence of SARS-CoV-2 in sludge is low to negligible, given the effectiveness of all treatments applied in wastewater treatment plants.²¹ However, it recommends enhanced control to verify the correct implementation of

purification treatments and compliance with safety work rules that must be taken by treatment plant workers and operators who place sludge on agricultural land (appropriate protective equipment, hand washing, shower after the end of the activity, etc.).

For sludge produced during the COVID-19 epidemic that has not undergone any disinfectant processing, there is currently insufficient data to accurately define the level of contamination with SARS-CoV-2. In addition, we do not currently have enough knowledge of the persistence and development of the infectiousness of coronavirus over time to define the period of sludge storage after which the virus may be inactivated. Consequently, it is recommended that such wastewater sludge is not administered without prior disinfection.

In view of the lack of data to accurately document virus contamination in the sludge and the entire waste water treatment system, special scientific studies should be conducted, primarily by further monitoring of bacteriophages that infect intestinal bacteria, and which are suggested as good indicators of fecal or viral contamination.

Ecotoxicology reports during the COVID-19 epidemic

In the Dr. Andrija Štampar Teaching Institute for Public Health, acute toxicity tests are performed on two indicator organisms: *Pseudokirchneriella subcapitata* and *Daphnia magna*. These are the standard procedures to assess the impact of wastewater on organisms living in a natural water recipient into which wastewater is being sent. *Pseudokirchneriella subcapitata* are single-cell freshwater algae and the primary producer in freshwater while *Daphnia magna* is the primary consumer. In ecotoxicology studies, it is particularly important to select organisms of different trophic levels for best assessment of the potential toxic effects of wastewater.

Increased use of disinfectants for personal hygiene and professional use during the epidemic brought an increased load of wastewater with disinfectant agents. Such wastewater arrives to wastewater treatment plants and, after treatment, enters the watercourse or reaches the natural recipient without prior treatment, thus presenting a risk to the aquatic environment. Ethanol-based agents are most commonly used for disinfection. For effective disinfection, a minimum ethanol concentration of 70% is required. It should be noted that disinfectant efficacy tests on SARS-CoV-2 can be performed in only a few laboratories in Europe. In addition to ethanol, the active substances used in disinfectants are isopropanol, hydrogen peroxide, sodium hypochlorite, quaternary ammonium compounds, etc. Each active substance has specific chemical characteristics such as acute and chronic toxicity, reactivity, biological degradation, environmental retention, etc. The degradation products of the active substances and their environmental impact should not be neglected. A particular problem is

presented by chlorine-based disinfectants that act by destroying the cell membrane in cells of exposed organisms or by damaging proteins through the oxidation process. They may also react with other molecules, e.g. dissolved organic substances in water, resulting in very dangerous and toxic compounds such as trihalomethane.²² According to the available literature, between 2,000 and 5,000 tons of disinfectant were used in the Chinese town of Wuhan for indoor and outdoor disinfection, which then reached watercourses through the drainage system, thus possibly contaminating sources of drinking water.²²

No adverse effects on indicator organisms have been observed so far, according to the Institute data and wastewater analysis. Chemical and environmental toxicology indicators should be continuously monitored in wastewater, with the cooperation of other laboratories in the Republic of Croatia that test water quality. It is also necessary to use disinfectants rationally to ensure the required disinfectant effect, while at the same time preserving the water we depend on.

Air quality during the COVID-19 epidemic

Isolation measures implemented by many European countries, including Croatia, to stop the spread of the COVID-19 disease have led to a sharp reduction in economic activity, including a decline in road traffic in many cities, which has contributed to better air quality. In the area of the City of Zagreb, air quality monitoring is conducted at state network stations for continuous monitoring of air quality at three locations and at six locations with automatic measurement stations, which form the municipal network of stations. In addition to the state and municipal network of measurement stations listed, air quality monitoring in the area of the City of Zagreb is also conducted in stations for special purposes located in the following locations: Vrhovec, Jakuševac, Central Wastewater Treatment Plant of Zagreb (MM CUPOVZ), and Mirogojska 16. The automatic measurement station for monitoring the impact of road traffic in the area of Mirogoj residential zone (SO₂, NO₂, O₃, CO parameters) is a special purpose station and under the competence of the Andrija Štampar Teaching Institute of Public Health.

Additionally, in 2017, the web GIS app 'Environmental Map of the City of Zagreb' was established as an innovative tool for monitoring, assessing and managing environmental impacts on and risks for health.^{23, 24} As part of the 'Environmental Map of the City of Zagreb' program, the existing network for permanent monitoring of air quality has been upgraded with eight automated sensors to measure air pollution parameters with an on-line collection of data on pollution levels every 15 minutes. The upgrade of the existing air quality-monitoring network in the City of Zagreb with automated sensors is a novelty to the previous air quality monitoring (Figure 2).²⁵

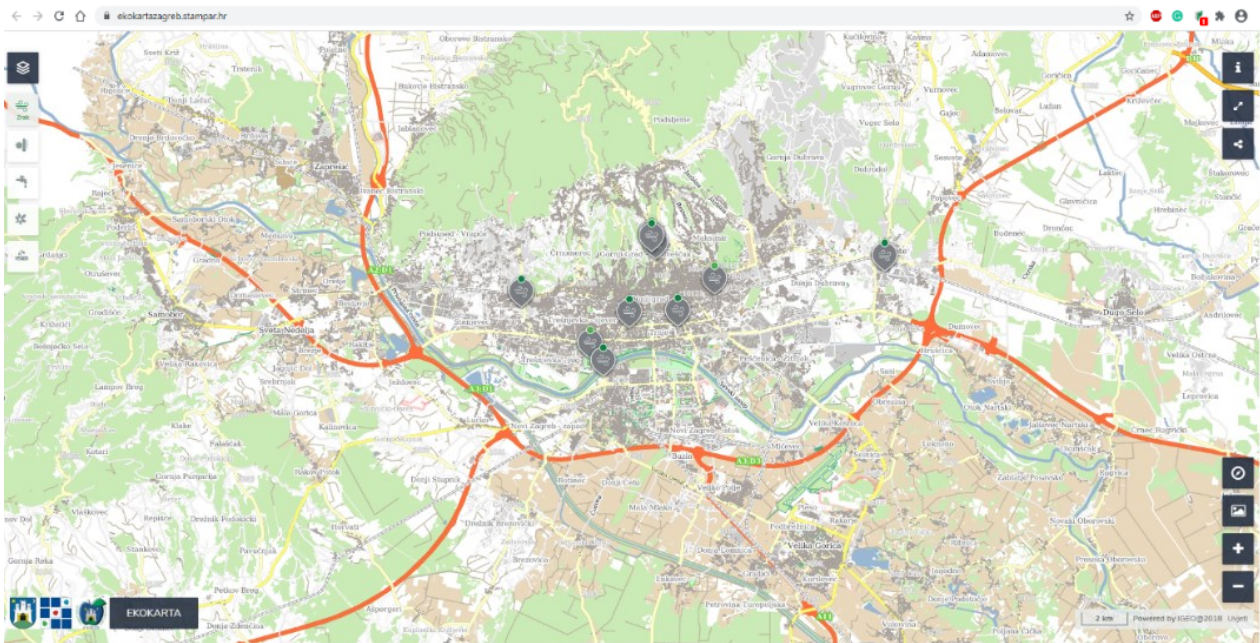


Figure 2. Web GIS app 'Environmental Map of the City of Zagreb', 'air' interface

The data from the specific purpose measurement station at Mirogojska location indicate that since the introduction of strict self-isolation measures, significant decreases in nitrogen dioxide concentrations (NO_2) have been observed in the air. Although a decrease of small floating $\text{PM}_{2.5}$ particles could also be expected, a significant reduction is still not possible to detect. This is probably since the main sources of $\text{PM}_{2.5}$ are diverse, while the main source of nitrogen dioxide (NO_2) is road transport.

In the first week since the start of self-isolation measures caused by the coronavirus, nitrogen dioxide levels (NO_2) dropped by as much as 70%. The loosening of measures has brought about a slight increase in concentrations, so we are now at about 50% of the usual concentrations in the City of Zagreb (Figure 3). This positive impact on air quality is the result of compliance with anti-epidemic measures at the level of the City of Zagreb and the Republic of Croatia.²⁶

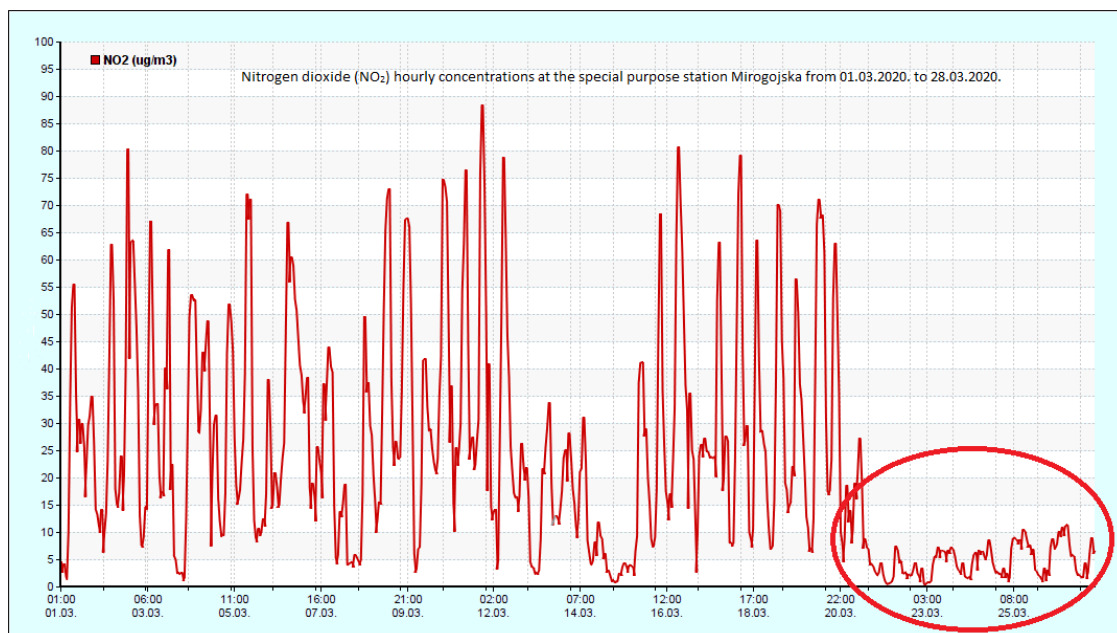


Figure 3. Nitrogen dioxide (NO_2) hourly concentrations at the special purpose station Mirogojska (source: Andrija Štampar Teaching Institute of Public Health)

Air quality of indoor premises

Health issues that can be caused by biological agents are as diverse as, for example, infections, disease of upper or lower respiratory system, occupational asthma, various forms of chronic obstructive pulmonary disease, various forms of allergies, poisoning, and malignancy. Infections are most often caused by parasites, viruses, fungi and bacteria. Inhalation of mould conidia, organic dust such as flour, dust from animal excretions, bacterial spores, can cause various respiratory diseases and the development of allergy symptoms.

Given the increased number of complaints about poor air quality in the working environment over the past few years, it has been decided to systematically approach the attempt to collect data that could contribute to the resolution of these problems. To this end, methods for microbiological analyses of air have been introduced according to internationally recognized ISO standards which relate to parameters of determination of the total number of bacteria and mould in air, based on the comparison of air between indoor and outdoor areas of living and working environment.

Institutional up-to-date method establishment testing of the air samples for viral RNA have been implemented recently. There is a great difference between biological agents and other risk substances. For example, some biological agents can replicate in certain favorable conditions. It is hard to establish exposure limits because of lack of documentation (such as guidelines, standards and documentation).

COVID-19 and hay fever

The coronavirus pandemic appeared at the time of the flu season and hay allergies. It is therefore important to know how to recognize the symptoms of hay fever and to distinguish them from the symptoms caused by COVID-19. Hay fevers represent a significant public health problem today. Pollen is defined as the most

powerful natural aeroallergen and the most common cause of allergic respiratory diseases, particularly in urban environments. In 2001, Croatia recognized the problem caused by allergenic pollen and established a network of public health institutes conducting monitoring of pollen in the air. In Zagreb, the program for monitoring allergenic pollen has been conducted since 2002.

The basic feature of hay fever is the yearly periodicity. Knowledge of the spatial and temporal dynamics of pollen grains in air over the year is one of the main factors for hay fever prevention and treatment. In this context, the best prevention is to reduce exposure to allergenic pollen, which is very difficult in practice. The Institute's website (www.stampar.hr) publishes daily data on the presence of allergenic pollen. Daily advice is also available for citizens through the free mobile app Pollen Forecast. Due to a high concentration of allergenic pollen in the air and overlapping with the coronavirus epidemic, the Institute's website has published a table of allergy symptoms and their similarity and diversity in relation to the symptoms caused by the COVID-19 virus (Table 1).

CONCLUSION

Under the legislative framework, the Institute quickly responded and adapted its activities to the new situation caused by the COVID-19 pandemic. Daily reporting via the media on the number of people tested on COVID-19 and the number of patients, and the publication of the latest guidelines in line with the crisis headquarters' recommendations have become routine. Throughout the coronavirus crisis, all Institute departments have carried out their regular activities continuously whilst adapting to the current conditions. The Institute's Department for Mental Health has opened eight call numbers for advisory assistance to the public either because of the epidemic or because of the earthquake (22nd March) that further affected Zagreb or because of any inquiries related to mental health.

Table 1. Symptom differences - cold, flu, COVID-19, and allergy

Symptom	Cold	Flu	Coronavirus	Allergy
high body temperature	rarely	often	yes	no
cough	rarely	yes	dry cough	sometimes
shortness of breath	rarely	yes	yes	sometimes
general weakness	sometimes	yes	yes	no
headache	rarely	yes	sometimes	sometimes
muscle aches	mild	frequent	yes	no
sneezing	often	sometimes	no	yes
sore throat	often	sometimes	sometimes	no
runny nose	sometimes	sometimes	rarely	yes
relief after use of allergy medicine	no	no	no	yes

Source: ECDC (available at: <https://www.ecdc.europa.eu/en/covid-19/questions-answers>)

The need for recognized guidelines, standards, eligibility criteria for the correct interpretation of environmental results is clear. In addition, sampling and testing methods should be standardized. Accreditation of the Department of Environmental Protection and Health Ecology according to HRN EN ISO/IEC 17025:2017 – General Requirements for the Competence of Testing and Calibration Laboratories is a step further in raising awareness of the environmental monitoring importance. Integrated management system in the Institute certified according the following standards: ISO 9001:2015 Certificate - Quality Management Systems, ISO 14001:2015 Certificate - Environmental Management Systems and ISO 45001:2018 Certificate - Occupational Health and Safety Management Systems helped significantly in faster and more efficient demands during the pandemic.

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