November 2018 ISSN 1650-3414 Volume 29 Number 3

# eJIFCC

Communications and Publications Division (CPD) of the IFCC

**Editor-in-chief: Prof. János Kappelmayer, MD, PhD**
Faculty of Medicine, University of Debrecen, Hungary
e-mail: ejifcc@ifcc.org

# The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

## In this issue

# Laboratory Medicine: Meeting the needs of the Mediterranean nations Part 1

## In this issue

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# Foreword from the editor-in-chief

János Kappelmayer, MD, PhD

The present issue of the eJIFCC is composed of articles focused on topics discussed at the conference entitled "**Laboratory medicine: meeting the needs of the Mediterranean nations**" The conference was held this year in Rome between July 2-4, with professor Sergio Bernardini as the Conference President.

The current issue is part one of a two-part series, and contains articles covering three sections of the conference, namely:

- Transmissible diseases in the Mediterranean area;
- Training and education in laboratory medicine;
- Improving health with emerging technologies.

*János Kappelmayer*

Editor

*In this issue: Laboratory medicine–meeting the needs of Mediterranean nations*

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# Laboratory medicine: meeting the needs of Mediterranean nations

Guest Editors: Sergio Bernardini[1], Bernard Gouget[2,3,4,5,6]

[1] *Department of Experimental Medicine, University of Tor Vergata, Rome, Italy*
[2] *Chair, IFCC Committee on Mobile Health and Bioengineering in Laboratory Medicine (C-MHBLM)*
[3] *SFBC-International Committee*
[4] *General Secretary of the International Francophone Federation of Clinical Biology and Laboratory Medicine (FIFBCML)*
[5] *Counselor for Public Health-FHF*
[6] *President Human Health Care Committee-COFRAC*

ARTICLE INFO

***Corresponding author:***
Sergio Bernardini
Department of Experimental Medicine
University of Tor Vergata
Rome
Italy

EDITORIAL

The Mediterranean Sea connects countries with different traditions, lifestyles and religions, but all have been shaped by this extraordinary geographical basin, which produced the oldest civilizations. Today, unfortunately, the Mediterranean countries also share wars, terrorism, poverty and large-scale migration. This conference highlights the value of laboratory medicine for a greater effectiveness and safety with the potential to impact numerous health system outcomes at national and regional level and to improve security in the Middle East region with on-site opportunities for diagnosis and care to help victims of war and sociopolitical instability as well as care of refugees. It will open a new dialogue for scientific solutions to improve healthcare delivery under these extraordinary circumstances as well as to demonstrate the capacity of laboratory medicine network of excellence to combine different expertise in a single joint action to become of added value. At the healthcare level,

South European Mediterranean countries are faced with a double burden. They must maintain common policies to fight against traditional diseases, which rely mainly on vaccination policy, while having to face new characteristic diseases from developed countries (cancer, cardiovascular diseases, HIV, hepatitis, obesity, etc.). In addition to the epidemiological transition, other transitions are adding up (demographic, organizational and democratic). Financial resources remain limited and the post-Arab spring context gave rise to an increasing demand of populations for better access to health care for all and at the lowest cost. Such is the complex health context facing South European Mediterranean countries. On the other hand, in the Middle East and North Africa region, non-communicable diseases such as cardiovascular disease (up by 44%), stroke (up 35%), metabolic diseases and diabetes (up 87%), obesity, maternal mortality are causing more premature death and disability than they did in the past. Potentially preventable risk factors such as poor diets, high blood pressure, high body mass index (an indicator of obesity and overweight), and smoking are contributing to the growing burden of non-communicable diseases in the region. Tuberculosis is still endemic and some transmissible diseases (HIV) may reoccur. In the last few months, the world has been vividly reminded by the Ebola epidemic and by the resurgence of polio and of MERS Coronavirus that health problems do not stop at borders. In the alarming context of risk of Zika virus (ZIKV) transmission in the Euro-Mediterranean area, there is a need to examine whether capacities to detect, diagnose and notify ZIKV infections in the region are in place and whether ongoing capacity-building initiatives are filling existing gaps. Moreover collaborating with Mediterranean countries is important, through the networks of pharmacovigilance, to be aware of antimicrobial resistance by extending the surveillance and laboratory experience, while reviewing and strengthening shared vaccination strategies. Countries in Europe and the Mediterranean face several common health challenges, including, to different extents, the double burden of diet- and physical inactivity related chronic diseases and of nutritional deficiency disorders. Migration and health is another common challenge where EU and non-EU countries in the Mediterranean are faced with large unexpected flows of migrants, refugees and asylum seekers many of whom have particular health needs. This constitutes a third and serious burden that we cannot underestimate and that should be addressed jointly by mobilizing needed resources within a shared framework. Deteriorating health and unnecessary deaths and suffering also due to the current turmoil in several areas in the region are indeed our main common challenges and we need to devise shared strategies to combat them and overcome the risk they impose on our societies. Finally, there is a need for more stringent relationships between Mediterranean countries to realize advancements in education and curriculum of laboratory professionals.

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# Communicable diseases in the Mediterranean region

Ghassan Shannan

*Medical Care Centre, Damascus, Syria*

ARTICLE INFO

***Corresponding author:***
Ghassan Shannan
Director of Laboratory
Medical Care Centre
Damascus
Syria
E-mail: ghassanshannan@gmail.com

***Key words:***
communicable diseases, tuberculosis, HIV/AIDS, hepatitis

ABSTRACT

Communicable diseases still pose a health hazard and are a major cause of death in many parts of the world. Tuberculosis is one of the top 10 causes of death worldwide with an annual burden of 1.7 million. Global deaths in 2016 for other communicable diseases included 1.34 million from hepatitis; 1.0 million from HIV-related causes; and almost half a million from malaria. Outbreaks of vaccine-preventable diseases like polio, measles, rubella and other life-threatening diseases also pose a significant threat to various communities around the world. In this paper, we shed some light on the epidemiology of communicable diseases in the Mediterranean Region and conclude that socioeconomic differences between the north and the south Mediterranean lead to differences in the epidemiology of communicable diseases.

## INTRODUCTION

Socioeconomic, environmental and behavioural factors, as well as international travel and migration, foster and increase the spread of communicable diseases.

Vaccine-preventable, foodborne, zoonotic, health care-related and communicable diseases pose significant threats to human health and may sometimes threaten international health security.

In cooperation with governments, WHO/Europe develops norms and standards, guidance and public health tools to help countries implement effective disease prevention and control programmes and address their risk factors.

Globally, an estimated 3 out of 10 deaths are attributed to communicable diseases. Among these, the World Health Organization (WHO) global burden of disease baseline projections in 2008 revealed that the African Region contributes the highest number of deaths with 41%, followed by the Eastern Mediterranean and South-East Asia Regions, each contributing 15% of the deaths due to infectious and parasitic diseases.

**Table 1** **Communicable diseases [* Vaccine-preventable communicable diseases]**

| | |
|---|---|
| HIV/AIDS | * Hepatitis |
| Influenza | * Measles |
| Rotavirus | * Rubella |
| Tuberculosis | * Poliomyelitis |
| Malaria | * Diphtheria |
| Brucellosis | * Japanese Encephalitis |
| Typhoid | * Mumps |
| Cholera | * Neonatal Tetanus |
| Sexually Transmitted Infections | * Tetanus |
| Vector-Born & Parasitic Diseases | * Yellow Fever |
| - | * Congenital Rubella Syndrome |

WHO defines communicable disease as infectious diseases which are caused by pathogenic microorganisms, such as bacteria, viruses, parasites or fungi, that can be spread, directly or indirectly, from one person to another.

Under this definition, one can include a long list of diseases. However, in this publication we will focus on the most important communicable diseases, which have significant impact on public health (Table 1).

**Table 2** List of the Mediterranean region countries included in the review

| Southern European Coast from West to East | Levantine Coast from North to South |
|---|---|
| Gibraltar | Syria |
| Spain | Cyprus |
| France | Lebanon |
| Monaco | Israel |
| Italy | Palestine, Gaza Strip |
| Malta | **Northern African Coast from East to West** |
| Slovenia | |
| Croatia | Egypt |
| Bosnia & Herzegovina | Libya |
| Montenegro | Tunisia |
| Albania | Algeria |
| Greece | Morocco |
| Turkey | - |

The review includes the countries listed in Table 2 in the Mediterranean Region.

## RESULTS

Table 3 presents the population of each Mediterranean country, the expenditure on health per capita and the expenditure on health as percentage of gross domestic product (GDP). The table shows clearly the big difference in spending on health between the Northern and Southern Mediterranean countries.

### *HIV/AIDS*

The incidence rate in northern Mediterranean countries is higher than in southern Mediterranean countries (Table 4).

Of the people living with HIV (PLWHIV), a variable percentage are receiving anti-retroviral therapy (ART) around the Mediterranean. The lowest percentage is in Egypt, with 27%; and the highest in Spain, with 77%. There is no correlation between the expenditure on health and the number of PLWHIV (Table 4).

**Table 3** Demographic and health care spending data for countries in the Mediterranean region

| Country | Population | Expenditure on health per capita Intl. $ | Expenditure on health as % of GDP |
|---|---|---|---|
| Albania | 2,897,000 | 615 | 5.90 |
| Algeria | 39,666,000 | 932 | 7.20 |
| Bosnia & Herz. | 3,810,000 | 957 | 9.60 |
| Croatia | 4,240,000 | 1,652 | 7.80 |
| Cyprus | 1,165,000 | 2,062 | 7.40 |
| Egypt | 91,508,000 | 594 | 5.60 |
| France | 64,365,000 | 4,508 | 11.50 |
| Gibraltar | N/A | N/A | N/A |
| Greece | 10,955,000 | 2,098 | 8.10 |
| Israel | 8,064,000 | 2,599 | 7.80 |
| Italy | 59,798 | 3,239 | 9.20 |
| Lebanon | 5,851,000 | 987 | 6.40 |
| Libya | 6,278,000 | 806 | 5.00 |
| Malta | 419,000 | 3,072 | 9.80 |
| Monaco | 38,000 | 7,302 | 4.30 |
| Montenegro | 26,000 | 888 | 6.40 |
| Morocco | 34,378,000 | 447 | 5.90 |
| Palestine | N/A | N/A | N/A |
| Slovenia | 2,068,000 | 2,698 | 9.20 |
| Spain | 46,122,000 | 2,966 | 9.00 |

| | | | |
|---|---|---|---|
| Syria | 18,502,000 | 376 | 3.20 |
| Tunisia | 11,254,000 | 785 | 7.00 |
| Turkey | 78,666,000 | 1,036 | 5.40 |

### *Hepatitis*

The incidence of hepatitis A (HAV) in south Mediterranean countries is much higher than in north Mediterranean countries, especially among school children. This is also the case for the hepatitis B virus (HBV) and the hepatitis C virus (HCV), with highest incidence rate of HCV (10.70%) being in Egypt (Table 4).

### *Tuberculosis*

The incidence rate of tuberculosis (TB) is much higher in the southern Mediterranean despite the increase of TB cases in European countries among PLWHIV. The same finding applies to multi-drug resistant (MDR) TB, where the incidence is higher in the southern Mediterranean (Table 4).

### *Schistosomiasis in Egypt*

Schistosomiasis is a parasitic disease caused by blood flukes (Trematodes) of the genus Schistosoma (S.). It is well documented that schistosomiasis haematobium was endemic in Ancient Egypt. Infection was diagnosed in mummies 3000, 4000 and 5000 years old.

Schistosomiasis haematobium was highly prevalent (60%) both in the Nile Delta and Nile Valley South of Cairo. However, by the end of 2010, in the whole country only 29 villages had a prevalence of >3% and none had more than 10%.

A recent study in 2016 by a group of scientists from Tanta University report that the prevalence of S. mansoni infection was found to be 1.8% among schoolchildren of the studied areas.

**Table 4** Incidence rates of communicable diseases in Mediterranean countries

| Country | HIV Incidence /$10^5$ | PLWHIV | Receiving ART % | HBV | HCV | TB Incidence /$10^5$ | MDR TB Incidence /$10^5$ |
|---|---|---|---|---|---|---|---|
| Albania | 0.15 | 1,700 | 30% | - | - | 16 | 0.42 |
| Algeria | 0.04 | 13,000 | 76% | **2.30%** | **8.90%** | 70 | 1.10 |
| Bosnia & Herz. | N/A | N/A | N/A | - | - | 32 | N/A |
| Croatia | 0.04 | 1,500 | 70% | 2.70/$10^5$ | - | 12 | N/A |
| Cyprus | N/A | N/A | N/A | 0.20 | 0.2/$10^5$ | 5.6 | N/A |
| Egypt | 0.03 | 11,000 | 27% | **1.40%** | **10.70%** | 14 | 2.20 |
| France | 0.21 | 180,000 | 78% | - | - | 7.7 | 0.13 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| Gibraltar | N/A | N/A | N/A | 0.20/$10^5$ | - | N/A | N/A |
| Greece | N/A | N/A | N/A | - | - | 4.4 | 0.10 |
| Israel | N/A | N/A | N/A | 0.20/$10^5$ | - | 3.5 | 0.30 |
| Italy | 0.12 | 130,000 | 58% | 0.60/$10^5$ | 0.3/$10^5$ | 6.1 | 0.24 |
| Lebanon | 0.04 | 1,200 | 51% | - | - | 12 | 0.48 |
| Libya | N/A | 4,400 | 73% | **3.20%** | **1.20%** | 40 | 1.90 |
| Malta | N/A | N/A | N/A | 4.20/$10^5$ | 0.7/$10^5$ | 13 | N/A |
| Monaco | N/A | N/A | N/A | - | - | 0 | N/A |
| Montenegro | 0.23 | <500 | 60% | - | - | 16 | N/A |
| Morocco | 0.05 | 22,000 | 48% | **1.80%** | **1.60%** | 103 | 1.80 |
| Palestine | N/A | N/A | N/A | - | - | N/A | N/A |
| Slovenia | N/A | N/A | N/A | - | - | 6.5 | N/A |
| Spain | 0.18 | 140000 | 77% | 1.10/$10^5$ | - | 10 | 0.50 |
| Syria | <0.01 | 175 | 70% | **3.10%** | **0.40%** | 21 | 2.00 |
| Tunisia | 0.05 | 2900 | 29% | **0.80%** | **1.90%** | 38 | 0.40 |
| Turkey | N/A | N/A | N/A | **4.00%** | **1.00%** | 18 | 0.70 |

### *Leishmaniosis in Syria*

Due to the unrest in Syria over the past seven years, the incidence of leishmaniosis cases has significantly increased: from 35,876 cases in 2014; to 50,972 cases in 2015; and to 48,311 cases in 2016. Scientists attribute the increase to poor sanitation and the accumulation of rubbish and garbage around towns and villages, as many municipals authorities are not able to clear the garbage regularly.

### Polio in Syria

Outbreaks of polio cases have been reported by WHO country offices in several parts of Syria (Table 5). These outbreaks are attributed to poor sanitation and the inability of International organisations to implement the immunization programme in the hard-to-reach areas.

## DISCUSSION

The epidemiology of communicable diseases is affected by the socioeconomic status of the populations of the north and south Mediterranean. HIV/AIDS prevalence is higher in the northern than the southern Mediterranean countries; largely attributed to the conservative societies and the sexual behaviour of the populations, as multi-partners sexual relationships and sex outside marriage are not widespread in southern Mediterranean countries.

**Table 5** Syria Polio (cVDPV2) outbreak*

| Governorate | District | Number of cVDPV2 cases to date |
|---|---|---|
| Deir Ez-Zor | Mayadeen | 58 |
| Deir Ez-Zor | | 01 |
| Boukamal | | 12 |
| Raqqa | Tell Abyad | 01 |
| Thawra | | 01 |
| Homs | Tadmour | 01 |
| **Total** | | **74** |

** Situation Report #34, February 2018*

The prevalence of hepatitis, brucellosis, typhoid, schistosomiasis and vaccine-preventable communicable diseases is much higher in the southern Mediterranean countries; mainly due to poor hygiene and inefficiency of the health systems to provide viable health services in all areas, especially in remote and hard-to-reach areas. The outbreak of Hepatitis A among schoolchildren is an example of the inefficiency of the health system to provide proper hygiene in schools and public places.

More effort and funds are needed to combat many killer diseases in southern parts of the Mediterranean to eradicate hepatitis, tuberculosis, typhoid, brucellosis and other communicable diseases.

## REFERENCES

1. Annual Epidemiological Report 2015, Hepatitis C, European Centre for Disease Prevention and Control.

2. Annual Epidemiological Report 2015, HIV and AIDS, European Centre for Disease Prevention and Control.

3. Asmaa Gomaa, et al, Hepatitis C infection in Egypt: prevalence, impact and management strategies, Hepatic Medicine: Evidence and Research 2017:9, PP. 17-25.

4. Communicable Disease Threats, European Centre for Disease Prevention and Control.

5. Elgharably et al, Hepatitis C in Egypt – past, present, and future, International Journal of General Medicine 2017:10, pp.1-6.

6. Epidemiological update: hepatitis A outbreak in the EU/EEA mostly affecting men who have sex with men, European Centre for Disease Prevention and Control.

7. EWARS, weekly bulletin of WHO Country Office, Syria

8. Global Tuberculosis Report 2017, World Health Organisation.

9. Haq Z., Communicable Diseases in the Eastern Mediterranean Region: Prevention and Control, 2010 – 2011, Eastern Mediterranean Health Journal, Vol. 19, No. 10, pp. 881 – 891.

10. Measles and Rubella Surveillance 2017, European Centre for Disease Prevention and Control.

11. Ministry of Health of Syria, CDC annual reports.

12. Mohamed A. Daw, et al, Hepatitis C Virus in North Africa: An Emerging Threat, The Scientific World Journal, Volume 2016 (2016), Article ID 7370524, 11 pages, http://dx.doi.org/10.1155/2016/7370524.

13. Rash Raslan et al, Re-Emerging Vaccine-Preventable Diseases in War-Affected Peoples of the Eastern Mediterranean Region—An Update, Front Public Health. 2017; 5: 2.

14. Rashida M.R. Barakat, Epidemiology of Schistosomiasis in Egypt: Travel Through Time, Journal of Advance Research, (2013) 4, 425 - 432

15. Syria's health crisis: 5 years on, the Lancet, Vol. 387, March 12, 2016, PP. 1042-1043.

16. Tozun N. et al, Seroprevalance of hepatitis B and C infections in Turkey, Turk J Gastroenterol 2017; 28: 147-8, pp.147-148

17. WHO international: http://www.who.int/, WHO Eastern Mediterranean: http://www.emro.who.int/index.php, WHO Europe: http://www.euro.who.int/en/home

*Meeting the needs of Mediterranean nations: transmissible diseases in the Mediterranean area*

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# Are medical laboratories ready for the diagnosis of parasitic diseases?

Ahmet Özbilgin

Department of Parasitology, Faculty of Medicine, University of Manisa Celal Bayar, Manisa, Turkey

ARTICLE INFO

***Corresponding author:***
Ahmet Ozbilgin
Department of Parasitology
Faculty of Medicine
University of Celal Bayar
Dekanlik Binasi, Uncubozkoy
45030 Manisa
Turkey
Phone: +90 236 233 19 20
Mobile: +90 532 134 35 31
Fax: +90 236 233 14 66
E-mail: a.ozbilgin@yahoo.com

***Key words:***
parasitic disease, refugee, mediterranean, migrant, diagnosis

***Disclosures:***
The author declares no conflicts of interest.

ABSTRACT

Economic instability, destabilisation with armed conflagration, religious and ethnic conflicts are the most important driving factors for migrations towards Europe. Mediterranean countries are important route for refugees who emigrate from the Middle East, Africa and Asia.

Viral, bacterial, parasitic and fungal diseases carried by these refugees constitute a significant health risk in Mediterranean countries.

Parasitic diseases, such as intestinal parasites, pediculosis, scabies, lymphatic filariasis, schistosomiasis, malaria and leishmaniasis, which may reach the Mediterranean region trough migrations, were briefly reviewed and the precautions to be taken were mentioned.

It is of utmost importance that laboratories in the Mediterranean countries pay particular attention to parasitic infections, especially by using experienced staff and appropriate diagnostic methods in combating such infections.

The diagnosis and screening of all these infections among the refugees can be done by some basic parasitological laboratory methods such as direct methods for stool samples (direct wet smear, concentration methods, permanent stains, special stains), methods

for blood samples (thick and thin blood films, blood concentration procedures), direct methods for urogenital specimens, serological methods, molecular methods and rapid diagnosis kits. These methods can be easily learned by laboratory employees.

We suggest that migratory related infections study group should be established in Mediterranean countries, which should inform each other by sharing their findings and observations with officials in congresses and symposiums, and should cooperate on this issue and prepare training and workshop programs and health education programs in these countries should be updated about the health and risk factors of refugees.

❖❖❖❖❖

## 1. INTESTINAL PARASITES

Parasitic diseases that do not require any intermediate hosts or vectors and are directly transmitted to humans such as Enterobiosis, hymenolepiosis, giardiosis, amoebic dysentery can be found in almost every area and every society in the world.

In order for these diseases to become widespread, people must be in a collective presence or in close contact. These diseases spread easily and even constitute epidemics in places like schools, factories, prisons, refugee camps, temporary tent camps that people live together during a war and nursing homes.

Amoebiosis, one of the most important intestinal parasitic diseases in the world, is seen in 10% of the world population and 40 million people die each year from this disease. Giardiosis has been reported in 200 million people worldwide. Other than these parasites, Strongyloides species are one of the most important intestinal parasites that should be considered in the refugee populations. Ascaris lumbricoides, Trichuris trichura, hookworms and Taenia species need to be kept in mind as well (1).

## 2. PEDICULOSIS

Every year over 100 million cases of pediculosis are reported in the world. In the United States, head lice are found in 6-12 million children aged between 3 to 12 years each year. In studies conducted on at least 1000 students in various countries of the world, it has been found that; 49% of the students in France, 20% in Israel, 25% in England and 37% in Nigeria have been subjected to head lice (2,3).

Pediculosis spread easily and even constitute epidemics in places such as schools, factories, prisons, refugee camps, temporary tent camps during the war and nursing homes (4,5).

## 3. SCABIES

Scabies is a common disease of the world and can be seen in every age, race, region, climate and social fraction. In particular, it is noteworthy that the frequency of the disease increases in autumn and winter seasons when people have to live together and decreases in summer seasons. Sporadic or epidemic occurrences may be seen while every individual shows a different resistance to scabies.

Scabies is responsible for confined epidemics nowadays but still it is one of the important diseases that may create epidemics for the countries that receive large amounts of refugees in the recent years. It is known that the number of people infected with scabies tends to rise where people have to live together (6,7).

## 4. VECTOR BORNE DISEASES

**There are four important tropical diseases associated with refugees which can cause epidemics in Mediterranean countries.**

### 4.1. Lymphatic filariasis

In Mediterranean countries, mosquito species which are vectors of lymphatic filariasis are commonly found in many countries. New foci of lymphatic filariasis may be formed in Mediterranean countries through people migrating from endemic areas or people staying in these areas during their migration process (8,17).

### 4.2. Schistosomiasis

Among migrants, especially those who complain of hematuria should be carefully examined in terms of schistosoma. It should be noted that intermediate host snails can be found in some Mediterranean countries. Special efforts should be made in order to prevent the spread of the currently controlled disease. Schistosomiasis is seen in northern parts of Syria, near the border to Turkey (9,10,17).

### 4.3. Malaria

Vector mosquito species are common in all Mediterranean countries. Considering that some countries have autochtonous cases, special attention should be given to combat malaria in these countries and immigrants should be screened in terms of Plasmodium species via laboratory tests (11).

### 4.4. Leishmaniasis

Visceral and cutaneous leishmaniasis are present in some of the Mediterranean countries while vector Phebotomus species have a wide distribution in the region. Syria, which has the biggest number of refugees nowadays is one of the top countries in the world for cutaneous leishmaniasis prevalance. For this reason, laboratory tests must be done carefully to screen for the visceral and cutaneous leishmaniasis among immigrants. In addition, resistance to meglumine antimonate (Glucantime) which is used in the treatment of cutaneous leishmaniasis in the Middle East, has been reported. If resistant cases can not be detected in time and the number of cases as well as areas increase, the fight against this disease will be very difficult and problematic (12-17).

## CONCLUSION

It is important that the laboratories in the Mediterranean countries specifically address this issue, especially in the case of parasitic infections by utilising experienced personnel and appropriate diagnostic methods to combat these infections. Gastrointestinal parasitic infections, Scabies and Pediculosis which can be transmitted directly between humans in places where people are living with the refugees and where there is a lack of hygiene, can cause epidemics in these countries. Unless necessary measures are taken, Malaria, Leishmaniasis, Lymphatic filariasis and Schistosomiasis, which are among the most important tropical diseases in the world and are encountered in the geographical paths where refugees live and migrate, can create new foci and epidemics in the Mediterranean countries.

## REFERENCES

1. Özcel,M. General Parasitology, Medical Parasitic Diseases (Eds. Özcel MA, Özbel Y, Ak M,), Turkish Society of Parasitology No: 22, 2007;s.3-75

2. Frankowski BL, Weiner LB. Head lice. Pediatrics. 2002;110(3): 638-643

3. Chosidow O. Scabies and pediculosis. Lancet. 2000;355: 819-26, 2000

4. Akısü Ç, Sarı B, Aksoy Ü, Özkoç S, Öztürk S. Investigation of Pediculus capitis prevalence in a primary school in Narlıdere and comparison with previous results. Turkish Journal of Parasitology, 2003; 27(1):45-48

5. Kokturk A, Bugdayci R, Sasmaz T, Tursen U, Kaya TI, Ikizoglu G. Pediculosis, Int.J Dermatol, 2003;42(9): 694-698

6. Arnold HL, Odom RB, James WD. Andrew's Disease of the skin. 8. Edition. s.523-527.(10). WB Saunders Comp. Philedelphia 1990

7. Budak S, Yolasığmaz A, Scabies. Medical Parasitic Diseases (Eds. Özcel MA, Özbel Y, Ak M,), Turkish Society of Parasitology No: 22, 2007;s.379-397

8. Kuman H.A. Filariasis., Medical Parasitic Diseases (Eds. Özcel MA, Özbel Y, Ak M,), Turkish Society of Parasitology No: 22, 2007;s.601-618

9. Berkin T, Berke Z. About Bilharzia disease. Turkish Hygiene and experimental biology journal, 1950;10(1): 145 – 164

10. Özcel,M. Schistosomiasis, Medical Parasitic Diseases (Eds. Özcel MA, Özbel Y, Ak M,), Turkish Society of Parasitology No: 22, 2007;s.475-497

11. Özcel,M. Malaria, Medical Parasitic Diseases (Eds. Özcel MA, Özbel Y, Ak M,), Turkish Society of Parasitology No: 22, 2007;s.79-132

12. Magill AJ, Grögl M, Gasser RA Jr, Sun W, Oster CN. Visceral infection caused by Leishmania tropica in veterans of Operation Desert Storm, N Engl J Med., 1993;13;328(19):1383-7.

13. AlSamarai, A.M., AlObaidi, H.S. Cutaneous leishmaniasis in Iraq. J Infect Dev Ctries. 2009;Mar 1; 3(2):123-9.

14. Tayeh A, Lama J, Cairncross S, Twenty years of cutaneous leishmaniasis in Aleppo, Syria, Trans R Soc Trop Med Hyg, 1997;91,657-659.

15. Hadighi, R, Mohebali M, Boucher P. et al, Unresponsiveness to glucantime treatment in Iranian cutaneous leishmaniasis due to drug-resistant Leishmania tropica parasites. PLoS Med, 2006;3:e162.

16. Report of Syria Ministry of Health, Damascus, Syria (2012)

17. Eckstein B.,Primary care for refugees. Am Fam Physician. 2011 Feb 15;83(4):429-36.

*Meeting the needs of Mediterranean nations: transmissible diseases in the Mediterranean area*

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# Travel, migration and emerging infectious diseases

Nicolas Vignier[1,2], Olivier Bouchaud[3,4]

[1] *INSERM, Sorbonne Université, Institut Pierre Louis d'Épidémiologie et de Santé Publique (IPLESP), Department of Social Epidemiology, Institut Convergences et Migration, Paris, France*

[2] *Groupe hospitalier Sud Ile-de-France, Department of Infectious and Tropical Diseases, Melun, France*

[3] *Paris 13 University, Sorbonne Paris Cité, Groupe hospitalier Paris-Seine-Saint-Denis, Avicenne Universitary Hospital, Assistance Publique-Hôpitaux de Paris (AP-HP), Department of Infectious and Tropical Diseases, Bobigny, France*

[4] *Laboratoire Éducations et Pratiques de Santé (LEPS EA 3412), Bobigny, France*

## ARTICLE INFO

***Corresponding author:***
Nicolas Vignier
Equipe de Recherche en Epidémiologie Sociale
Institut Pierre Louis d'Épidémiologie
et de Santé Publique (IPLESP)
27 rue de Chaligny
75012 Paris
France
Phone: 00 33 1 78 94 98 74
Fax: 00 33 1 78 94 98 79
E-mail: vigniernicolas@yahoo.fr

***Key words:***
migration, travel, emerging infectious diseases, transient and migrants

***Competing interests:***
The authors declare that they have no competing interests.

The manuscript is in compliance with the ethical principles for medical research involving human subjects and in accordance with the Declaration of Helsinki.

## ABSTRACT

Emerging infectious diseases (EID) threaten public health and are sustained by increasing global commerce, travel and disruption of ecological systems. Travelers could play a role in importing EIDs and could be a sentinel of major epidemics. In connection with the extension of poverty, urbanization, extensive livestock rearing and globalization, we could be exposed to a third epidemiological transition characterized by zoonotic diseases and infections with multidrug-resistant bacteria. The risk appears low for emerging infectious diseases, or very low for high-risk emerging infectious diseases, but higher for multidrug-resistant enterobacteriaceae carriage with possibly limited consequences. The role played by migrants is weaker than imagined. Immigrants don't play the role of sentinel epidemic so far. They could play a role in importing multidrug-resistant enterobacteriaceae, but it is poorly evaluated.

Emerging infectious diseases (EIDs) have led to cooperation between countries, the first international epidemic response conference in 1851 and the establishment of WHO in 1948.

EIDs are diseases that have appeared recently or that have recently increased in frequency, geographical distribution or both (1). Since the end of the 20th century, there has been a constant stream of newly identified pathogens and an increasing occurrence of pandemic threats to global health (2).

These infections are due to new agents (HIV-1, Severe Acute Respiratory Syndrome CoronaVirus -SARS-CoV- (2003), avian influenza virus H5N1 (2005), H1N1 (2009)), geographical area in extension (West Nile, Dengue, Chikungunya, and Zika viruses), increased incidence of infectious disease (HIV, tuberculosis, plague), modification of virulence (*Neisseria meningitidis*) or acquisition of resistance (Extended-spectrum beta-lactamases -ESBL- or carbapenemase producing enterobacteriaceae and multidrug-resistant -MDR- tuberculosis).

We can also compare the re-emerging infections (polio virus (2014), Ebola virus (2014), etc.) (3, 4).

EIDs threaten public health and are sustained by increasing global commerce, travel and disruption of ecological systems and in particular urbanization. Urbanization is characterized by rapid intensification of agriculture, socioeconomic change, and ecological fragmentation, which can have profound impacts on the epidemiology of infectious disease (5). However, their interactions with travel and migrations are less well known.

Travelers could play a role in importing EIDs and could be a sentinel of major epidemics.

In France, there are more than 20 million travelers every year, 4.5 million of which are destined for areas at high risk for health. There are several modes of travel: tourist, business or visiting friends and relatives. Trips can be very short or extended in time.

Infectious diseases are rare health events, with the exception of common infectious diseases such as traveler's diarrhea and are a single cause of death, far behind accidents and cardiovascular disease (6).

The risk of emerging infections such as dengue in a risk zone was estimated at 1% for one month of travel (7).

We have seen (re-)emergence of diseases imported by travellers in Europe, such as chikungunya and dengue in France and Itay, and malaria in Greece (8-10). Apart from these examples, these are rare situations. However, with global travel growth, the risk could become more tangible (11).

A particular concern is that of Multidrug Resistant Enterobacteriaceae (MRE) carriage. MRE acquisition is very frequent among travellers to tropical regions (12). The acquisition was higher in Asia (72%) than in sub-Saharan Africa (48%) or Latin America (31%). However, the same study showed that MRE carriage was limited in time and disappeared after a few months.

Migration is a global phenomenon that influences the health of individuals and populations over the course of their lives (13). Migrants are special travellers who, in most case, do not migrate by choice. Migrants are considered at higher risk for a range of health problems including infectious diseases as HIV, hepatitis B, tuberculosis, schistosomiasis and malaria (14, 15). This higher risk is partly due to poor socioeconomic conditions and, in some countries, is due to the lack of rights to health coverage for undocumented migrants (16-19).

Existing evidence from different European countries highlights the difficulties to access health services that migrants are facing (20-23). These infectious diseases unequally expose the majority

population, from none at all (e.g., malaria) to a little (e.g., tuberculosis).

One can take the examples of epidemics of Middle East Respiratory Syndrome Coronavirus -MersCov- and Ebola, for which no secondary case has been reported in France.

Among the published studies on migrants and infectious diseases, the majority were non-emergent diseases with the exception of MDR tuberculosis and multidrug-resistant bacteria (24, 25).

In connection with the increased use of antibiotics in low-resource countries, there is a worrying increase in the prevalence of multidrug-resistant bacteria (26, 27). This increase could lead to an increased risk for migrants and their relatives, but there are few data on this point (28). The risk seems particularly increased when they return home to visit friends and relatives (29). While antimicrobial resistance is of concern, the prospects for pandemic spread of a bacterial or fungal emerging pathogen by migrants seem less likely (30).

Endemic disease, as tuberculosis, impose a far higher public health burden than epidemic disease (31). Denmark experienced an increase in the incidence of tuberculosis in the 1990s in relation to the increase in the number of cases among migrants (32). The rate of tuberculosis in France is 10 times higher among immigrants than in the majority population. Refugees and asylum seekers may have a heightened risk of MDR-TB infection and worse outcomes but the data remains poor (33).

Thus, there is little evidence to support the theories by which migrants would expose the host population to significant infectious risk. However, human diseases acquire a social status based on their perceived risk that determines their acceptability (31).

In a study that we conducted with a number of 347 doctors in France (infectious diseases and general practitioners), they were asked if first-time migrant people represent a vector of infectious diseases different from the majority population: 8% answered no, 13% yes but weakly, 44% yes but moderately, 27% yes significantly and 9% did not know.

Thereby, apart from infections such as tuberculosis and multidrug-resistant bacteria, the introduction of EIDs into human populations seems to be more often a consequence of economic development that brings zoonotic reservoirs in closer proximity to people.

Indeed, most pandemic threats are caused by viruses from either zoonotic sources or vector-borne sources (30). There is a need for rapid diagnosis of EIDs. Systems biology approaches can lead to a greater understanding of EIDs pathogenesis and facilitate the evaluation of newly developed vaccine-induced immunity in a timely manner (30, 34).

Close collaboration is therefore needed between specialists in tropical medicine, in public health, immunologists and biologists to anticipate the risk of EIDs in order to achieve the Sustainable Development Goals established by the United Nations in 2015 (35).

The WHO established a Department of Pandemic and Epidemic Diseases in 2011 to better prepare for and respond to EIDs.

In conclusion, in connection with the extension of poverty, urbanization, extensive livestock rearing and globalization, we could be exposed to a third epidemiological transition characterized by zoonotic diseases and infections with multidrug-resistant bacteria (36).

The risk appears low for EIDs, or very low for high-risk EIDs, but higher for MRE carriage with possibly limited consequences. The role played by migrants is weaker than imagined (except for tuberculosis). Immigrants don’t play the role of

sentinel epidemic so far. They could play a role in importing MRE, but it is poorly evaluated.

## REFERENCES

1. Metcalf CJE, Lessler J. Opportunities and challenges in modeling emerging infectious diseases. Science. 2017;357(6347):149-52.

2. Fauci AS, Morens DM. The perpetual challenge of infectious diseases. N Engl J Med. 2012;366(5):454-61.

3. Drosten C, Gunther S, Preiser W, van der Werf S, Brodt HR, Becker S, et al. Identification of a novel coronavirus in patients with severe acute respiratory syndrome. N Engl J Med. 2003;348(20):1967-76.

4. Ungchusak K, Auewarakul P, Dowell SF, Kitphati R, Auwanit W, Puthavathana P, et al. Probable person-to-person transmission of avian influenza A (H5N1). N Engl J Med. 2005;352(4):333-40.

5. Hassell JM, Begon M, Ward MJ, Fevre EM. Urbanization and Disease Emergence: Dynamics at the Wildlife-Livestock-Human Interface. Trends Ecol Evol. 2017;32(1):55-67.

6. Hargarten SW, Baker TD, Guptill K. Overseas fatalities of United States citizen travelers: an analysis of deaths related to international travel. Ann Emerg Med. 1991;20(6):622-6.

7. Steffen R, Amitirigala I, Mutsch M. Health risks among travelers--need for regular updates. J Travel Med. 2008;15(3):145-6.

8. Delisle E, Rousseau C, Broche B, Leparc-Goffart I, L'Ambert G, Cochet A, et al. Chikungunya outbreak in Montpellier, France, September to October 2014. Euro Surveill. 2015;20(17).

9. Tseroni M, Baka A, Kapizioni C, Snounou G, Tsiodras S, Charvalakou M, et al. Prevention of Malaria Resurgence in Greece through the Association of Mass Drug Administration (MDA) to Immigrants from Malaria-Endemic Regions and Standard Control Measures. PLoS Negl Trop Dis. 2015;9(11):e0004215.

10. Marchand E, Prat C, Jeannin C, Lafont E, Bergmann T, Flusin O, et al. Autochthonous case of dengue in France, October 2013. Euro Surveill. 2013;18(50):20661.

11. World Tourism Organization. UNWTO Annual Report 2016. Madrid: UNTWO; 2017.

12. Ruppe E, Armand-Lefevre L, Estellat C, Consigny PH, El Mniai A, Boussadia Y, et al. High Rate of Acquisition but Short Duration of Carriage of Multidrug-Resistant Enterobacteriaceae After Travel to the Tropics. Clin Infect Dis. 2015;61(4):593-600.

13. Zimmerman C, Kiss L, Hossain M. Migration and health: a framework for 21st century policy-making. PLoS Med. 2011;8(5):e1001034.

14. Simon J, Kiss N, Laszewska A, Mayer S. Public Health Aspects of Migrant Health: A Review of the Evidence on Health Status for Labour Migrants in the European Region. WHO Health Evidence Network Synthesis Reports. Copenhagen2015.

15. De Vito E, de Waure C, Specchia ML, Ricciardi W. Public Health Aspects of Migrant Health: A Review of the Evidence on Health Status for Undocumented Migrants in the European Region. WHO Health Evidence Network Synthesis Reports. Copenhagen2015.

16. Vazquez ML, Vargas I, Jaramillo DL, Porthe V, Lopez-Fernandez LA, Vargas H, et al. Was access to health care easy for immigrants in Spain? The perspectives of health personnel in Catalonia and Andalusia. Health policy. 2016;120(4):396-405.

17. Derose KP, Bahney BW, Lurie N, Escarce JJ. Review: immigrants and health care access, quality, and cost. Med Care Res Rev. 2009;66(4):355-408.

18. Magalhaes L, Carrasco C, Gastaldo D. Undocumented migrants in Canada: a scope literature review on health, access to services, and working conditions. J Immigr Minor Health. 2010;12(1):132-51.

19. Vignier N, Desgrees du Lou A, Pannetier J, Ravalihasy A, Gosselin A, Lert F, et al. Access to health insurance coverage among sub-Saharan African migrants living in France: Results of the ANRS-PARCOURS study. PLoS One. 2018;13(2):e0192916.

20. Rechel B, Mladovsky P, Ingleby D, Mackenbach JP, McKee M. Migration and health in an increasingly diverse Europe. Lancet. 2013;381(9873):1235-45.

21. Scheppers E, van Dongen E, Dekker J, Geertzen J, Dekker J. Potential barriers to the use of health services among ethnic minorities: a review. Fam Pract. 2006;23(3):325-48.

22. Gray BH, van Ginneken E. Health care for undocumented migrants: European approaches. Issue Brief (Commonw Fund). 2012;33:1-12.

23. Dourgnon P, Jusot F, Silva J, Sermet C. Immigrants' access to ambulatory care in France. Questions d'économie de la santé. 2009;146.

24. Napoli C, Dente MG, Karki T, Riccardo F, Rossi P, Declich S, et al. Screening for Infectious Diseases among Newly Arrived Migrants: Experiences and Practices in Non-EU Countries of the Mediterranean Basin and Black Sea. Int J Environ Res Public Health. 2015;12(12):15550-8.

25. Kentikelenis A, Karanikolos M, Williams G, Mladovsky P, King L, Pharris A, et al. How do economic crises affect

migrants' risk of infectious disease? A systematic-narrative review. Eur J Public Health. 2015;25(6):937-44.

26. Laxminarayan R, Matsoso P, Pant S, Brower C, Rottingen JA, Klugman K, et al. Access to effective antimicrobials: a worldwide challenge. Lancet. 2016;387(10014):168-75.

27. Karanika S, Karantanos T, Arvanitis M, Grigoras C, Mylonakis E. Fecal Colonization With Extended-spectrum Beta-lactamase-Producing Enterobacteriaceae and Risk Factors Among Healthy Individuals: A Systematic Review and Metaanalysis. Clin Infect Dis. 2016;63(3):310-8.

28. Angeletti S, Ceccarelli G, Vita S, Dicuonzo G, Lopalco M, Dedej E, et al. Unusual microorganisms and antimicrobial resistances in a group of Syrian migrants: Sentinel surveillance data from an asylum seekers centre in Italy. Travel Med Infect Dis. 2016;14(2):115-22.

29. Khawaja T, Kirveskari J, Johansson S, Vaisanen J, Djupsjobacka A, Nevalainen A, et al. Patients hospitalized abroad as importers of multiresistant bacteria-a cross-sectional study. Clin Microbiol Infect. 2017;23(9):673 e1- e8.

30. Graham BS, Sullivan NJ. Emerging viral diseases from a vaccinology perspective: preparing for the next pandemic. Nat Immunol. 2018;19(1):20-8.

31. Medley GF, Vassall A. When an emerging disease becomes endemic. Science. 2017;357(6347):156-8.

32. Carballo M, Nerukar A. Migration, refugees, and health risks. Emerg Infect Dis. 2001;7(3 Suppl):556-60.

33. Hargreaves S, Lonnroth K, Nellums LB, Olaru ID, Nathavitharana RR, Norredam M, et al. Multidrug-resistant tuberculosis and migration to Europe. Clin Microbiol Infect. 2017;23(3):141-6.

34. Oh SJ, Choi YK, Shin OS. Systems Biology-Based Platforms to Accelerate Research of Emerging Infectious Diseases. Yonsei Med J. 2018;59(2):176-86.

35. United Nations. Resolution adopted by the General Assembly on 25 September 2015. Transforming our world: the 2030 Agenda for Sustainable Development. 2015.

36. Zuckerman MK, Harper KN, Barrett R, Armelagos GJ. The evolution of disease: anthropological perspectives on epidemiologic transitions. Glob Health Action. 2014;7:23303.

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# Is the profession of laboratory medicine uniform across the North Mediterranean countries?

Konstantinos Makris

*Clinical Biochemistry Department, KAT General Hospital, Kifissia, Greece*

ARTICLE INFO

***Corresponding author:***
Konstantinos Makris
Clinical Biochemist
Clinical Biochemistry Department
KAT General Hospital
2 Nikis street, Kifissia, 14561
Greece
Email1: kostas.makris.km@gmail.com
Email2: kostas.makris.km@icloud.com

***Key words:***
EFLM, UEMS, training, laboratory medicine, EFLM syllabus

ABSTRACT

Harmonization of the postgraduate training of both Clinical Scientists and Physicians, in Laboratory Medicine (LM) has been a goal for many years, for the **European Federation of Clinical Chemistry and Laboratory Medicine (EFLM)** and the **Union Européenne de Médecins Spécialistes (UEMS), Section of Laboratory Medicine/ Medical Biopathology.** This was based on the concept of free movement of people within the European Union.

Much has been achieved within the respective European organizations in the development of curricula that will harmonize the postgraduate training at least within the European Union (EU).

Advances in the area of diagnostics and the need for particular expertise in distinct areas have led to the emergence of laboratory scientists and physicians specialized in hematology (including transfusion medicine), clinical biochemistry, immunology, and microbiology.

However, the training and specialization is varying and practice is of laboratory medicine is polyvalent in some countries and single specialties in others countries.

Moreover, these advances have led to the involvement of non-medical scientists in the clinical laboratories.

However, the training and the roles of Medical Doctors and Clinical Scientists in a Clinical laboratory, differ from country to country. These differences still remain today throughout Europe and even within the EU.

❖❖❖❖❖

## INTRODUCTION

The profession of clinical chemistry and laboratory medicine is practiced in all of the North Mediterranean countries. However, it differs among countries in many respects, such as background training, fields of interest, legal status and professional organization.

In Europe, there are two organizations that represent national professional organizations in the field of Laboratory Medicine and Clinical Chemistry: **European Federation of Clinical Chemistry and Laboratory Medicine (EFLM)** and the **Union Européenne de Médecins Spécialistes (UEMS), Section of Laboratory Medicine/Medical Biopathology**.

Both have as objective to promote mutual recognition of laboratory specialists within the European Union (EU). This is closely related to the free movement of people, a major goal of European integration. However, in order this policy to be accomplished in the field of Laboratory Medicine needs equivalence of standards and harmonization of the training curriculum among member states, both the central tasks of the European professional organizations.

Harmonization of the profession has been an important objective, and has a long history. In 1958 – one year after the treaty of Rome was signed – representatives of the professional organizations of medical specialists of the six-member states of the very new European Economic Community (EEC), met in Brussels and created the European Union of Medical Specialists [Union Européenne des Médecins Spécialistes (UEMS)]. The main objectives of the UEMS are: to promote the highest level of training of the medical specialists, medical practice and health care within the EU and to promote free movement of specialist doctors within the EU. The UEMS represents the medical specialist profession in the member states of the EU, to EU authorities and any other authority dealing with questions concerning the medical profession. Laboratory Medicine as a medical specialty is represented in UEMS.

In 1988 the Federation of European Societies for Clinical Chemistry (FESCC) was founded, joining the different national societies for clinical chemistry and laboratory medicine. The harmonization and recognition of laboratory specialists, particularly of scientific and pharmaceutical background, was the main objective from the start. The EC4 was subsequently founded, with the goal to harmonize the training through the production of a syllabus that will be commonly accepted by all members and establishing a European register for highly trained specialist, whatever their background.

Until the early 90's when European Community started to issue directives to the state members to adopt a system on a uniform basis of graduation and of post-graduation/specialization for all medical graduates, the training of medical doctors in LM was not only variable but in some countries was missing. To be specialized became, over a few years, a pre-requisite for practicing the medical activity in a specific professional field.

However, in most European Hospital Laboratories, are employed not only medical doctors but even pharmacy, biology and chemistry graduates (the so called scientists).

The result was that on the one side we had specialized MDs, on the other side non-specialized, non-MD graduates, creating the potential for serious issues. Each country developed its own solution. Several countries issued by law the training

of non-MD graduates in order to comply with the EU directives while other countries did not. In the latter, professional societies started initiatives of volunteer training programs based on EC4 syllabus. The result is a tremendous diversity not only concerning who is qualified to practice Laboratory Medicine, but also what is the definition of LM and what disciplines should be included in the curriculum. This diversity still exists.

The European Federation of Clinical Chemistry and Laboratory Medicine developed a syllabus for the postgraduate education and training for all Specialists in LM in order to provide a framework for training for all EU countries (now it is in latest version 5 – 2018).

A common syllabus has many useful applications. First, it describes in detail the education and training associated with high-quality, specialist practice but also can help in defining the common set of skills, knowledge and competence for non-medical Specialists in Laboratory Medicine under EU Directive 2013/55/ EU (The recognition of Professional Qualifications).

Second, it helps to give a definition to LM through the training. What disciplines should be included under the umbrella of LM? This is not a static definition and should be evolved as clinical research and technology evolve. and finally, it will provide the specialists with professional qualifications able to work without restrictions throughout EU (complying with EU Directive 2013/55/EU in providing safeguards to professional mobility across European borders).

## THE DEFINITION OF THE PROFESSION ACCORDING TO EFLM AND UEMS

IFCC defines clinical chemistry and laboratory medicine as "*the application of chemical, molecular and cellular concepts and techniques to the understanding and the evaluation of human health and disease*".

At the core of the discipline is the provision of "*results of measurements and observations, together with interpretation and informed clinical advice relevant to the maintenance of health, the cause of disease, the diagnosis of disease, predicting and monitoring the response to therapy, and follow-up investigations*". The discipline is committed to deepening the understanding of health and disease through fundamental and applied research.

High technical skills and ability to interpret results and provide consultancy to clinicians are the top requirements for LM scientists

In the latest version of EFLM syllabus the following disciplines are included in the training that could be common for medical and non-medical origin scientists: clinical chemistry, immunology, haematology and blood transfusion (including blood cells, haemostasis, cellular immunology and transfusion serology), microbiology (bacteriology, mycology, virology and parasitology), genetics, genomics and cytogenetics and finally *in vitro* fertilisation. Only anatomic pathology is outside this framework (Figure 1).

While EFLM provides with the present version a uniform framework for all scientists regardless of background, UEMS provides training only for medical doctors and recognizes different specializations for the section of Laboratory medicine.

Under the umbrella of the more recent name of the section, Laboratory Medicine/Medical Biopathology, several subspecialty divisions have been acknowledged.

At present, the section consists of the following divisions (see also Figure 2): the polyvalent General Laboratory Medicine/Polyvalent Medical Biopathology, and the monovalent specialities of Laboratory Medicine-Clinical Chemistry, Clinical and Laboratory Haematology, Clinical and Laboratory Immunology, and Laboratory Genetics (Genetic Pathology/Medical Genomics).

Since 2008 a separate section of Medical Microbiology was created within the UEMS with its own training program. In 1988 Anatomic Pathology was the first specialist section that split off from laboratory medicine section of UEMS, and was recognized as separate section with the main objective to harmonize the practice of pathology in Europe.

It is important to understand that it is not only that the two professional organization have differences when they try to define the profession of laboratory medicine.

There are huge differences between EU (and of course non-EU) countries. State regulations also allow or not to non-medical scientists to practice the profession of laboratory medicine.

Many specialties are defined differently from country to country within the European Union. For example, in the Netherlands and Scandinavia, Clinical Chemistry includes biochemistry, immunology, and hematology (most of the subspecialties included in General Laboratory Medicine/ Polyvalent Medical Biopathology but excluding Laboratory Microbiology). In other countries (Ireland, UK) LM is named chemical pathology (for the medicine derived scientists) and includes biochemistry and endocrinology or Clinical Biochemistry (for the non-medical clinical scientists.

The clinical component of practice varies among different specialties, with some having a strong clinical emphasis. For example, in the UK, chemical pathologists and immunologists may have independent responsibility for patient care and Clinical Scientists at the consultant level are allowed to provide clinical advice. This is not a common practice in other EU countries.

**Figure 1** **Disciplines of LM according to EFLM syllabus**

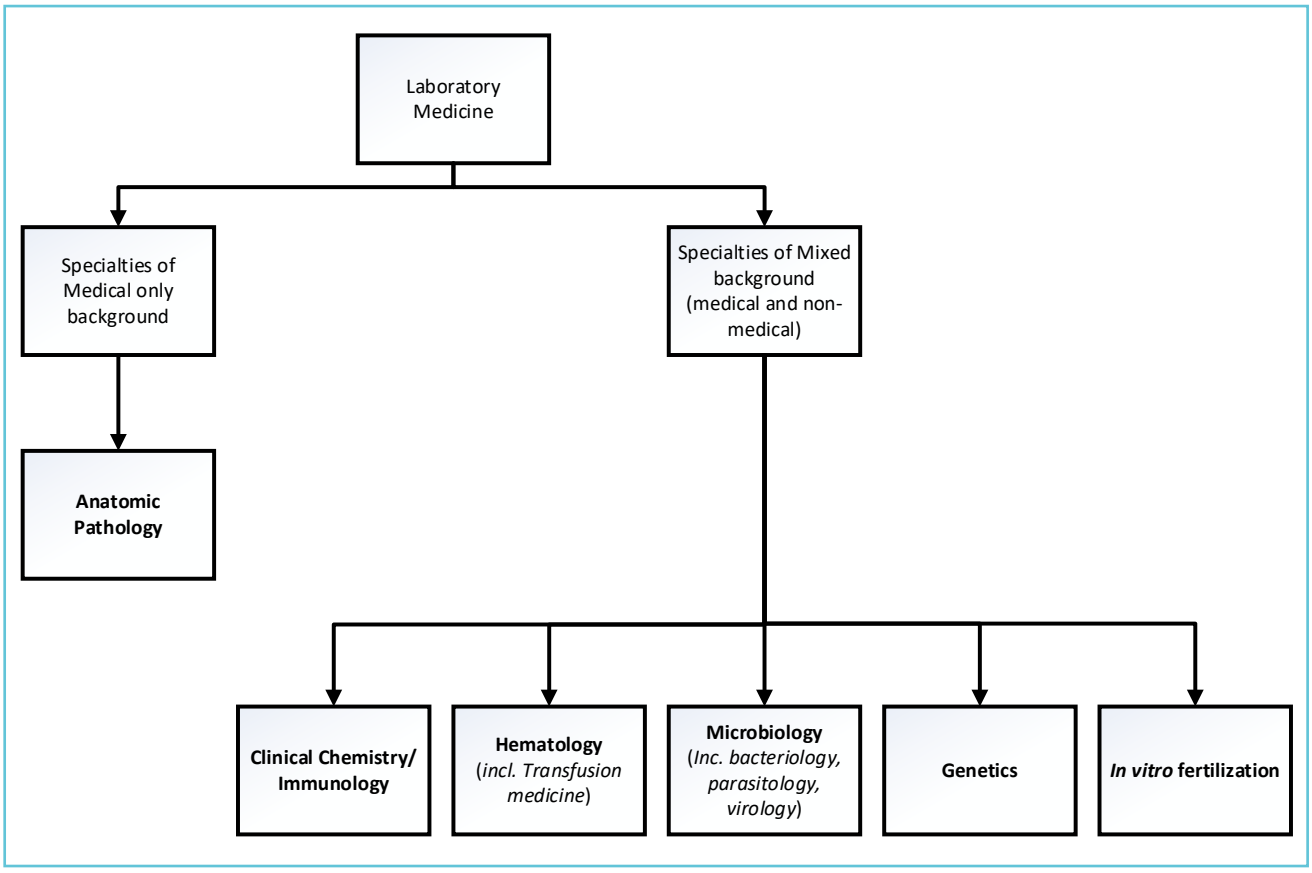
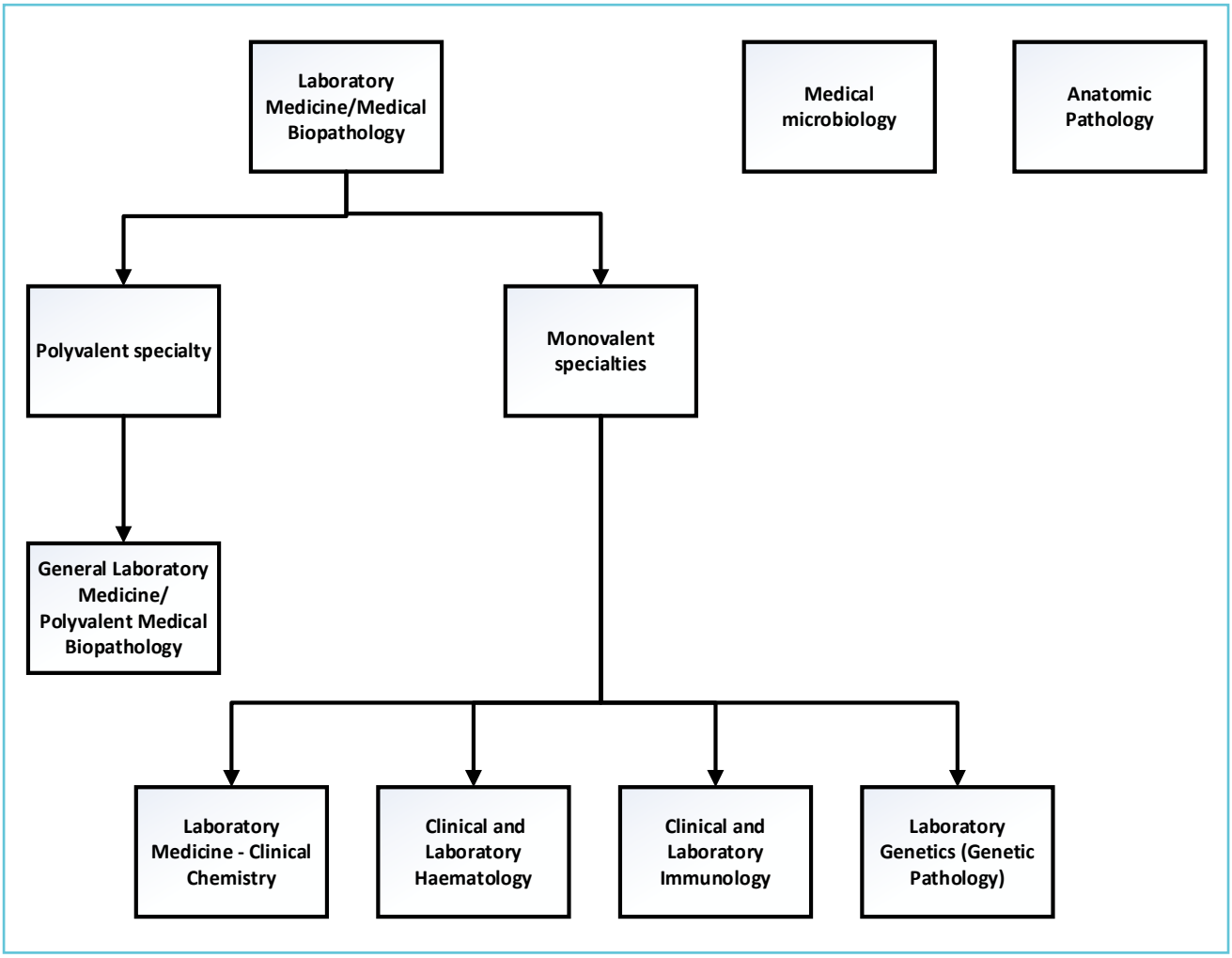


**Figure 2** Polyvalent and monovalent specialties and disciplines of LM according to UEMS syllabus




The name of the discipline also varies between countries and clearly reflects the differences that exist throughout Europe making particularly challenging the task to harmonize specialist training within the EU and Europe in general.

## WHAT IS THE SITUATION NOW IN NORTH MEDITERRANEAN COUNTRIES?

The countries that comprise the North Mediterranean region are (from west to east):

- Spain
- France
- Italy
- Slovenia
- Croatia
- Bosnia and Herzegovina
- Serbia
- Kosovo
- Montenegro
- Albania
- FYROM
- Greece
- Turkey
- Cyprus

All of the above are European countries, with seven of them being EU member states.

In the following paragraphs we try to describe the situation in all north Mediterranean countries. Source of our information was the replies to a questionnaire we sent to all north Mediterranean countries (*see* Appendix A - Questionnaire *on next page*) and the published documents we found in the internet and scientific journals.

Most of the countries responded to our questionnaire however we did not got responses from Croatia and Montenegro. And while we were able to retrieve information for Croatia from published documents we were unable to have any information from Montenegro. It is not easy to group the practices from these countries since there are huge differences. However, and most interestingly all countries claim that their training is EFLM syllabus compliant.

In Spain there are two specialties which are closer to what EFLM describes as LM are **Bioquímica Clínica (**which is the monovalent specialty**),** **and Análisis Clínicos (**the polyvalent specialty**)**. Medical Genetics, Transfusion Microbiology-Virology, Immunology, Laboratory Haematology, Medical Microbiology, and Haemostasis constitute separate autonomous monovalent specialties requiring special training. Medical doctors and non-medical scientists (of pharmacy, biology, biochemistry and chemistry background) are eligible for training in LM and are accepted after examination. Training is state regulated the duration is 4 years for everybody and licensed specialists have equal rights in releasing final results and in providing clinical consultancy to clinicians. From the remaining separate specialties only Haematology is strictly a medical specialty and genetics is not yet a medical specialty. The training program is compliant to EFLM syllabus.

In France we discovered that while MD's and Pharmacy graduates can work as LM specialists in all labs and the name of the specialty here is **Biologie médicale**, non-medical scientists can work only in University Hospital labs and when it comes to who has the rights in releasing final results this is not granted to all clinical scientists and they cannot provide any clinical consultancy. Training for medical and pharmacy scientists is state regulated is ELFM syllabus compliant and the total duration of studies is 10 years (including the undergraduate studies).

In Italy the name is **Laboratory Medicine**, the situation is closer to Spain concerning the type of university degrees that are accepted for training. Non-medical trained specialists share the same rights in respect of releasing the final results but are not allowed to provide clinical consultancy. Only medical genetics represent an autonomous specialty. The training program is state regulated and is (at least in part) EFLM syllabus compliant.

Clinical Chemistry and Laboratory Medicine (CCLM) is a scientific discipline within medicine in Slovenia, is the largest sub-discipline of laboratory medicine, and is named **Medical Biochemistry (Medicinska Biokemija)**. The practice of Medical Biochemistry in Slovenia includes clinical biochemistry (including toxicology, therapeutic drug monitoring, endocrinology, molecular diagnostics, immunology), hematology and coagulation. The training is 4 years for all, it is state regulated and open to medicine, pharmacy, chemistry, biochemistry, or other relevant university studies degrees and finally is ELFM syllabus compliant.

**Clinical chemistry** and **Medical biochemistry** is the terms that used in Bosnia and Herzegovina to describe the profession. the specialty is open to scientists from Medical schools, Pharmacy and non-medical (biologists, biochemists, chemists and other graduates from life-sciences). State regulated training is provided for both

## APPENDIX A - *Questionnaire*

*What is the term that describes best the specialty (Clinical Chemistry and Laboratory Medicine) in your country? (please check all appropriate replies)*

1. Clinical Chemistry
2. Clinical Biochemistry
3. Laboratory medicine
4. Medical Biochemistry
5. Clinical Biology
6. Chemical Pathology
7. Other (please describe)……

*What is the educational background of the scientific staff working in clinical laboratories in your country (please check all appropriate replies)*

1. Medical (MD)
2. Clinical Scientists (of non-medical, Life Sciences background)
3. Pharmacist
4. Other
5. Mixed (please describe in detail)……

*If the scientific staff is of mixed-background, can the non-medical scientist*

1. final release of results
   a. YES
   b. NO
2. provide clinical consultancy
   a. YES
   b. NO

*If the scientific staff is only of medical origin, what is the name of the medical specialty that is required from an MD in order to be eligible to work?*

1. Medical Biochemistry
2. Medical Biopathology
3. Chemical Pathology
4. Pathology
5. Clinical Biology
6. Biological Chemistry
7. Other (please specify)……

*Are the following laboratory medicine specialties constitute autonomous medical specialties requiring special and autonomous training?*

1. Medical Genetics
2. Molecular diagnostics
3. Transfusion Microbiology-Virology
4. Immunology
5. Laboratory haematology
6. Medical microbiology
7. Other (please specify)……

*Is the training common in content and duration form medical and non-medical scientists?*

1. Yes
2. No

*What is the total number of years of training required in order to be eligible to work in the clinical lab.*

A. for medical background
   1. pre-graduate
   2. post-graduate
B. for non-medical background
   3. pre-graduate
   4. post-graduate

*Is the training program:*

1. State regulated
2. Scientific Society regulated
3. Other……

*Is your training syllabus EFLM compliant?*

1. Yes
2. No

medical and non-medical background candidates. However non-medical scientists are not allowed to do clinical consultancy. Molecular diagnostics, immunology, laboratory hematology medical microbiology and transfusion medicine are separate monovalent medical specialties in this country. Finally, the training program is not EFLM syllabus compliant.

**Medical biochemistry** and **Medical biochemist** (**Specijalist Medicinske Biokemije I Laboratorijske Medicine**) are terms commonly used in Croatia, equivalent to Clinical Chemistry and Clinical Chemist, respectively, in most European countries. Medical biochemistry is almost exclusively practiced by medical biochemists. Medical biochemistry in Croatia comprises clinical biochemistry, haematology and coagulation, immunology, toxicology and therapeutic drug monitoring and endocrinology. Blood-banking, microbiology and cytogenetics are separate entities. Medical Biochemistry is studied at the Faculty of Pharmacy and Biochemistry. University degree (Master of Science) is earned after the 5 years of the studies and postgraduate training which is 1 year follows, it is state regulated and is ELFM syllabus compliant. The Faculty of Pharmacy and Biochemistry provides the postgraduate education within the three years of Doctoral studies for: (a) Pharmacy sciences; and (b) Biomedical sciences. PhD degree is requirement for a head of the clinical laboratory at the university hospital. However, PhD degree is not a requirement for medical biochemistry specialists to practice the profession in a laboratory.

In Serbia, the terms used are **Clinical Biochemistry, Laboratory Medicine**, and **Medical Biochemistry** and only Medical Doctors and Pharmacists are allowed to enter the profession. The training is 3-4 years depending on background, it is also state regulated compliant to EFLM syllabus but medical genetics transfusion medicine immunology and microbiology are separate medical specialties. And finally all specialists have the right to both release results and provide clinical consultancy. Kosovo although it is part of Serbia is a recent full member of IFCC. Regulation of the profession in kosovo is similar to that in Serbia only recently only Medical Doctors can enter the profession.

Small but significant differences we can see in FYROM where the term **Medical Biochemistry** and **Clinical Chemistry** are used. While Medical Doctors, Pharmacists and non-medical scientists are allowed to enter the profession only MD's and Pharmacists have the right to release results and provide consultancy. Moreover, training is state regulated but only for medical and pharmacists it is 4 years and it is EFLM syllabus compliant. No post-graduate training is provided for non-medical scientists.

In Albania, the name is **Clinical Biochemistry** is only open to medical doctors and the training is four years it is state regulated and compliant to EFLM and UEMS syllabus. Clinical biochemistry as a specialty here incorporates Haematology and Immunology but Microbiology is a seperate medical specialty.

In Greece and Cyprus, **Medical Biopathology** (**Iatriki Biopathologia**) is the state regulated multidisciplinary medical specialty that is given only to Medical Doctors. It is 5 years in duration and incorporates microbiology, laboratory haematology, immunology, biochemistry and transfusion medicine. However this training program is partly EFLM syllabus compliant. Medical Genetics is a new state regulated medical specialty which is going to be open to both medical and non-medical scientists, but the training process has not been decided yet. A medical specialty under the name **Clinical Chemistry** (open to both medical and non-medical scientists) is inactive and no regulatory laws are in practice to govern the training of medical and non-medical scientists. The Greek society for clinical chemistry and clinical biochemistry understanding the

gap in non-medical scientists issued a training program that is compliant to EFLM syllabus and it is available for all non-medical scientists.

In Turkey, the situation is a bit more complicated. Training is performed either at hospitals or medical faculties in the universities. Although there is a core curriculum for the training from ministry of health each hospital or medical faculty has its own training program. The graduates from the Medical, Pharmaceutical, Chemistry faculties can participate at an examination in order to start training, but only the medical graduates can be trained at the Medical faculties. Non-medical graduates can be trained only at the state hospitals. They all take exams organised by their organisations in order get the diploma. The graduates with the Medical Biochemistry Diplomas all can work at the Hospital laboratories by law. But the MoH does not allow them for working at the hospital laboratories belonged to the MoH because the MoH and the medical laboratory associations or societies except the Turkish Biochemistry Society are strongly against to the training of non-medical graduates for becoming Medical Biochemist.

## IS THE PROFESSION OF LABORATORY MEDICINE CHANGING?

The definition of Laboratory medicine is not a static one. Changes in technology and in the economic environment drive the changes in the profession. Techniques that a few years ago were available only for research now are ready for everyday clinical use. Moreover, the need for better and individualised care together with these emerging technologies has driven many laboratory tests outside the safe laboratory environment to the clinicians and in certain cases to the patients themselves. Who has the right to perform laboratory tests? This also needs to be re-defined in the next years.

### *Changes due to technology*

During the last 10-20 years, major scientific and technological advances were the cause of evolutional changes that happened in the practice of laboratory medicine:

First, the **rise of new technologies** that produce biomedical "big data" (next generation sequencing, multiparameter/multiplex flow cytometry, high-throughput proteomics and metabolomics, systems biology analysis) has caused us to rethink the best approach to diagnostics. Whereas formerly one could easily spend one's clinical and investigative career developing expertise in just a few analytes, we now have the opportunity to begin to crack the incredible redundant complexity of living organisms. Moreover, the implementation and growth of clinical testing using mass spectrometry and molecular diagnostics. Once only basic research tools, now these technologies provide same-day measurement of proteins, nucleic acids, and therapeutic drugs, improving patient care in complex medical cases.

Second, the advent of the **electronic medical record** (EMR) has added to this potential but, more importantly, has made it much more possible to carry out "cost-efficient" clinical consultation in laboratory diagnostics on specific patients across a wider geographic area. At least theoretically, one pathologist can now consult quickly on multiple patients from a remote location.

Third, **point-of-care** laboratory testing is advancing at a furious pace, resulting in both potential great benefits (imagine each clinical doctor with a handheld device) and potential dangers (the clinician might not be able to tell when the instrument is pout of control or not working at all).

Finally, **high-throughput automation**, combined with electronic identification technologies, provides a platform for reduction of laboratory test–related medical errors.

All of these changes work toward progressively greater centralisation. The risk of our profession to become "big business" in its underlying structure is evident, and this makes our second objective (the research) even more difficult.

### Changes due to increasing economic restrictions

Governments and private health-care providers try to reduce the cost of healthcare in general and laboratory were easy targets. Laboratory consolidations, outsourcing of services, and hostile takeovers of hospital laboratories by commercial companies were common occurrences in the US in the mid-1990s. These measures led to a reduction not only in the number of positions for clinical laboratory staff, but also to the closing of many medical technology schools, and downsizing of postdoctoral training programs.

These practices crossed the ocean and are now common practices in many European countries. Consolidation and, in some cases, regionalisation of laboratory services with the creation of individual laboratories serving multiple healthcare facilities is now the driving force in many countries. Healthcare in Europe used to be different than in US but is facing the dangers of an increased privatisation. The private–public competition also contributed to the increased perception of health as a commodity (the product of clinical laboratories was no exception). This is a dangerous path often ignoring the importance and the true impact of diagnostic testing in patient's health.

### Changes due to regulatory requirements

Furthermore, the regulatory requirements, quality assessment programs, compliance issues, and general administrative responsibilities of laboratory directors have significantly increased over the past decade. As a result of these clinical service demands, the academic aspects of the profession and the time to participate in research have seemingly suffered.

❖❖❖❖❖

***Acknowledgements***

I would like to thank the following colleagues from North Mediterranean countries that responded to my questionnaire and devoted some time and discussed with me the training in LM in their respective countries:

José Queralto – Spain

Philippe Gillery – France

Mario Plebani – Italy

Evgenija Homsak – Slovenia

Jozo Coric – Bosnia-Herzegovina

Danica Labudovic – FYROM

Sanja Stankovitch – Serbia

Anyla Bulo – Albania

Gramos Begolli – Kosovo

Diler Aslan – Turkey

Charis charilaou – Cyprus

Dimitris Rizos – Greece

❖❖❖❖❖

## REFERENCES

1. Jassam N, Lake J, Dabrowska M, Queralto J, Rizos D, et al. The European Federation of Clinical Chemistry and Laboratory Medicine syllabus for postgraduate education and training for Specialists in Laboratory Medicine: version 5 – 2018. CCLM in press

2. https://www.uems.eu/__data/assets/pdf_file/0018/44433/UEMS-2013.25-European-Training-Requirements-Medical-Microbiology.pdf (accessed 14/9/2018)

3. https://europathol.wordpress.com/about/ (accessed 14/9/2018)

4. https://www.uems.eu/__data/assets/pdf_file/0019/44452/UEMS-2016.15-European-Training-Requirements-Laboratory-Medicine.pdf (accessed 14/9/2018)

5. http://mahse.co.uk/wp-content/uploads/2016/03/HSST-v.2.pdf (accessed 14/9/2018)

6. https://www.healthcareers.nhs.uk/explore-roles/healthcare-science/roles-healthcare-science/life-sciences/clinical-biochemistry (accessed 14/9/2018)

7. https://www.healthcareers.nhs.uk/explore-roles/doctors/roles-doctors/pathology/chemical-pathology/training-and-development-chemical-pathology (accessed 14/9/2018)

8. https://www.rcpath.org/trainees/training/training-by-specialty/chemical-pathology.html (accessed 14/9/2018)

9. Wieringa G, Zerah S, Jansen R, Simundic AM, Queralto J, Solnica B, et al. The EC4 European syllabus for post-graduate training in clinical chemistry and laboratory medicine: version 4 – 2012. Clin Chem Lab Med 2012;50:1317–28.

10. McMurray J, Zerah S, Hallworth M, Koeller U, Blaton V, Tzatchev K, et al. The European Register of Specialists in Clinical Chemistry and Laboratory Medicine: Code of Conduct, Version 2 – 2008. Clin Chem Lab Med 2009;47:372–5.

11. McMurray J, Zerah S, Hallworth M, Schuff-Werner P, Haush- ofer A, Szekeres T, et al. The European Register of Specialists in Clinical Chemistry and Laboratory Medicine: guide to the Register, version 3 – 2010. Clin Chem Lab Med 2010;48: 999–1008

12. Misbah SA, Kokkinou V, Jeffery K, et al The role of the physician in laboratory medicine: a European perspective Journal of Clinical Pathology 2013;66:432-437.

13. Kricka L J, Polsky T G, Jason Y. Park J Y, Fortina P. The future of laboratory medicine — A 2014 perspective. Clinica Chimica Acta 438 (2015) 284–303

14. Orth M, Averina M, Chatzipanagiotou S, et al Opinion: redefining the role of the physician in laboratory medicine in the context of emerging technologies, personalised medicine and patient autonomy ('4P medicine') Journal of Clinical Pathology Published Online First: 22 December 2017. doi: 10.1136/jclinpath-2017-204734

15. Q&A - Moderators: Mitchell G. Scott and Nader Rifai, Experts: Brian Smith, Michael Oellerich, Mauro Panteghini, Fred Apple, Ken Sikaris, and Ian Young. The Changing Face of Laboratory Medicine: A More Service and Less Academically Oriented Profession? Clinical Chemistry 61:2 322–329 (2015)

16. Wytze P. Oosterhuis and Simone Zerah. Laboratory medicine in the European Union Clin Chem Lab Med 2015; 53(1): 5–14

*Meeting the needs of Mediterranean nations: training and education in laboratory medicine*

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# EFLM project "Exchange of practical knowledge and skills in Laboratory Medicine" – EFLMLabX

Evgenija Homšak[1,2]

[1] *Chair, EFLM WG-CPE*

[2] *Department for Laboratory Diagnostics, University Clinical Centre Maribor, Slovenia*

ARTICLE INFO

***Corresponding author:***
Evgenija Homšak
Department for Laboratory Diagnostics
University Clinical Centre Maribor
Ljubljanska 5, 2000 Maribor
Slovenia
Email: evgenija.homsak@ukc-mb.si

***Key words:***
EFLM LabX, exchanging practice, Laboratory Medicine, website, tool

ABSTRACT

***Background***

In many laboratories/institutions of the European Federation for Clinical Chemistry and Laboratory Medicine (EFLM) there is a need to acquire additional practical knowledge and skills in different fields of Laboratory Medicine (LM).

Until now, there were no possibilities, in official and open ways, to find the link to such additional but very important education, which may be obtained in other laboratories in the country or abroad. The aim of this EFLM project is to create and operate a network of medical laboratories willing and able to offer practical training in various fields/aspects of Laboratory Medicine.

***Methods***

We conducted the survey among EFLM members, with the aim to identify the EFLM members' needs and possibilities to offer different practical training in laboratories in the country or abroad. We created the EFLMLabX website portal within the main EFLM website for offering, searching the practices and establishing direct contacts between applicants and providers institutions.

***Results***

According to the very positive survey results (146 responses) about needs (128) and interest to offer different practices (87) on the field of LM, that we have obtained from EFLM National Societies (28) in 2015, we noted a great interest for training and exchange of practical knowledge and skills in LM amongst EFLM countries. In January 2018 we have launched EFLMLabX portal. Since now we have already 13 providers/ partners institutions, from 10 different countries, which offer 17 different practice positions. Four practices are already ongoing and two were already finished.

***Conclusions***

With this project, according to obtained higher level of knowledge/experience on different field of laboratory diagnostics of general professional population (of EuSpLM), better networks between professionals, experts, and scientists we will gain higher general quality of our profession.

❖❖❖❖❖

## INTRODUCTION

The Education for the specialists of Laboratory Medicine (LM) is very complex and according to common Syllabus concerns the knowledge on several different fields of diagnostics, methods, including practical laboratory training and skills (1,2). LM is undergoing continuing changes, due to medical information improvement, novel analytical technologies development and introduction of new tests. In many laboratories/institutions of EFLM Member Societies, to achieve complete competences of their specialist, there is a need to acquire additional practical knowledge and skills in different fields of Laboratory Medicine, due to the lack of opportunity and adequate professional experts (3). These may be obtained in other laboratories in the country or abroad. Until now, there were no possibilities, on the official and open way to find the link to such additional but very important education, especially for young trainees, but also for all other European Specialists of Laboratory Medicine (EuSpLM), who want to share the knowledge of LM on a different level. Under the umbrella of EFLM, as a main and central European professional organization, there is now an option which could help to address this problem: the project "**Exchange of practical knowledge and skills in Laboratory Medicine"** which the Working Group for Congresses & Postgraduate Education (WG-CPE) is developing.

## THE AIM OF EFLMLABX PROJECT

The aim of this EFLM project is to create and operate a network of medical laboratories willing and able to offer practical training in various fields/aspects of Laboratory Medicine; additionally, to enable the connections and direct communication between providers and potential users of practices. With this project, we would be able to establish the big nets-communication between LM professionals and contribute to the higher exchange of knowledge and experience and general quality of our profession. This project is also in a scope of our several year's attempts to get official recognition of our profession, according to the new EU Council Directive on recognition of professional qualifications and Regulation with the established free movement of professionals among EU member/European countries (4).

## EFLM SURVEY ON THE NEED AND INTEREST FOR PRACTICAL TRAINING IN LM

The first phase of the project was to conduct the survey among EFLM members with the aim to identify the EFLM member needs for practical training in laboratories in the country or abroad and set up possibility and readiness of

laboratories offering such practical training in EFLM countries. We have prepared the questionnaire, which included two part of questions that were related to needs and possibilities to offer different practical experience and knowledge in EFLM laboratories and institutions. In March 2015 the related questionnaire was disseminated to all national representatives and society presidents and to a number of laboratories in EFLM Countries. According to the very positive survey results (146 responses) about needs (128) and interest to offer different practices (87) on the field of LM, that we have obtained from EFLM National Societies (28) in 2015, we noted a great interest for training and exchange of practical knowledge and skills in LM amongst EFLM countries.

Therefore, we were stimulated for the next further step to create the website program to start and running the project.

## EFLMLABX WEBSITE-PROGRAM
(https://eflmlabx.eflm.eu/en)

A dedicated website for the EFLMLabX program was developed within the framework of the main EFLM website (Figure 1), offering the possibility to search and apply to the several practical training

**Figure 1** EFLMLabX project on EFLM website

positions, and to establish direct links and communication opportunities between both providers and users/applicants of practices (Figure 2).

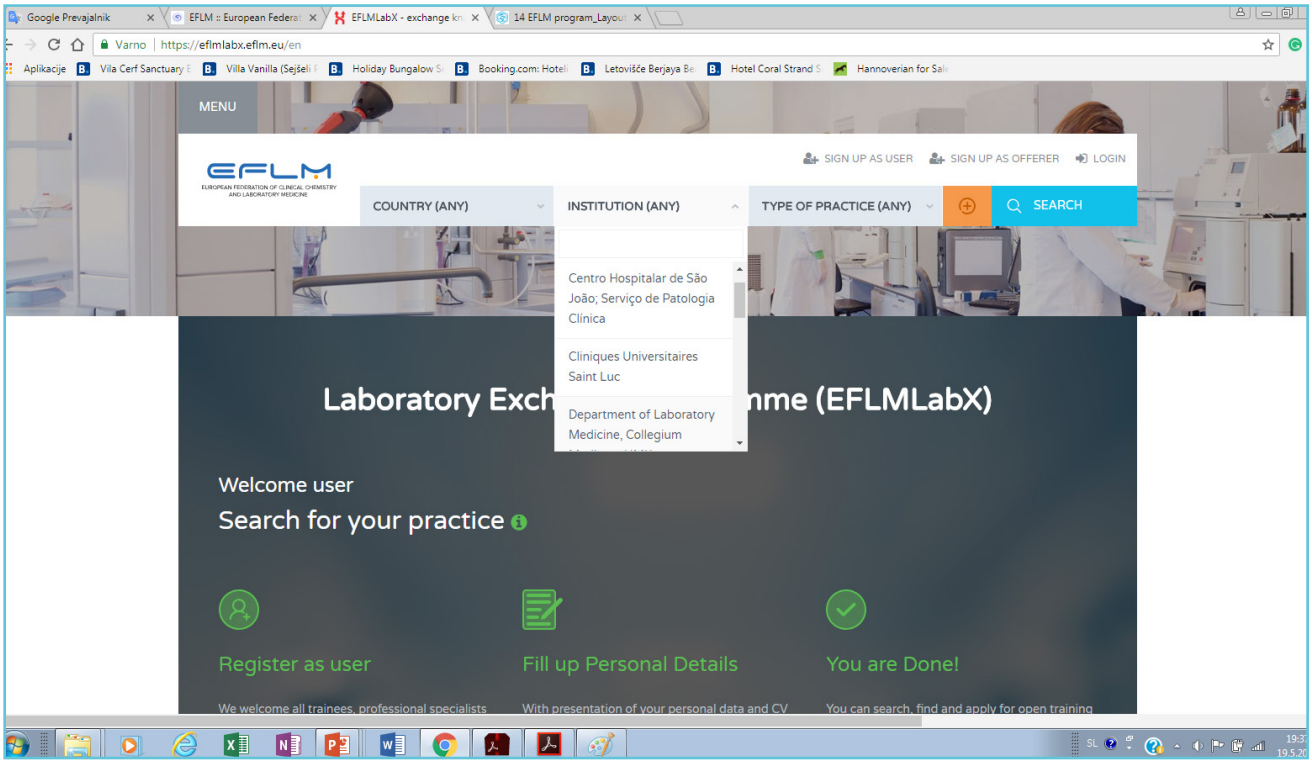
Training opportunities in the database can range from visiting, general specialist training to gaining skills necessary for specialized measurement methods or systems (introduction of new IVD systems), research methods provided by groups in laboratories and practical courses in laboratory medicine related topics. According to the added guide, here are the opportunities to sign up/ login as a user or an offerer of practices, with the generation of the own user/offerer profile. Each provider/offerer of the practice, after application and evaluation/confirmation from the EFLM WG-CPE and signed contract (Memorandum of agreement) from both (EFLM and institution) site, become a trustable partner of the project, with the obligation to manage and update all information regarding institution and offered practice positions. At the end of each concluded practice, each participant will receive Certificate of attendance. The future task of the WG-CPE will include also evaluation of these practices, with the possibility to gain CPD points for continuing professional development (5).

## GOALS OF THE PROJECT

With such a project we will be able on EFLM level:

- to achieve **higher level of experience *on different field*** of laboratory diagnostics
- to **share the knowledge and experience** among practice ***in different lab-institution***
- to learn and get/ exchange the knowledge and skills of *different* ***diagnostic methods*** (specific-GC –MS, new IVD systems...)
- to get the knowledge and skills of ***specific field of diagnostics*** (in terms of development and introduction of new diagnostics as an interest of participant/applicant's Laboratory-institution)

**Figure 2** EFLMLabX project website



- to offer the opportunity for the young trainees and specialist to expanding **contacts with (between) the experts (motivation for work and research**)
- to get the opportunity for **research work** and as part of that to get additional knowledge and skills on academic level (eg writing scientific articles)
- to find the **new potential co-workers on diagnostic fields/research science**

## FUNDATION FOR BURSARIES

To support this important exchange of knowledge and skills in LM, especially for young trainees, we would like to create the fundation for bursaries, that will be supported by IVD partners/sponsors. With bursaries would be possible to cover expences for traveling, accomodation and possible fee for education of participants. On that way we would be able to stimulate the exchange of knowledge between different professionals/ laboratories/institutions and EFLM countries.

## PILOT PROJECT AND FIRST EXPERIENCES

The EFLMLabX was launched in January 2018. The invitation to participate the project and join as a user or offerer was sent to all national representatives of EFLM. Since now we have already 13 providers/ partners institutions, from 10 different countries, which offer 17 different practice positions. Four practices are already ongoing and two were already finished.

## CONCLUSIONS

EFLMLabX project of exchanging practice is/could be a useful tool for the young and already experienced professionals (EuSpLM) in LM to obtain higher level of knowledge/experience on different field of laboratory diagnostics and to establish better networks between professionals, experts, and scientists. With this project we could gain higher general quality of our profession.

## REFERENCES

1. Wieringa G, Zerah S, Jansen R, Simundic AM, Queralto J, Solnica B, et al. The EC4 European syllabus for post-graduate training in clinical chemistry and laboratory medicine: version 4 – 2012. Clin Chem Lab Med 2012;50:1317–28

2. Jassam N, Lake J, Dabrowska M, Queralto J, Rizos D, Lichtinghagen R, et.al. The European Federation of Clinical Chemistry and Laboratory Medicine syllabus for postgraduate education and training for Specialists in Laboratory Madicine: version 5-2018. Clin Chem Lab Med 20018;doi: 10.1515/cclm-2018-0344

3. ISO 15189:2012 Medical laboratories – Requirements for quality and competence. Available from: https://www.iso.org/standard/56115.html.

4. European Parliament and EU Council. Directive 2013/55/EU of the European Parliament and of the Council of 20 November 2013 amending Directive 2005/36/EC on the recognition of professional qualifications and Regulation (EU) No 1024/2012 on administrative cooperation through the Internal Market Information System ('the IMI Regulation') 2013: Available from: http://eur-lex.europa.eu/legal-content/EN/TXT/?uri=OJ:L:2013:354:TOC.

5. Topic E, Beletic A, Zima T. Continuing professional development crediting system for specialists in laboratory medicine within 28 EFLM national societies. Biochemia Medica 2013;23(3):332-41. http://dx.doi.org/10.11613/BM.2013.041

*Meeting the needs of Mediterranean nations: improving health with emerging technologies*

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# Economic evaluation as a tool in emerging technology assessment

Nataša Bogavac-Stanojević

*Department of Medical Biochemistry, University of Belgrade-Faculty of Pharmacy, Belgrade, Serbia*

ARTICLE INFO

***Corresponding author:***
Nataša Bogavac-Stanojević
Department of Medical Biochemistry
Faculty of Pharmacy
POB 146, 11000 Belgrade
Serbia
Phone: +381 11 3951279
Fax: + 381 11 3972840
E-mail: naca@pharmacy.bg.ac.rs

***Key words:***
in vitro diagnostics, economic evaluations, point of care testing, genetic testing

***Acknowledgements:***
The work was financially supported by the Ministry of Education, Science and Technological Development, Republic of Serbia (Project No. 175035).

***Conflict of interest***
The author reports no conflict of interest, including any financial, personal or other relationships with other people or organisation.

ABSTRACT

Medical technologies are part of health technologies and they include medical devices (MD) and in vitro diagnostics (IVD). They have become a vital part of modern healthcare. Framework for introduction of new technology in the healthcare system includes a few steps: analytical and clinical accuracy assessment, clinical utility determination and economic evaluation. In addition, payers are interested whether new technology is adequate for reimbursement. There are fairly enough specific guidelines for implementation of economic methods at the early stage of IVD development. Searching the available literature in this field, this paper discusses the economic evaluations of emerging medical technologies with focus on point of care testing (POCT) and genetic testing.

Results of POCT economy studies depend on investigated perspective (payers, policy makers or society), used effectiveness values (utility, effectiveness or consequence estimated as monetary value) and understanding of clinical pathway. There is a need for better understanding of the care pathway, resource utilisation and how they change with the introduction of POCT.

Introduction of genetic testing before drug therapy was recommended with the aim to improve treatment benefit and to reduce costs of adverse drug reaction.

Clinical utility and cost-effectiveness analyses should be considered for novel genetic testing – guided treatments. Most of the studies considering genetic testing – guided treatments showed that those combinations were cost-saving or cost-effective compared to standard care.

For medical tehnology there is no universal guidance for outcomes measurement, cost calculation, performance requirements, use of a certain type of economic studies and economic thresholds.

❖❖❖❖❖

## INTRODUCTION

Medical technologies consist of both medical devices (MD) and in vitro diagnostics (IVD) and represent an important group of health technologies. It was estimated that more than 500,000 medical technologies are available today (1, 2). They have become a vital part of modern healthcare and practically no diagnosis or treatment is possible without them.

Introduction of medical technologies in the healthcare system and their reimbursement is the result of available evidence assessment with the scope of ensuring rational resources allocation (3). Payers are now requiring data about both clinical and economic value before they will consider reimbursing and using any new technology. They initially assess the clinical benefit of the technology with purpose to determine whether it is adequate for reimbursement. Payers then evaluate the added clinical benefit of the technology in comparison to existing or alternative technologies (4). This approach is obstructed by small number of available clinical studies for emerging technologies. European Union regulations consider premarket evidence, but because of the lack of appropriate data about new MD and IVD effectiveness, expectations of decision makers included in reimbursement process have rarely been met (5). Consequently, numerous applications submitted to the payers each year get rejected or withdrawn due to insufficient data (6). In addition, there is a lack of information concerning how, whether or not stakeholder should perform economic evaluations for MD and IVD or how cost-effectiveness should be applied in the health care setting (3). Number of factors, depending on medical technology, complicate economic evaluation and limit its informative value. Some of these factors are result of the fact that technologies have multiple indications or purposes and so they have distinctive features. All those require different or modified methods for economic evaluation compared to pharmaceuticals (7). Consequently, in 2016 the National Institute of Health and Care Excellence (NICE) in the United Kingdom published 50 pharmaceutical appraisals, but only 3 MD and 6 diagnostic technologies appraisals (2). The European parliament decided to reform the EU legislation for MD and IVD. In 2020, Europe's Medical Device Regulation and in 2022 In Vitro Diagnostic Regulation will come into effect, which will impact all medical technologies (8).

There are fairly enough specific guidelines for implementation of economic methods at the early stage of test development. Drawing on the available literature in this field, this paper discusses economic evaluations of emerging medical technologies with focus on point of care testing (POCT) and genetic testing.

## ECONOMIC EVALUATION OF POINT OF CARE TESTING

In 2013, St John and Price published a review paper on economic evaluation of POCT. They analysed five studies which included classical cost effectiveness analysis and two studies which applied cost consequence analysis (9). Few economic studies also analysed self-monitoring

(SM) POCT. Simon et al. applied cost – utility analyse to evaluate SM of blood glucose in patients with type 2 diabetes (10). The study showed that SM of blood glucose was more expensive than usual care. Patients had modest improvements in HbA1c levels and consequently non-significant health benefit. On the other hand in the study of Claes and colleagues, results of cost effectiveness analyses of various interventions related to INR testing by SM POCT showed that the intervention after INR levels measurement in GP surgery combined with multifaceted education was dominant over usual care. They showed increased quality (expressed as "more patients with INR values closer to the target value") and less cost (11). Contrary to the previous results, in the Parry et al. study, effectiveness of SM of INR which was expressed as "proportion of people with INR values in the therapeutic range" and costs were higher than standard care (12). Connock et al. developed a Markov model to evaluate cost-effectiveness of SM of INR in comparison to clinical care. They estimated costs from the UK NHS perspective and calculated incremental cost of SM per QALY. According to the results of Conneck study SM was cost effective using a threshold of £30,000 /QALY (13). Findings of St John and Price review study were confusing regarding economic analysis of POCT because of limited quality and availability of clinical effectiveness of POCT. Various studies used different effectiveness or utility values and compared them to costs of resource utilisation across different elements of the care pathway (9). In 2014, Ulf Martin Schilling explained main steps in calculation of direct and indirect POCT costs. He pointed out that major advantages of POCT are short turnaround times (TATs) and no requirement for dedicated laboratory staff for routine analysis (14). In addition, long TAT correlates with late diagnosis, less successful treatment and higher associated costs (cost for prolonged therapy, increased morbidity and mortality). Few studies have showed that primary testing costs are increasing while costs of complete patient pathway are decreasing and consequently adoption of POCT was cost effective (14, 15, 16). An Australian study published in 2018 examined cost effectiveness of POCT as a tool for triage of acutely ill patients in rural communities. Results showed that POCT for patients with acute chest pain, for patients with CRF who missed one or more dialysis sessions and for patients with acute diarrhea, were more expensive but more effective than Usual Care strategies. Adopting of POCT in these patients would lead to cost savings (due to unnecessary medical evaluations avoided) in rural communities (17).

Results of POCT economy studies depend on investigated perspective (payers, policy makers or society), used effectiveness values (utility, effectiveness or consequence estimated as monetary value) and understanding of clinical pathway. Most often the analysed effectiveness was from clinical studies with relatively short duration. Accordingly, main outcomes were not detected. There is a need for better understanding of the care pathway, resource utilisation and how they change with the introduction of POCT (9).

## ECONOMIC EVALUATION OF GENETIC TESTING

In the USA, cost of adverse events has been estimated at US$177 billion per year and drug's efficacy was approximate on 50%. Potential waste of money related to low drug efficacy was approximately $700 billion (18). Introduction of genetic testing before drug therapy was recommended with the aim to improve treatment benefit and to reduce costs of adverse drug reaction. For novel genetic testing – guided treatments clinical utility and cost-effectiveness should be considered. The majority of genetic testing – guided

treatments were cost-effective or even dominant (cost saving), but with notification that there was large heterogeneity in methodology between studies (19, 20).

According to systematic review conducted by Verbelen and co-workers, from 68 drugs that met inclusion criteria for study (FDA-approved drugs along with the biomarker gene - presented in The FDA Table of Pharmacogenomic Biomarkers in Drug Labeling lists), only 10 were economically evaluated. 44 economic evaluations were implemented for those 10 drugs. Over half of the 44 economic evaluations took cost utility and cost effectiveness analyses and they favoured genetic testing-guided therapy (21). Most of the studies considering genetic testing – guided treatments showed that those combinations were cost-saving or cost-effective compared to standard care (22). Rest of publications found genetic testing was not cost-effective or did not reach a definitive conclusion (21). The majority of studies evaluated genetic testing for azathioprine, clopidogrel, irinotecan and clozapine with positive economic assessment. Warfarin was evaluated in most economic studies, but they reached diverging conclusions (21).

Verbelen and co-workers concluded that cost of genetic testing is an important parameter of economic evaluations because the price of genetic tests decreased over time. In addition, genetic testing costs may depend on the method used to determine genetic variants (for example, PCR or measuring enzyme activity). If alternative drug for test-positive patients is expensive and if genetic test has a high proportion of false positive results, genetic test is not cost effective (21).

HER2, EGFR and KRAS testing are reimbursed in the UK and such approval was sponsored by pharmaceutical industry. In 2008, a French transparency committee recommended the use of Amgen's (CA, USA) Vectibix for metastatic colorectal cancer treatment for wild-type *KRAS* patients only. Similarly, Herceptin has been reimbursement since 2007. In Italy, HER2 and *KRAS* are publicly funded and available via a network of public hospital laboratories. *HER2*, *KRAS*, *EGFR* and *BCR-ABL* test reimbursement involved pharmaceutical subsidization or sponsorship (18).

## CONCLUSION

For medical tehnology there is no universal guidance for outcomes measurement, cost calculation, performance requirements, use of a certain type of economic studies and economic thresholds. There is no appropriate recommendation which medical technology should undergo formal national reimmbursement system. It is uncler how existing health technology criteria for medicines can be translated to medical tehnology reimbursment decision making. Whether it should be analysed using „real-world" observational evidence rather than experimental data? Assessment of cost-effectiveness is primarily of use to the policymaker and the purchaser, while healthcare provider needs to adopt technology in order to satisfy a recognised unmet need.

## REFERENCES

1. MedTech Europe - European IVD Market Statistics Report 2016: https://www.vdgh.de/media/file/5153.European_IVD_Market_Statistics_Report_2015.pdf. Assesed 20 June 2018.

2. Tarricone R, Torbica A, Drummond M. Challenges in the Assessment of Medical Devices: The MedtecHTA Project: Challenges in the assessment of medical devices. Health Economics 2017; 26(S1): 5-12.

3. Schnell-Inderst P, Mayer J, Lauterberg J et al. Health technology assessment of medical devices: What is different? An overview of three European projects. Z Evid Fortbild Qual Gesundhwes 2015; 109: 309-18.

4. Huot L, Decullier E, Maes-Beny K et al. Medical device assessment: scientific evidence examined by the French national agency for health – a descriptive study. BMC Public Health 2012;12:585.

5. Kramer DB, Xu S, Kesselheim AS. Regulation of medical devices in the United States and European Union. N Engl J Med 2012; 366: 848–55.

6. Chevreul K, Durand-Zaleski I, Bahrami SB et al. France: health system review. Health Syst Transit 2010; 12: 1–291, xxi–xxii

7. Drummond, M. F. Twenty years of using economic evaluations for drug reimbursement decisions. What has been achieved? Journal of Health Politics, Policy and Law, 2013. 38, 1081–1102.

8. The new Regulations on medical devices. https://ec.europa.eu/growth/sectors/medical-devices/regulatory-framework_en. Assesed 20 June 2018.

9. St John A, Price C. Economic Evidence and Point-of-Care Testing. Clin Biochem Rev 2013; 34: 61-74

10. Simon J, Gray A, Clarke P et al. Diabetes Glycaemic Education and Monitoring Trial Group. Cost effectiveness of self monitoring of blood glucose in patients with non-insulin treated type 2 diabetes: economic evaluation of data from the DiGEM trial. BMJ 2008; 336: 1177-80.

11. Claes N, Moeremans K, Frank B et al. Estimating the cost-effectiveness of quality-improving interventions in oral anticoagulation management within general practice. Value Health 2006; 9: 369-76.

12. Parry D, Fitzmaurice D, Raftery J. Anticoagulation management in primary care: a trial-based economic evaluation. Br J Haematol 2000; 111: 530-3.

13. Connock M, Stevens C, Fry-Smith A et al. Clinical effectiveness and cost-effectiveness of different models of managing long-term oral anticoagulation therapy: a systematic review and economic modelling. Health Technol Assess 2007; 11: iii-iv, ix-66.

14. Schilling UM: Time is money – the economic impact of point of care on the emergency department of a tertiary care university hospital. Point Care 2014; 13:21–3.

15. Asha SE, Chan AC, Walter E et al. Impact from point-of-care devices on emergency department patient processing times compared with central laboratory testing of blood samples: a randomized controlled trial and cost-effectiveness analysis. Emerg Med J 2014; 31:714–29.

16. Schilling UM. Chest pain at the emergency department. Hosp Healthcare Eur 2014; 182–6.

17. Spaeth BA, Kaambwa B, Shephard MD et al. Economic evaluation of point-of-care testing in the remote primary health care setting of Australia's Northern Territory. Clinicoecon Outcomes Res. 2018; 10: 269-277.

18. Miller I, Ashton-Chess J, Spolders H et al. Market access challenges in the EU for high medical value diagnostic tests. Per Med. 2011; 8: 137-148.

19. Phillips KA, Ann Sakowski J, Trosman J et al. The economic value of personalized medicine tests: what we know and what we need to know. Genet Med 2014; 16: 251–7.

20. Beaulieu M, de Denus S, Lachaine J. Systematic review of pharmacoeconomic studies of pharmacogenomic tests. Pharmacogenomics 2010; 11: 1573–90.

21. Verbelen M, Weale ME, Lewis CM. Cost-effectiveness of pharmacogenetic-guided treatment: are we there yet? Pharmacogenomics J. 2017;17:395-402.

22. Altar CA, Carhart J, Allen JD et al. Clinical utility of combinatorial pharmacogenomics-guided antidepressant therapy: evidence from three clinical studies. Mol Neuropsychiatry 2015; 1: 145–55.

*Meeting the needs of Mediterranean nations: improving health with emerging technologies*

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# Who or what is SHERLOCK?

Ann M. Gronowski

*Department of Pathology and Immunology, Washington University in St. Louis, MO, USA*

ARTICLE INFO

***Corresponding author:***
Ann M. Gronowski
Washington University School of Medicine
425 S. Euclid Ave., Campus Box 8118
St. Louis, MO 63110
USA
Phone: 314.362-0194
Fax: 314.362-1461
E-mail: gronowski@wustl.edu

***Key words:***
CRISPR, PCR, nucleic acid

ABSTRACT

***Background***

Polymerase chain reaction (PCR) is the most commonly used method for detecting nucleic acids. However, PCR requires specialized and expensive equipment, as well as specially trained personnel. Recently, new innovative diagnostic methods have been developed to detect nucleic acids using the Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) gene editing technology.

***Objective***

This manuscript reviews the newly emerging diagnostic methods that exploit the CRISPR technology.

***Results***

The programmable endonuclease properties of CRISPR have been harnessed for use in diagnostic testing.

Specific High-sensitivity Enzymatic Reporter un-LOCKing (SHERLOCK) and DNA Endonuclease Targeted CRISPR Trans Reporter (DETECTR) are diagnostic tools that can be used to detect specific RNA/DNA at low attomolar concentrations. Heating Unextracted Diagnostic Samples to Obliterate Nuclease (HUDSON), is a process of heat and chemical reduction that allows for direct detection of nucleotides in body fluids.

HUDSON and SHERLOCK can be combined to detect RNA/DNA directly from urine, saliva, serum, plasma, and whole blood with limited sample preparation or equipment with results in 1 to 2 hours. In addition, a lateral flow readout has been developed to facilitate assay detection.

***Conclusions:***

Potential uses of this emerging technology are numerous due to the analytical sensitivity and specificity, simplicity, speed, and flexibility.

❖❖❖❖❖

The ability to measure nucleic acids with highly sensitive methods is important for a number of applications such as: environmental monitoring, food safety, and detection of biological threats. Ultra-sensitive methods also have utility in clinical diagnostics for early detection of infectious diseases, testing the blood supply, and screening for cancer.

The most commonly used method for detecting nucleic acids is polymerase chain reaction (PCR). PCR is capable of routinely detecting <100 copies of nucleic acid per sample, which is in the low attomolar to zeptomolar range. PCR, however, requires specialized and expensive equipment, as well as specially trained personnel. For laboratories without this equipment or personnel, sending samples to a specialized laboratory takes time. In addition, in some cases, such as certain viral infections, the viral load is so low that even PCR is not sensitive enough.

Recently, new innovative diagnostic methods have been developed to detect nucleic acids using the CRISPR gene editing technology. In order to understand these new diagnostic methods, it is important to understand CRISPR.

CRISPR stands for "Clustered Regularly Interspaced Short Palindromic Repeats". (1) This is a naturally occurring genome editing tool that is part of the bacterial immune system used to fight against invading viruses. This system has been harnessed by scientists to modify genes in living cells.

Bacteria store genetic elements from infectious agents in genomic loci called CRISPR arrays as memories for adaptive immunity. In a very simplified explanation, this immune system works as follows. When bacteria senses the presence of viral DNA it produces a unique RNA, which matches that of the invading virus. This RNA forms a complex with a protein enzyme called CAS9 (**C**RISPR **as**sociated protein 9). CAS9 is an endonuclease (an enzyme that cuts DNA). The endonuclease is guided by the RNA to its DNA target. When the bacterial guide RNA finds its match DNA the Cas9 enzyme cuts the DNA in a specific location. Hence, Cas 9 is referred to as a "programmable endonuclease" because one can program it to cut a specific DNA by providing a unique RNA.

Over the past few years, researchers have realized that they can harness this system to cut, not just viral DNA, but any DNA at a specific location by changing the guide RNA to match the target of interest. In addition, this can be done, not just in a test tube, but in the nucleus of a living cell. This technology has enabled scientists to do incredible things such as edit the human genome to correct naturally occurring mutations.

Recently, this CRISPR technology has been harnessed for use as a diagnostic test. In 2017, a group of scientists reported the development of a technology called SHERLOCK which stands for: "Specific High-sensitivity Enzymatic Reporter unLOCKing". (2) Their goal was to develop a method to rapidly detect nucleic acids with high sensitivity, single base specificity, on a portable platform.

Instead of using the Cas9 endonuclease, SHERLOCK uses a related, but different protein, Cas13a. Cas13a binds and cleaves RNA rather than DNA substrates. After Cas13a cleaves its

target RNA, it adopts an enzymatically "active" state rather than reverting to inactive state, like Cas9. Cas13a then binds and cleaves additional RNAs regardless of homology. This is referred to as "collateral cleavage". It is this property of Cas13a that opens up the possibility of using Cas13a as a diagnostic tool.

SHERLOCK works by amplifying RNA (or DNA with a reverse transcriptase) using recombinase polymerase amplification (RPA) which is an isothermal nucleic acid amplification. Isothermal amplification does not require specialized instrumentation, as it uses a single temperature. The amplified nucleotides are combined with the Cas13a nuclease, a guide RNA that matches the nucleic acid sequence of interest, and a short nucleotide sequence that is coupled to a fluorescent reporter and a quencher. If the target sequence is present in the pool of amplified nucleotides, the non-specific RNAse activity of Cas13a becomes activated and the RNA reporter will be cleaved resulting in activation of the fluorophore. Therefore, the fluorescent signal is used as an indicator to determine whether the target sequence is present in the original pool of nucleotides. Hence the name "Specific high-sensitivity enzymatic reporter unlocking".

The authors demonstrated that SHERLOCK could distinguish between Zika virus (ZIKV) and dengue virus (DENV) in clinical isolates (serum or urine) where concentrations can be as low as 2000 copies/mL (3.2aM). They were also able to distinguish between several pathogenic bacterial strains, genotype human DNA, and identify mutations in cell-free tumor DNA. (2) One year later, in an issue of science, three papers were published that further advanced field of molecular diagnostics using CRISPR technology. (3-5) A group from Jennifer Doudna's group at the University of California Berkley reported on a detection system similar to SHERLOCK that they termed DETECTR for: "DNA Endonuclease Targeted CRISPR Trans Reporter". Their system utilizes Cas12a which cleaves double stranded DNA and activates non-specific cleavage of single stranded DNA. They use the system to detect carcinoma associated HPV from clinical specimens also using RPA with attomolar sensitivity.

In another paper in the same issue of Science, Myyhrvold *et al* reported on a new method to release and protect from degradation viral nucleic acid's from clinical specimens, thereby bypassing the need for nucleic acid extraction. (4) This method, called Hudson for: "Heating Unextracted Diagnostic Samples to Obliterate Nuclease", is a process of heat and chemical reduction that inactivates the high amount of ribonucleases found in body fluids and then lyses viral particles by disrupting the viral envelope, thereby releasing nucleic acid's into solution. The authors combined HUDSON and SHERLOCK to detect ZIKV and DENV directly from urine, saliva, serum, plasma, and whole blood with limited sample preparation or equipment with results in 1 to 2 hours.

In the third paper, Gootenberg *et al* advanced this technology even further with SHERLOCKv2. (5) The authors describe the following four advances to SHERLOCK:

1. four-channel multiplexing;
2. quantitative measurement as low as 2 aM;
3. 3.5-fold increase in signal sensitivity;
4. development of a lateral flow readout.

The flexibility of this revolutionary new technology has enormous potential. Proposed uses have included:

- Rapid detection of pneumonia pathogens (viral and bacterial) in one assay
- Monitoring viral load in HIV patients receiving therapy in resource limited areas
- Liquid biopsy to detect mutations in cell free DNA

- Rapid TB results before patient is lost to follow-up
- Rapid detection of pathogen resistance genes
- CRISPR to gene - edit mutations - SHERLOCK to determine proportion of genes successfully edited

In conclusion, the programmable endonuclease properties of CRISPR have been harnessed for use in diagnostic testing.

SHERLOCK and DETECTR are diagnostic tools that can be used to detect specific RNA/DNA.

HUDSON pairs with SHERLOCK for direct detection of nucleotides in body fluids.

Potential uses of this technology are numerous due to the analytical sensitivity and specificity, simplicity, speed, and flexibility.

## REFERENCES

1. Jinek M, Chilynksi K, Fonfara I, Hauer M, Doudna J, Charpentier E. A programmable dual-RNA-guided DNA endonuclease in adaptive bacterial immunity. Science 2012;337: 816–821.

2. Gootenberg JS, Abudayyeh OO, Lee JW, Essletzbichler P, Dy AJ, Joung J, Verdine V, Donghia N, Daringer NM, Freije CA, Myhrvold C, Bhattacharyya RP, Livny J, Regev A, Koonin EV, Hung DT, Sabeti PC, Collins JJ, Zhang F. Nucleic acid detection with CRISPR-Cas13a/C2/c2. Science 2017;356:438-42.

3. Chen JS, Ma E, Harrington LB, Da Costa M, Tian X, Palefsky JM, Doudna JA. CRISPR-Cas12a target binding unleashes indiscriminate single-stranded DNase activity. Science 2018;360:436-9.

4. Myhrvold C, Freije CA, Gootenberg JS, Abudayyeh OO, Metsky HC, Durbin AF, Kellner MJ, Tan AL, Paul LM, Parham LA, Garcia KF, Barnes KG, Chak B, Mondini A, Nogueira ML, Isern S, Michael SF, Lorenzana I, Yozwiak NL, MacInnis BL, Bosch I, Gehrke L, Zhang F, Sabeti PC. Field-deployable viral diagnostics using CRISPR-Cas 13. Science 2018;360:444-8.

5. Gootenberg JS, Abudayyeh OO, Kellner MJ, Joung J, Collins JJ, Zhang F. Multiplexed and portable nucleic acid detection platform with Cas13, Cas12a, and CSM6. Science 2018;360:439-44.

*Meeting the needs of Mediterranean nations: improving health with emerging technologies*

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# Advancement in POCT molecular testing: the multiplex PCR POCT devices for infectious diseases

Alpaslan Alp

*Department of Medical Microbiology, Faculty of Medicine, Hacettepe University, Ankara, Turkey*

ARTICLE INFO

***Corresponding author:***
Alpaslan Alp
Department of Medical Microbiology
Faculty of Medicine
Hacettepe University
Ankara
Turkey
E-mail: alp1086@gmail.com

***Key words:***
point of care test, multiplex point of care device, microfluidics, lab-on-a-chip, infectious disease

ABSTRACT

Rapid and accurate diagnostic tests are very important for the global control of infectious diseases. The point of care diagnosis has become a promising strategy in recent years. Different kind of point of care testing devices has been introduced into the market in the last decade. These devices must provide a low-cost, robust, sensitive, specific, and practical analysis in order to replace the conventional clinical laboratory diagnostic test algorithms when needed. The successful implementation of point of care diagnostics has a potential to increase the strength of infectious diseases surveillance programs. Finally, the rapid progress in point of care diagnosis can stimulate a shift from a centralized diagnostic model to a decentralized patient-centered approach.

## INTRODUCTION

It is critically important to reduce the global burden of infectious diseases and the drug resistant cases. Because of this reason, new diagnostic methods and instruments are continuously being developed through basic research. Rapid diagnosis of infections is very important for the initiation of an effective treatment.

After the discovery of polymerase chain reaction (PCR), there are many important milestones including sequencing, microchip technology, real-time PCR, and cartridge tests in the history of diagnostic molecular tests. Actually this amazing history is not more than 40 years that is a tiny space in the history of human being. The first step of technological progress is dreaming as stated in a paper published in 2002 by Dunne et al (1).

In that paper the authors introduced a mythical take on the future of the clinical microbiology laboratory based on the technological progress and describe the clinical microbiology in the year 2025. One of their most important predictions was the reversion of the trend of centralized laboratory services towards a decentralized testing approach that was basically point of care testing. Based upon the evolutionary pace of the technology, it will not be surprising to see that most of their predictions will become true until that time.

## MULTIPLEX POINT OF CARE TECHNOLOGIES

Multiplex point-of-care diagnostic technologies (MPOCTs) can test the presence of multiple infectious pathogens within a specimen (such as blood, urine, or sputum) (2). Proteins, cells, DNA, RNA, exosomes and metabolites can be detected by using MPOCT devices that contain hybridization papers, array settings, bead technology or microfluidic systems. This theoretically complex high technology design actually provides a very practical application that can be performed in places outside the routine laboratories like clinics, wards or doctor's offices. The favorable outcome for an ideal MPOCT device is to have a high sensor performance at low system complexity. The test results can be obtained within 15 minutes to several hours. The development of new molecular panel diagnostics that can provide results in 15 minutes would provide both clinical and economic benefits. Analysis of the multiplexed results provides the clinician with an opportunity to administer personalized therapies in a short time.

Potential benefits of MPOCTs for infectious diseases include improved patient health care and management, more appropriate use of antibiotics, improved ability to limit the spread of disease, health care cost savings and increased access to testing in remote or low-resource settings (3).

The set of tests on a multiplex technology is known as a test panel. Syndromic test panels are designed to test for multiple diseases associated with a similar set of symptoms, or a syndrome. These panels help the evaluation of the cause of the disease at the point of care. Respiratory panels and gastrointestinal panels are two examples of syndromic panels (4,5).

The main performance characteristics of commercially available MPOCTs (6) include:

1. panel size, or the number of pathogen targets that can be tested in one sample run;
2. time to test result;
3. throughput, or the number of patient samples that can be run simultaneously; and
4. physical size of the device.

The molecular diagnostic methods were initially expensive due to high investment costs, had long turnaround time (hours), and needed experienced user. However, recent developments in isothermal DNA amplification have made great contribution to the workflow of molecular diagnostics (7,8).

The successful migration of high sensitivity molecular diagnostics from the routine diagnostic laboratories to the field could dramatically improve the accuracy and sensitivity of MPOCTs, enhance public health reporting, and facilitate outbreak containment in difficult settings (9).

The key design features for MPOCTs in resource-limited settings included: loop-mediated isothermal amplification to eliminate the need for a thermal cycler, lyophilized reagents for long-term stability at high temperature, and relatively simple procedures for ease-of-use by operators in a field laboratory (10).

Microfluidic devices can provide a fully integrated MPOCT device for sample processing, fluid handling, and signal generation (11,12). A major goal is a low-cost diagnostic test for use in remote settings. Microfluidics-based devices use channels to transport small amounts of fluid by actuation forces. Solid phase nucleic acid extraction and isothermal enzymatic nucleic acid amplification steps are combined on microfluidic cartridges or chips that contain pre-stored, paraffin-encapsulated lyophilized reagents (13).

The use of microfluidic technologies reduces assay complexity and enables multiplex analysis and high-throughput screening (14). On-chip nucleic acid analysis is particularly promising because it miniaturizes and integrates the various assay steps, including the lysis or extraction of target cells, the purification of nucleic acids, the amplification of nucleic acids, and on-chip detection of reaction products (15). Current efforts in the development of lab-on-chip diagnostics include the identification of new biomarkers, as well as integrated microfluidic design, construction materials, and detector technologies (16).

A particular concern is the per-test cost and the need for instrumentation to drive the devices and product detection. One novel approach to assay construction is the use of layered paper to construct three-dimensional microfluidic devices that can distribute fluids vertically and horizontally and enable streams of fluid to cross one another without mixing (17).

With regard to detector technologies, a universal mobile electrochemical detector was recently described that can communicate results to distant sites using a mobile phone (13). These and similar developments will be critical for lab-on-chip diagnostics for resource-limited settings (18,19).

Effective communication of results for disease surveillance can be best accomplished if there is standardization for result recording and reporting. Ideally, multiple detection technologies might be combined in a single instrument.

The communication of results from MPOCT assays will require the abilities to digitally capture data and to communicate results to a central database. But using an electronic reader to scan point of care tests and store or transmit patient data remain as a major concern for data privacy and security.

The immediate goal of a MPOCT assay is to use the information gained from the test to impact the care of the patient. For many diseases, particularly, communicable diseases such as influenza or emerging infectious diseases, the use of MPOCT assays can provide a key element of disease surveillance (9). Linking data to specific geographical locations can provide information regarding disease emergence, disease spread, or progress toward control.

There are some studies that evaluate the effectiveness of MPOCTs compared to routine, laboratory-based detection methods in order to assess the impact on length of stay and antibiotic usage. In one of these studies, for the first time, the ward staff performed the MPOCT (20). The authors found no association between respiratory PCR MPOC testing and length of stay or most of the secondary outcomes except the antimicrobial prescribing decision. They concluded

that this was probably due to a delay in initiating MPOC testing. MPOC testing allowed time-critical antivirals to be given significantly faster, and results were available considerably faster than routine laboratory-based testing. It seems that in order to obtain the most beneficial results from MPOCTs, the tests must be performed immediately after the collection of the specimens. This action requires at least one personnel to be allocated for the application of MPOCTS. As highlighted by the authors, new technology itself is not enough, it should be incorporated into the routine algorithms in a correct manner.

## CONCLUSION

An ideal device for multiplexed point-of-care testing should offer a high sensor performance, like high sensitivity and multiplexing capability, as well as short turnaround times, at low system complexity, including low-cost fabrication and minimized user intervention. The future technology challenges will be the standardization and further miniaturization of the system components for the most effective use of this tool as a part of infectious diseases surveillance programs. It seems that the more widely use of MPOCT devices will bring diagnostic testing closer to patients and will be a driving force for a shift from a centralized model to a decentralized patient-centered approach.

## REFERENCES

1. Dunne DW, Pinckard JK, and LV Hooper. Clinical microbiology in the year 2025. J Clin Microbiol. 2002; 40: 3889-3893.

2. Dincer C, Bruch R, Kling A, et al. Urban multiplexed point-of-care testing-xPOCT. Trends in Biotechnol. 2017; 35:728-742.

3. Maffert P, Reverchon S, Nasser W, et al. New nucleic acid testing devices to diagnose infectious diseases in resource-limited settings. Eur J Clin Microbiol Infect Dis. 2017; 36:1717-1731.

4. Brendish NJ, Malachira AK, Armstrong L, et al. Routine molecular point-of-care testing for respiratory viruses in adults presenting to hospital with acute respiratory illness (ResPOC): a pragmatic, open-label, randomised controlled trial. Lancet Respir Med. 2017; 5: 401-11.

5. Duchesne L, Lacombe K. Innovative technologies for point-of-care testing of viral hepatitis in low-resource and decentralized settings. J Viral Hepat. 2018; 25:108-117.

6. United States Government Accountability Office Center for Science, Technology, and Engineering Health Care. Medical devices: Capabilities and challenges of technologies to enable rapid diagnoses of infectious diseases. August 2017.

7. Ahmad F, Hashsham SA. Miniaturized nucleic acid amplification systems for rapid and point-of-care diagnostics: A review. Analytica Chimica Acta. 2012; 733: 1-15.

8. Giuffridaa MC, Spoto G. Integration of isothermal amplification methods in microfluidic devices: Recent advances. Biosensors and Bioelectronics. 2017; 90:174-186.

9. Deshpande A, McMahon B, Daughton AR, et al. Surveillance for emerging diseases with multiplexed point-of-care diagnostics. Health Security. 2016; 14: 111-121.

10. Schreckenberger PC, McAdam AJ. Point-counterpoint: large multiplex PCR panels should be first-line tests for detection of respiratory and intestinal pathogens. J Clin Microbiol. 2015; 53:3110-3115.

11. Kozel TR, Burnham-Marusich AR. Point-of-care testing for infectious diseases: past, present, and future. J Clin Microbiol. 2017; 55:2313-2320.

12. Song Q, Gao Y, Zhu Q, et al. A nanoliter self-priming compartmentalization chip for point-of-care digital PCR analysis. Biomed Microdevices. 2015; 17: 64.

13. Mauk MG, Song J, Liu C and Bau HH. Simple approaches to minimally-instrumented, microfluidic-based point-of-care nucleic acid amplification tests. Biosensors. 2018; 8: 17.

14. Nemiroski A, Christodouleas DC, Hennek JW, et al. Universal mobile electrochemical detector designed for use in resource-limited applications. Proc Natl Acad Sci. 2014; 111:11984-11989.

15. Robinson T, Dittrich PS. Microfluidic technology for molecular diagnostics. Adv Biochem Eng Biotechnol. 2013; 133: 89-114.

16. Sackmann EK, Fulton AL, Beebe DJ. The present and future role of microfluidics in biomedical research. Nature. 2014; 507: 181-189.

17. Martinez AW, Phillips ST, Whitesides GM. Three-dimensional microfluidic devices fabricated in layered paper and tape. Proc Natl Acad Sci. 2008; 105:19606-19611.

18. Sharma S, Zapatero-Rodriguez J, Estrela P, and O'Kennedy R. Point-of-care diagnostics in low resource

settings: Present status and future role of microfluidics. Biosensors. 2015; 5: 577-601.

19. Geng Z, Zhang X, Fan Z, et al. Recent progress in optical biosensors based on smartphone platforms. Sensors. 2017; 17: 2449.

20. Andrews D, Chetty Y, Cooper BS, et al. Multiplex PCR point of care testing versus routine, laboratory-based testing in the treatment of adults with respiratory tract infections: a quasi-randomised study assessing impact on length of stay and antimicrobial use. BMC Infect Dis. 2017; 17: 671.

*Meeting the needs of Mediterranean nations: improving health with emerging technologies*

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# New solutions for the sample transport and results delivery: a digital lab

Damien Gruson[1,2]

[1] *Department of Laboratory Medicine, Cliniques Universitaires St-Luc and Université Catholique de Louvain, Brussels, Belgium*

[2] *Pôle de recherche en Endocrinologie, Diabète et Nutrition, Institut de Recherche Expérimentale et Clinique, Cliniques Universitaires St-Luc and Université Catholique de Louvain, Brussels, Belgium*

ARTICLE INFO

***Corresponding author:***
Damien Gruson
Service de Biochimie Médicale
Cliniques Universitaires Saint-Luc
Tour Rosalind Franklin
10F Avenue Emmanuel Mounier
B-1200 Brussels
Belgium
Phone: +32-(0)2-7646747
Fax: +32-(0)2-7646930
E-mail: damien.gruson@uclouvain.be

***Key words:***
big data, automation, machine learning, augmented reality, blockchain, drones, robots, user experience

ABSTRACT

The consolidation of laboratories, the evolution to integrated care network as well as an environment of consumerization are disrupting laboratory services and operations. The switch to SMART (Speed Metrics Automation Remote Technologies) digital laboratories based health ecosystems depends on several prerequisites for successes. Intelligent processes, integration of big data and real-time data management, automation, blockchain, Internet of things and enhancement of user experiences are key element of the smart digital laboratory. Safety, security and cost-effectiveness are pillars for the credibility and transferability of such smart digital laboratory environment. This transforming ecosystem will also trigger novel human – machine interfaces and we will be the gatekeepers for this new "click to brick" ecosystem.

## INTRODUCTION: TRANSPORTATION AND DELIVERY

At first sight, we could limit our discussion to "classical" pathways of laboratory transportation of samples and delivery of results to physicians.

Focusing on this perimeter, we can already observe that our laboratory environment is changing and consolidating (1,2). Laboratories and hospitals are operating as networks with some consolidated hubs for specialty testing. The complexity of these networks are increasing as well as the need to meet the accreditation requirements, leveraging the need for real-time monitoring, traceability, safety, temperature control and economy of scale (1-3). These networks and large size consolidated core-laboratories have triggered the evolution of transportation, supply chain management and current progresses are impacting the intra-laboratory / intra-hospital logistics but also the inter-laboratory / inter-hospital logistics (1,4). Emerging technologies offer clear gains for temperature control and monitoring, holistic supply chain management, inventory management, sample tracking, standardized connectivity between laboratory information systems, and for a broader control of whole healthcare facilities through the latest generations of electronic medical records (1,5). Innovations make processes more fluids and operations more cost effective, meeting the constraints coming from health care systems under stronger economical pressure.

However, broadening the vision led us to consider that our whole ecosystem is transforming. We are living in smart, inclusive and connected cities where the paradigms of mobility, motility and logistics are transforming (12). We are also citizens of an "accelerated world" where our relation to ordering and transport is influence by "uberization" of goods and services and wishes of "consumerization". Finally, we are actors of the ongoing digitalization wave and users of applications aimed to facilitate services, empowerment, tracking and monitoring. The efficiency of our transforming SMART (Speed Metrics Automation Remote Technologies) based health ecosystems will depend on several prerequisites for successes.

## THE PREREQUISITES OF SUCCESS FOR A SMART DIGITAL LABORATORY

### *The processes and logistics will be integrated, intelligent and scalable*

The effects of consumerization, competition and the switch to care pathways trigger real time reporting, integrated workflows, speed and cost-effectiveness (1,5). Solutions will need to take a greater control over the supply chain, to ensure fast and smooth processes, to guarantee quality and ISO requirements. Intelligent systems will analyze and sense demands, measure performances, monitor the status of systems and will respond in real-time to manage capacities, predict needs and avoid disruption and complains. An example of transformation could be the evolution from temperature maintenance to intelligent cold chain management.

### *The efficient use of big data will improve operations*

Our new ecosystems are driven by data and our new healthcare systems are made of opportunities to harness new forms of data to improve practices (6-9). The use of big data to capture and analyze, to streamline the operations and optimize the supply chain will add tremendous value to laboratory logistics and services. The application of algorithms and machine learning to crunch data, unlocking insights and opportunities as never imagine before. The perspectives include a data based automation to optimize the use of resources, reduce waste and facilitate lean operation, an effective real time management, a comprehensive data base inventory and supply

chain management, the analysis of data to control carbon footprints and contribute to a greener environment and the use of machine learning to build actionable and intelligent network and services (6-9). The benefits of machine learning have already been demonstrated in electronic health records, omics and mobile data and wearables. The efficient use of data and data flows is essential to face our transforming biotope and provide a dynamic response to disruption.

### *Relying on automation and blockchain to improve security and safety*

The recent reformation of the Global Data Protection Regulation bring to light for a safe and secure use and management of data, and this is clearly sensitive when considering healthcare data. Automation will continue to secure processes and reduce the rate of errors (6-9). The use of cognitive technologies will prevent mistakes and increase safety (13,14). Inventories, real-time supply chain management, transfer of results to caregivers will be stronger and more secure. Blockchain represent a way to improve the security and safety of data monitoring, sample transfer and results communication (16,17). Blockchain technologies have already demonstrated their benefits in the also sensitive field of economics and cryptocurrencies. Blockchain technologies are offering a decentralized network maintained by users and allowing the development of a stable and secure data set with which users can interact through transactions of various types. The opportunities offered by blockchain technologies to the clinical laboratory and biobanking fields seem therefore evident.

### *The use of tags and the internet of things to enhance operations*

Intelligent, integrated and reactive systems require the capture of data. The applications of RFID and internet of things (IoT) allow this capture of data. Perspectives include connected and autonomous transportation solutions, accurate tracking and real time management of the supply chain and services. The science of cloud computing will be the glue to collect, transfer and agglomerate the data generated by all the smart sensors and IoT.

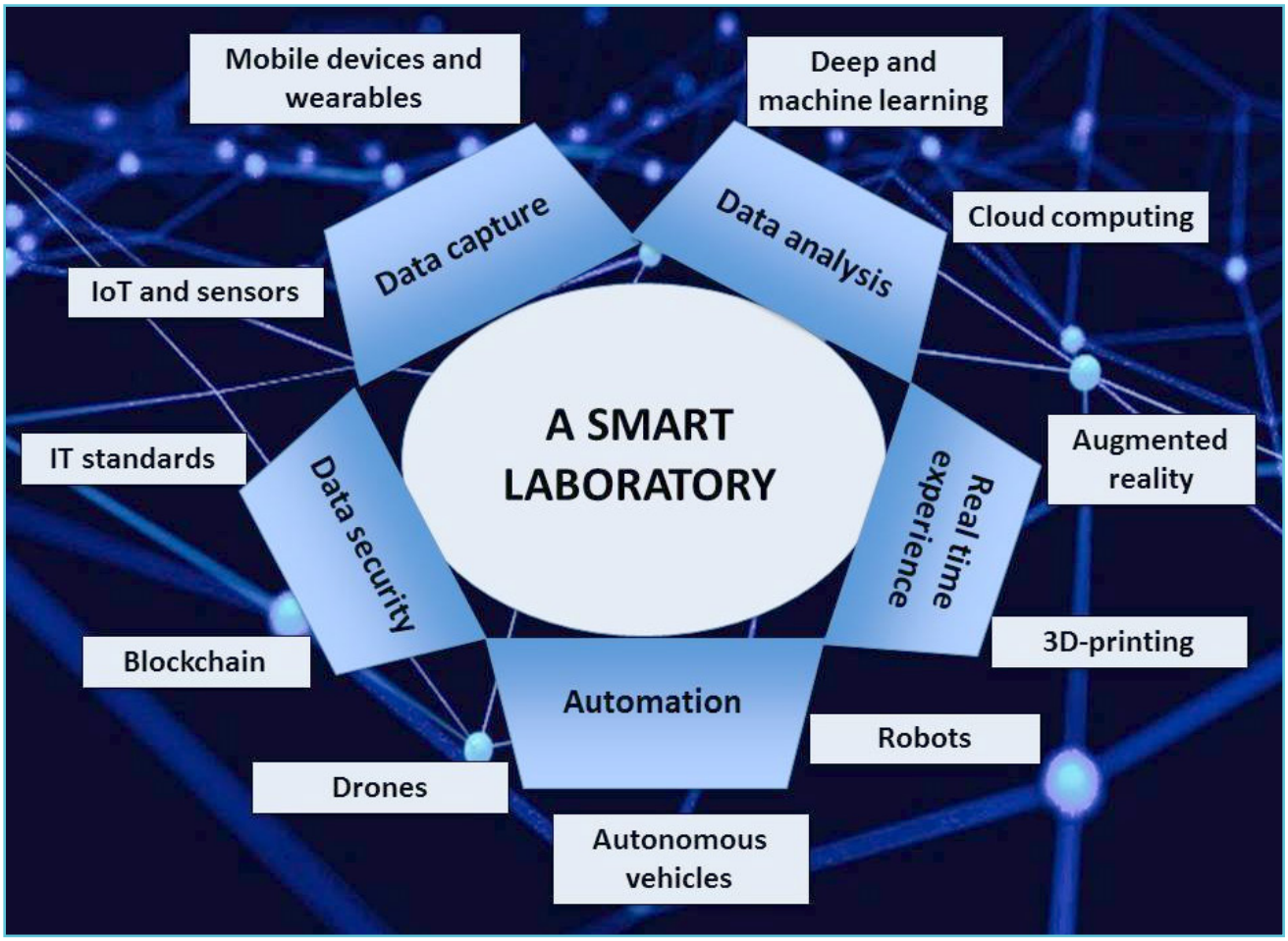
### *Emerging technologies to unlock new possibilities*

Emerging technologies are helping us to transform the way (10,11). Even if the validation and harmonization of pneumatic tube systems remains a subject of discussion, this solution remains the preferred choice for hospital. Innovative pneumatic systems are carrier free, faster and could offer more options for mega-laboratories. The use of robots and drones are also tested in hospital and laboratory networks for human samples transportation and transfer. Like cars, these new transportation solutions are becoming more and more autonomous. Transportation is also a field where sharing vehicles and means is current practices and where public – private partnerships for a greener and cost-efficient ecosystem could stimulate.

### *Co-creation and user experience*

Meeting the satisfaction and priorities of users is key and the evolution of the user experience is fundamental (15). Meeting the user's expectations means more personalized, faster and timeless services. The transcription of direct field observations to user-centered experiences is also an opportunity to improve patient safety. Our digital environment and hyper-connectivity reinforce the quest for more user centric experiences. Therefore the use of agile and users experiences methodologies will offer the possibility to re-design and co-create with the users themselves more users-centric solutions. The progresses of augmented reality will provide more option to sense the future experience and discover the benefits of future solutions, products

**Figure 1** The prerequisites for a smart laboratory (IoT, internet of things)




or services. Augmented reality, defined as the expansion of physical reality by adding layers of computer generated information to the real environment, allow now to have the information at the right time at the right place and offer mixed reality simulations for exploring the virtual and the real.

## CONCLUSIONS

The transforming ecosystem, the era of digitalization and the needs to improve efficiency and users experiences are disrupting laboratory services and operations. Such major ongoing changes allow us to implement emerging technologies into practices, develop integrated and intelligent solutions, streamline and standardize structures and processes, and provide more personalized and real-time experiences. The transformation will also offer us the digitalization and integration of vertical and horizontal value chains through cloud computing, mobile devices and IoT platforms. Safety, security and cost-effectiveness are pillars for the credibility and transferability of such smart digital laboratory environment. The future will also trigger novel human – machine interfaces and we will be the gatekeepers for this new “click to brick” ecosystem.

## REFERENCES

1. Jonas Vollanda Andreas Fügenerb Jan Schoenfeldera Jens O.Brunnera. Material logistics in hospitals: A literature review. Omega Volume 69, June 2017, Pages 82-101 https://doi.org/10.1016/j.omega.2016.08.004

2. Nybo M, Lund ME, Titlestad K, Maegaard CU. Blood Sample Transportation by Pneumatic Transportation Systems: A Systematic Literature Review. Clin Chem. 2018 May;64(5):782-790. doi: 10.1373/clinchem.2017.280479.

3. Lippi G, Banfi G, Church S, Cornes M, De Carli G, Grankvist K, Kristensen GB, Ibarz M, Panteghini M, Plebani M, Nybo M, Smellie S, Zaninotto M, Simundic AM; European Federation for Clinical Chemistry and Laboratory Medicine Working Group for Preanalytical Phase. Preanalytical quality improvement. In pursuit of harmony, on behalf of European Federation for Clinical Chemistry and Laboratory Medicine (EFLM) Working group for Preanalytical Phase (WG-PRE). Clin Chem Lab Med. 2015 Feb;53(3):357-70. doi: 10.1515/cclm-2014-1051.

4. Lippi G, Lima-Oliveira G, Nazer SC, Moreira ML, Souza RF, Salvagno GL, Montagnana M, Scartezini M, Picheth G, Guidi GC. Suitability of a transport box for blood sample shipment over a long period. Clin Biochem. 2011 Aug;44(12):1028-9. doi: 10.1016/j.clinbiochem.2011.05.028.

5. Zini A. Robots expand delivery options with seamless integration. A growing number of forward-thinking hospitals in search of greater efficiencies are embracing automated delivery robots. Health Manag Technol. 2011 Mar;32(3):10-2..

6. Matías JM, Taboada J, Ordóñez C, Nieto PG. Machine learning techniques applied to the determination of road suitability for the transportation of dangerous substances. J Hazard Mater. 2007 Aug 17;147(1-2):60-6.

7. Pashazadeh A, Jafari Navimipour N. Big data handling mechanisms in the healthcare applications: A comprehensive and systematic literature review. J Biomed Inform. 2018 Apr 12. pii: S1532-0464(18)30056-X. doi: 10.1016/j.jbi.2018.03.014.

8. Gibbons C, Greaves F. Lending a hand: could machine learning help hospital staff make better use of patient feedback? BMJ Qual Saf. 2018 Feb;27(2):93-95. doi: 10.1136/bmjqs-2017-007151.

9. Beam AL, Kohane IS. Big Data and Machine Learning in Health Care. JAMA. 2018 Apr 3;319(13):1317-1318. doi: 10.1001/jama.2017.18391..

10. Piva E, Tosato F, Plebani M. Pre-analytical phase: The automated ProTube device supports quality assurance in the phlebotomy process. Clin Chim Acta. 2015 Dec 7;451(Pt B):287-91. doi: 10.1016/j.cca.2015.10.010.

11. Lippi G, Plebani M. Laboratory applications for smartphones: risk or opportunity? Clin Biochem. 2011 Mar;44(4):273-4. doi: 10.1016/j.clinbiochem.2010.12.016.

12. Alvear O, Calafate CT, Cano JC, Manzoni P. Crowdsensing in Smart Cities: Overview, Platforms, and Environment Sensing Issues. Sensors (Basel). 2018 Feb 4;18(2). pii: E460. doi: 10.3390/s18020460. Review.

13. Cerruela García G, Luque Ruiz I, Gómez-Nieto MÁ. State of the Art, Trends and Future of Bluetooth Low Energy, Near Field Communication and Visible Light Communication in the Development of Smart Cities. Sensors (Basel). 2016 Nov 23;16(11). pii: E1968.

14. Cresswell KM, Mozaffar H, Lee L, Williams R, Sheikh A. Safety risks associated with the lack of integration and interfacing of hospital health information technologies: a qualitative study of hospital electronic prescribing systems in England. BMJ Qual Saf. 2017 Jul;26(7):530-541. doi: 10.1136/bmjqs-2015-004925.

15. Allyse MA, Robinson DH, Ferber MJ, Sharp RR. Direct-to-Consumer Testing 2.0: Emerging Models of Direct-to-Consumer Genetic Testing. Mayo Clin Proc. 2018 Jan;93(1):113-120. doi: 10.1016/j.mayocp.2017.11.001.

16. Yli-Huumo J, Ko D, Choi S, Park S, Smolander K. Where Is Current Research on Blockchain Technology?-A Systematic Review. PLoS One. 2016 Oct 3;11(10):e0163477. doi: 10.1371/journal.pone.0163477. eCollection 2016.

17. Roman-Belmonte JM, De la Corte-Rodriguez H, Rodriguez-Merchan EC. How blockchain technology can change medicine. Postgrad Med. 2018 May 10:1-8. doi: 10.1080/00325481.2018.1472996.

*Meeting the needs of Mediterranean nations: improving health with emerging technologies*

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# Next generation sequencing: from research area to clinical practice

Chiara Di Resta[1,2], Maurizio Ferrari[1,2,3]

[1] *Vita-Salute San Raffaele University, Milan, Italy*

[2] *Genomic Unit for the Diagnosis of Human Disorders, Division of Genetics and Cell Biology, IRCCS San Raffaele Hospital, Milan, Italy*

[3] *Laboratory of Clinical Molecular Biology and Cytogenetics, IRCCS San Raffaele Hospital, Milan, Italy*

## ARTICLE INFO

***Corresponding author:***
Maurizio Ferrari
Genomic Unit for the Diagnosis
of Human Pathologies
Division of Genetics and Cellular Biology
IRCCS San Raffaele Hospital
Via Olgettina 60, 20132 Milan
Italy
Phone: 02-26432303
Fax: 02-26434351
E-mail: ferrari.maurizio@hsr.it

***Key words:***
next generation sequencing, clinical laboratory medicine, medical care, high-throughput approach

***Disclosures:***
The authors declare no conflict of interest.

## ABSTRACT

Translating the power of high-throughput sequencing technologies from research area into clinical medicine is one of the major goal for several researchers and health-care providers. One of the important advantages of these technologies is that they can be successfully used in a numerous range of clinical applications. The efficiency of sequencing, that can now be achieved, is leading impressive progress in the diagnostics of common and rare genetic disorders, inherited forms of cancer, prenatal testing or infectious diseases, to cite some examples. Despite several challenges and limitations still remain to overcome, the high-throughput sequencing technologies are leading to real and unprecedented benefits for the medical care of patients.

## GENERAL OVERVIEW

Over the past decade great advances have been done in sequencing technologies. After Sanger Sequencing, the current gold standard approach, also known as dideoxy method [1], high-throughput sequencing has been developed and widespread in biomedical laboratories. The first one allows to analyse one DNA segment at time in laborious and time-consuming way while the second approach has the great advantage of performing a simultaneous analysis of several genomic regions, with a dramatic reduction also of the cost of sequencing per base [2].

Today the high-throughput next generation sequencing (NGS) instruments mainly used in biomedical laboratories are the Ion Torrent sequencers (Life Technologies) and the Illumina platforms (Illumina) [3,4].

All NGS technologies are based on the same general process, comprising template preparation, sequencing and data analysis. The unique combination of specific technical details distinguishes one technology from another and determines the type of data produced from each platform [5].

After the extraction of DNA, the first step of the sequencing process is the library preparation, which consists on the ligation of DNA fragments to platform specific oligonucleotide adapters [2]. After that each fragment is immobilized and clonally amplified. In Life Technologies approach, clonal amplification is performed by emulsion PCR, in which DNA fragments are amplified on the beads surface in oil-aqueous mixture [6–8]. Illumina approach otherwise is based on a unique "isothermal bridge amplification" reaction that occurs on the surface of the flow cell [9].

For sequencing Life Technologies exploited the native dNTP chemistry during base incorporation by DNA polymerase, that relies hydrogen ions, causing the pH modification that is detected by a modified silicon chip [10]. Illumina sequencing is instead performed on a flow-cell and it is based on the existing Solexa *sequencing by synthesis* chemistry, based on the fluorescent detection released when the complementary fluorescently tagged nucleotides are incorporated [11].

In recent years, the advent of these benchtop NGS platforms on the marketplace has had an impressive impact in *-omics*, thanks to the huge amount of data obtained with a significant reduction of time and costs [12]. Indeed NGS has been applied in varied contexts, restricted not only to genomics but also to transcriptomics and epigenomics, such as in non-coding RNA expression profiling, finding transcription factor binding sites, RNA seq, ChIp-Seq or MeDIP, to cite few examples [13].

In research genetic studies NGS has been successfully exploited to identify new causative genes or variants associated to inherited diseases, especially in genetically heterogenous disorders, whose genetic basis was partially unknown and in which gene-by-gene Sanger sequencing approach would not have been economical or efficient [2]. For this purpose, NGS has been applied to whole-genome, exome or targeted sequencing, leading to the improvement of the current knowledge of genetic basis of several pathologies, such as retinitis pigmentosa, cardiomyopathies or inherited cancer [14–17].

More recently the widespread use of these rapid high-throughput technologies, the improvement of their performance and the overcoming of initial technical limitations are encouraging their transition from basic research into clinics with important benefits for routine patient management.

## USE OF NGS IN THE CLINICAL PRACTICE

Now NGS is an established test method in many clinical laboratories, in particular for the detection of germline and somatic genetic mutations.

The analysis of causative mutations in inherited diseases is performed using different approaches, exploiting targeted panel, whole exome, whole genome or mitochondrial DNA sequencing [3,18,19]. More in details, targeted panel analysis is usually applied to genetic test for different genetically heterogeneous disorders, such as renal, neurologic, connective tissue disorders, cardiomyopathies, immune deficiencies, blindness, deafness, and several forms of inherited cancer [15,18,20–23].

Even if the analyzed gene panel may vary between laboratories, target sequencing is the first approach of genetic test for inherited disorders, while whole exome sequencing is exploited for negative cases, in which targeted testing has not been informative. Moreover, whole exome approach is useful in rare diseases for trio testing, sequencing the child and both parents [24–26].

In oncology, targeted testing is widely used, exploiting two different approaches. In the first one the targeted panel may be focused only on principle genes associated to a particular type of malignancy, for example *BRCA1* and *BRCA2* gene for breast and ovarian cancer, while in the other one NGS approach allows to analyze a broader panel including genes associated with other cancers. Given the clinical overlapping between different forms of cancer, for example between ovarian cancer and Lynch syndrome, this latter approach may be useful to enhance the diagnostic yield [27–29]. In oncology, whole exome and whole genome sequencing are not currently used for clinical purpose, in order to avoid the potential risk of unactionable incidental findings [19,29].

More recently, several new NGS applications moved to the research area to clinical use, citing for example the analysis of cell-free DNA in the prenatal genetic-testing [30], circulating tumor DNA testing [31,32], human leukocyte antigen (HLA) typing [33], microbial analysis [19], RNA sequencing and expression [19], and methylation[19], even if there are yet some challenges to overcome.

For example in HLA typing, it is difficult to differentiate low-frequency alleles from high-frequency artifacts and newer data analysis approach or the development of instruments for single-molecule sequencing, called third-generation sequencers, are solving these limitations [33,34].

Today testing of circulating tumor DNA (ctDNA), often referred to as “liquid biopsy”, is now available in clinics [35,36]. One possible approach is NGS, that presents a lot of potential applications, including diagnosis of cancer, monitoring for progression or relapse, and targeted therapy for a patient with a known cancer diagnosis [32,37,38]. Indeed, several studies have shown that ctDNA sequencing allows at first to detect somatic mutation in patients with known cancer diagnosis and then to monitor it in correlation with the relapse and progression of disease [39]. Without doubt the detection of ctDNA using NGS presents the great advantage to be a reasonable alternative to the repeated invasive biopsies for patients with metastatic cancers. However, it still presents some limitations due to a low sensitivity to detect early-stage cancer (false negatives), limiting until now the practical use of ctDNA for early cancer diagnosis or screening [38].

Other clinical applications of NGS include pharmacogenetics and microbial sequencing but these topics are beyond the scope of this article.

Although NGS is now widely used in clinics, several challenges still remain to overcome.

The main issues are, for example, the sequencing of genomic portions that are difficult to analyze, due their intrinsic features (pseudogenes, homologous regions, repetitive regions, GC-rich regions) and the limited ability to detect structural gene variation and copy number variation

(CNV) [40–43]. Sometimes the storage and the interpretation of huge amounts of sequence data, mainly of several novel or rare mutations, by trained health care professionals may be still an open challenge [44,45]. Moreover, a successful NGS testing need a collaborative effort between geneticists and physicians to combine and integrate clinical data and genetic analyses to guide medical care of patient.

## CONCLUSIONS

NGS technologies have revolutionized biological research and have deeply transform the fields of diagnostic pathology and clinical medicine. In the future, the use of NGS in clinical laboratories will surely increase as technology and bioinformatics, in order to address the current limitations, improve the quality of results, and increase the number of possible clinical applications.

However, the challenge for clinical laboratories will be to perform the most appropriate approach of NGS testing taking into account the clinical relevance, cost-effectiveness and clinical care of patient.

## REFERENCES

1. Sanger F, Nicklen S, Coulson AR. DNA sequencing with chain-terminating inhibitors. Proc Natl Acad Sci U S A. 1977;74: 5463–7. Available: http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=431765&tool=pmcentrez&rendertype=abstract

2. Jamuar SS, Tan E-C. Clinical application of next-generation sequencing for Mendelian diseases. Hum Genomics. 2015;9: 10. doi:10.1186/s40246-015-0031-5

3. Di Resta C, Galbiati S, Carrera P, Ferrari M. Next-generation sequencing approach for the diagnosis of human diseases: open challenges and new opportunities. EJIFCC. 2018;29: 4–14. Available: http://www.ncbi.nlm.nih.gov/pubmed/29765282

4. Mardis ER. Next-generation sequencing platforms. Annu Rev Anal Chem (Palo Alto Calif). 2013;6: 287–303. doi:10.1146/annurev-anchem-062012-092628

5. Reuter JA, Spacek D V, Snyder MP. High-throughput sequencing technologies. Mol Cell. 2015;58: 586–97. doi:10.1016/j.molcel.2015.05.004

6. McKernan KJ, Peckham HE, Costa GL, McLaughlin SF, Fu Y, Tsung EF, et al. Sequence and structural variation in a human genome uncovered by short-read, massively parallel ligation sequencing using two-base encoding. Genome Res. 2009;19: 1527–41. doi:10.1101/gr.091868.109

7. Wheeler DA, Srinivasan M, Egholm M, Shen Y, Chen L, McGuire A, et al. The complete genome of an individual by massively parallel DNA sequencing. Nature. 2008;452: 872–6. doi:10.1038/nature06884

8. Margulies M, Egholm M, Altman WE, Attiya S, Bader JS, Bemben LA, et al. Genome sequencing in microfabricated high-density picolitre reactors. Nature. 2005;437: 376–80. doi:10.1038/nature03959

9. Bentley DR, Balasubramanian S, Swerdlow HP, Smith GP, Milton J, Brown CG, et al. Accurate whole human genome sequencing using reversible terminator chemistry. Nature. 2008;456: 53–9. doi:10.1038/nature07517

10. Merriman B, Rothberg JM. Progress in ion torrent semiconductor chip based sequencing. Electrophoresis. 2012;33: 3397–417. doi:10.1002/elps.201200424

11. Quail M, Smith ME, Coupland P, Otto TD, Harris SR, Connor TR, et al. A tale of three next generation sequencing platforms: comparison of Ion torrent, pacific biosciences and illumina MiSeq sequencers. BMC Genomics. BioMed Central; 2012;13: 341. doi:10.1186/1471-2164-13-341

12. Williams ES, Hegde M. Implementing genomic medicine in pathology. Adv Anat Pathol. 2013;20: 238–44. doi:10.1097/PAP.0b013e3182977199

13. Choi BY, Kim BJ. Application of next generation sequencing upon the molecular genetic diagnosis of deafness. Korean J Audiol. Korean Audiological Society; 2012;16: 1–5. doi:10.7874/kja.2012.16.1.1

14. Carrera P, Di Resta C, Volonteri C, Castiglioni E, Bonfiglio S, Lazarevic D, et al. Exome sequencing and pathway analysis for identification of genetic variability relevant for bronchopulmonary dysplasia (BPD) in preterm newborns: A pilot study. Clin Chim Acta. 2015;451: 39–45. doi:10.1016/j.cca.2015.01.001

15. Di Resta C, Spiga I, Presi S, Merella S, Pipitone GB, Manitto MP, et al. Integration of multigene panels for the diagnosis of hereditary retinal disorders using Next Generation Sequencing and bioinformatics approaches. EJIFCC. 2018;29: 15–25. Available: http://www.ncbi.nlm.nih.gov/pubmed/29765283

16. Doyle MA, Li J, Doig K, Fellowes A, Wong SQ. Studying Cancer Genomics Through Next-Generation DNA Sequencing and Bioinformatics. Methods in molecular biology (Clifton, NJ). 2014. pp. 83–98. doi:10.1007/978-1-4939-0847-9_6

17. Di Resta C, Pietrelli A, Sala S, Della Bella P, De Bellis G, Ferrari M, et al. High-throughput genetic characterization

of a cohort of Brugada syndrome patients. Hum Mol Genet. 2015;24: 5828–5835. doi:10.1093/hmg/ddv302

18. Strom SP. Current practices and guidelines for clinical next-generation sequencing oncology testing. Cancer Biol Med. Chinese Anti-Cancer Association; 2016;13: 3–11. doi:10.28092/j.issn.2095-3941.2016.0004

19. Yohe S, Thyagarajan B. Review of Clinical Next-Generation Sequencing. Arch Pathol Lab Med. 2017;141: 1544–1557. doi:10.5858/arpa.2016-0501-RA

20. Kamps R, Brandão RD, Bosch BJ van den, Paulussen ADC, Xanthoulea S, Blok MJ, et al. Next-Generation Sequencing in Oncology: Genetic Diagnosis, Risk Prediction and Cancer Classification. Int J Mol Sci. Multidisciplinary Digital Publishing Institute (MDPI); 2017;18. doi:10.3390/ijms18020308

21. Celestino-Soper PBS, Gao H, Lynnes TC, Lin H, Liu Y, Spoonamore KG, et al. Validation and Utilization of a Clinical Next-Generation Sequencing Panel for Selected Cardiovascular Disorders. Front Cardiovasc Med. 2017;4: 11. doi:10.3389/fcvm.2017.00011

22. Weerakkody RA, Vandrovcova J, Kanonidou C, Mueller M, Gampawar P, Ibrahim Y, et al. Targeted next-generation sequencing makes new molecular diagnoses and expands genotype–phenotype relationship in Ehlers–Danlos syndrome. Genet Med. 2016;18: 1119–1127. doi:10.1038/gim.2016.14

23. Nijman IJ, van Montfrans JM, Hoogstraat M, Boes ML, van de Corput L, Renner ED, et al. Targeted next-generation sequencing: A novel diagnostic tool for primary immunodeficiencies. J Allergy Clin Immunol. 2014;133: 529–534.e1. doi:10.1016/j.jaci.2013.08.032

24. Fernandez-Marmiesse A, Gouveia S, Couce ML. NGS Technologies as a Turning Point in Rare Disease Research , Diagnosis and Treatment. Curr Med Chem. 2018;25: 404–432. doi:10.2174/0929867324666170718101946

25. Yang Y, Muzny DM, Reid JG, Bainbridge MN, Willis A, Ward PA, et al. Clinical Whole-Exome Sequencing for the Diagnosis of Mendelian Disorders. N Engl J Med. 2013;369: 1502–1511. doi:10.1056/NEJMoa1306555

26. Sawyer SL, Hartley T, Dyment DA, Beaulieu CL, Schwartzentruber J, Smith A, et al. Utility of whole-exome sequencing for those near the end of the diagnostic odyssey: time to address gaps in care. Clin Genet. 2016;89: 275–284. doi:10.1111/cge.12654

27. Syngal S, Brand RE, Church JM, Giardiello FM, Hampel HL, Burt RW, et al. ACG clinical guideline: Genetic testing and management of hereditary gastrointestinal cancer syndromes. Am J Gastroenterol. NIH Public Access; 2015;110: 223–62; quiz 263. doi:10.1038/ajg.2014.435

28. Park HS, Park S-J, Kim JY, Kim S, Ryu J, Sohn J, et al. Next-generation sequencing of BRCA1/2 in breast cancer patients: potential effects on clinical decision-making using rapid, high-accuracy genetic results. Ann Surg Treat Res. Korean Surgical Society; 2017;92: 331–339. doi:10.4174/astr.2017.92.5.331

29. O'Leary E, Iacoboni D, Holle J, Michalski ST, Esplin ED, Yang S, et al. Expanded Gene Panel Use for Women With Breast Cancer: Identification and Intervention Beyond Breast Cancer Risk. Ann Surg Oncol. Springer; 2017;24: 3060–3066. doi:10.1245/s10434-017-5963-7

30. Ordulu Z, Kammin T, Brand H, Pillalamarri V, Redin CE, Collins RL, et al. Structural Chromosomal Rearrangements Require Nucleotide-Level Resolution: Lessons from Next-Generation Sequencing in Prenatal Diagnosis. Am J Hum Genet. Elsevier; 2016;99: 1015–1033. doi:10.1016/j.ajhg.2016.08.022

31. Thompson JC, Yee SS, Troxel AB, Savitch SL, Fan R, Balli D, et al. Detection of Therapeutically Targetable Driver and Resistance Mutations in Lung Cancer Patients by Next-Generation Sequencing of Cell-Free Circulating Tumor DNA. Clin Cancer Res. 2016;22: 5772–5782. doi:10.1158/1078-0432.CCR-16-1231

32. Vendrell JA, Mau-Them FT, Béganton B, Godreuil S, Coopman P, Solassol J. Circulating Cell Free Tumor DNA Detection as a Routine Tool forLung Cancer Patient Management. Int J Mol Sci. Multidisciplinary Digital Publishing Institute (MDPI); 2017;18. doi:10.3390/ijms18020264

33. Weimer ET, Montgomery M, Petraroia R, Crawford J, Schmitz JL. Performance Characteristics and Validation of Next-Generation Sequencing for Human Leucocyte Antigen Typing. J Mol Diagnostics. 2016;18: 668–675. doi:10.1016/j.jmoldx.2016.03.009

34. Szolek A, Schubert B, Mohr C, Sturm M, Feldhahn M, Kohlbacher O. OptiType: precision HLA typing from next-generation sequencing data. Bioinformatics. 2014;30: 3310–3316. doi:10.1093/bioinformatics/btu548

35. Brambati C, Galbiati S, Xue E, Toffalori C, Crucitti L, Greco R, et al. Droplet digital polymerase chain reaction for DNMT3A and IDH1/2 mutations to improve early detection of acute myeloid leukemia relapse after allogeneic hematopoietic stem cell transplantation. Haematologica. 2016;101: e157–e161. doi:10.3324/haematol.2015.135467

36. Whale AS, Devonshire AS, Karlin-Neumann G, Regan J, Javier L, Cowen S, et al. International Interlaboratory Digital PCR Study Demonstrating High Reproducibility for the Measurement of a Rare Sequence Variant. Anal Chem. 2017;89: 1724–1733. doi:10.1021/acs.analchem.6b03980

37. Ai B, Liu H, Huang Y, Peng P. Circulating cell-free DNA as a prognostic and predictive biomarker in non-small cell lung cancer. Oncotarget. 2016;7: 44583–44595. doi:10.18632/oncotarget.10069

38. Hofman P. Liquid biopsy for early detection of lung cancer. Curr Opin Oncol. 2017;29: 73–78. doi:10.1097/CCO.0000000000000343

39. Yamada T, Iwai T, Takahashi G, Kan H, Koizumi M, Matsuda A, et al. Utility of KRAS mutation detection using circulating cell-free DNA from patients with colorectal cancer. Cancer Sci. 2016;107: 936–943. doi:10.1111/cas.12959

40. Wall JD, Tang LF, Zerbe B, Kvale MN, Kwok P-Y, Schaefer C, et al. Estimating genotype error rates from high-coverage next-generation sequence data. Genome Res. 2014;24: 1734–9. doi:10.1101/gr.168393.113

41. Yamamoto T, Shimojima K, Ondo Y, Imai K, Chong PF, Kira R, et al. Challenges in detecting genomic copy number aberrations using next-generation sequencing data and the eXome Hidden Markov Model: a clinical exome-first diagnostic approach. Hum genome Var. 2016;3: 16025. doi:10.1038/hgv.2016.25

42. Li J, Dai H, Feng Y, Tang J, Chen S, Tian X, et al. A Comprehensive Strategy for Accurate Mutation Detection of the Highly Homologous PMS2. J Mol Diagn. 2015;17: 545–53. doi:10.1016/j.jmoldx.2015.04.001

43. Zhang T-H, Wu NC, Sun R. A benchmark study on error-correction by read-pairing and tag-clustering in amplicon-based deep sequencing. BMC Genomics. 2016;17: 108. doi:10.1186/s12864-016-2388-9

44. Richards S, Aziz N, Bale S, Bick D, Das S, Gastier-Foster J, et al. Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. Genet Med. 2015;17: 405–24. doi:10.1038/gim.2015.30

45. Amendola LM, Jarvik GP, Leo MC, McLaughlin HM, Akkari Y, Amaral MD, et al. Performance of ACMG-AMP Variant-Interpretation Guidelines among Nine Laboratories in the Clinical Sequencing Exploratory Research Consortium. Am J Hum Genet. 2016;98: 1067–1076. doi:10.1016/j.ajhg.2016.03.024

*Meeting the needs of Mediterranean nations: improving health with emerging technologies*

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# miRNA and other non-coding RNAs as promising diagnostic markers

Dorota Trzybulska[1], Eleni Vergadi[2] and Christos Tsatsanis[1,2]

[1] *Molecular Reproductive Medicine, Department of Translational Medicine, Lund University, Malmö, Sweden*

[2] *Department of Clinical Chemistry, School of Medicine, University of Crete, Heraklion, Greece*

ARTICLE INFO

***Corresponding author:***
Christos Tsatsanis
Medical School
University of Crete
Voutes, 70013 Heraklion, Crete
Greece
Phone: +30 2810394833
E-mail: tsatsani@uoc.gr

***Key words:***
miRNA, lncRNA, piRNA, serum, biomarkers, cancer, inflammation

***Ackgnowledgements:***
This article is part of the ReproUnion collaborative study, co-financed by the European Union, Interreg V ÖKS.

ABSTRACT

Since the discovery of non-coding RNAs (ncRNAs) a new area has emerged in the field of biomarkers. NcRNAs are RNA molecules of different sizes that are transcribed as independent genes or as part of protein coding genes and are not translated, therefore they do not produce proteins. They have been classified according to their size and function and include microRNAs (miRNAs), piwiRNAs (piRNAs), snoRNAs and long non-coding RNAs (lncRNAs). These non-coding RNAs are present in different cell compartments participating in multiple cell functions, but they have also been identified in biological fluids, also known as cell-free or circulating ncRNAs, where they can be detected in exosomes, bound on lipoproteins as well as free circulating molecules. The role of circulating ncRNAs is still under investigation but are believed to be paracrine or endocrine messengers to systematically deliver signals between cells and tissues. Detecting ncRNAs in biological fluids has opened a new field in Clinical Chemistry utilizing them as biomarkers of diseases or prognostic markers for different pathological conditions. Herein, the different types of ncRNAs and their potential in the field of diagnostics are outlined.

## INTRODUCTION

Protein-coding genes have been studied thoroughly during the last decades, even though they represent only 1.5% of the genome, which can increase to 2% if untranslated regions (UTRs) are included. The remaining genome, the non-protein coding region has remained a 'black box' until recently when characterization of non-coding RNAs (ncRNAs) has emerged, due to the development of novel nucleotide sequencing technologies. The importance of the ncRNAs has become increasingly apparent and our knowledge on the significance and contribution of ncRNAs in development and disease pathogenesis is expanding rapidly (1). NcRNAs can also explain phenotypic diversity between species given the fact that i.e. protein-coding genes are very similar among mammals while their ncRNAs do not exhibit the same level of similarity. The functional role of a class of ncRNAs, this of microRNAs (miRNAs), in cell physiology and human disease has been widely studied and acknowledged. In nearly all diseases miRNA expression pattern has been shown to differ both in tissues and extracellularly, contributing to disease pathogenesis. However, miRNAs are only the tip of the iceberg since additional ncRNAs are emerging as contributors of tissue homeostasis and regulators of cell function and fate. Hence, PIWI-interacting RNAs (piRNAs), transcribed small nucleolar RNAs (snoRNAs), ultraconserved regions (T-UCRs), large intergenic non-coding RNAs (lincRNAs) and the heterogeneous group of long non-coding RNAs (lncRNAs), are also key contributors to tissue homeostasis and disease pathogenesis. Among those, a significant number has been detected in biological fluids, allowing their use as circulating biomarkers.

Emergence of novel technologies has allowed to characterize sequences of ncRNAs in different healthy and diseased tissues as well as biological fluids from healthy and diseased individuals. The most prominent and widely studied ncRNAs are miRNAs which are already utilized as disease biomarkers.

## miRNAs AS BIOMARKERS OF HUMAN DISEASE

After the discovery of miRNAs and their first association with human disease sixteen years ago (2), their contribution in human disease has been established. miRNAs are ncRNAs 19-24 nucleotides(nt) long, and control gene expression by targeting the 3'UTR of mRNAs leading them to degradation or inhibiting translation. Alternatively, miRNAs stabilize mRNA molecules and lead to more efficient translation, thus positively affecting gene expression (3). Changes in intracellular miRNA expression have been causally associated with multiple diseases including cancer (4), neurodegenerative diseases (5), cardiovascular diseases (6) and more. Similar as well as distinct changes in miRNA expression pattern has been observed in the serum of patients, introducing detection of serum miRNAs as biomarkers of human diseases (7). MiRNAs are present in the serum in different configurations; they are transported inside exosomes, bound on lipoproteins such as LDL and HDL, and also bound on proteins such as Argonaut 2 (Ago2) (8).

All configurations allow miRNAs to be uptaken by distant tissues and cells, altering gene expression in the target cells. Initial studies were focusing in functionally relevant miRNAs but utilization of serum-specific miRNA arrays has led to the identification of additional diagnostic and prognostic miRNAs. Recent work from our group has shown that miRNA levels in the serum are potential markers linking different diseases. For example, changes in miR-155-5p, miR-200a-3p, miR122-5p and miR-200c-3p are common determinants of male subfertility and metabolic disease, suggesting potential

common causal events and mechanisms (9, 10). MiRNAs in the circulation can transmit information similar to that of cytokines, chemokines and hormones but with more specificity in targeting gene expression in the recipient cell. In the context of inflammation, expression of miRNAs such as miR-155-5p and let-7 family in the serum may modulate expression of their target genes such as SOCS1 or TLR4, thus limiting detrimental effects of inflammation or augmenting anti-pathogen responses from distant cells and organs (11-14).

## piRNAs AS BIOMARKERS

PiRNAs are ncRNAs slightly larger than miRNAs having size of 24–30 nt and are characterized by their ability to bind to the PIWI subfamily of Argonaut family proteins which are involved in maintaining genome integrity in germline cells, primarily in those of the male (15, 16). PiRNAs are transcribed from genomic regions that contain transposable elements and other repetitive elements and is believed that their function is to suppress those. PiRNA/PIWI complexes bind on transposable elements and inhibit their mobilization either by cleavage of transposable element transcripts by PIWI proteins using a mechanism based on recognition of homology between piRNAs and transposable elements or through heterochromatin-mediated silencing of transposable element transcription. PiRNAs, even though they are not abundant, they utilize the 'ping-pong' amplification cycle based on PIWI proteins (such as PIWILI1, PIWIL1 and PIWIL4) resulting in generation of antisense molecules that repress transcripts. PIWI proteins are also involved in DNA methylation and piRNAs mediate epigenetic changes (17). PiRNAs are also expressed in additional cell types to germ cells including endothelial cells but their function remains to be elucidated (18). Recent reports have shown that piRNAs are present in biological fluids, primarily in the seminal fluid and their expression has been associated to fertility (19). Whether soluble piRNAs are signals to mediate information imprinting between cells is not known. Profiling of soluble piRNAs can provide functionally significant and, more importantly, cell-source specific biomarkers for diseases.

## snoRNAs AS POTENTIAL BIOMARKERS

SnoRNAs is another family of ncRNAs of intermediate size ranging from 60 to 300 nucleotides. They are components of small nucleolar ribonucleoproteins (snoRNPs), which are complexes controlling sequence-specific 2' *O*-methylation and pseudouridylation of ribosomal RNA (rRNA). RRNAs are post-transcriptionally modified in the nucleolus facilitating rRNA folding and stability. SnoRNA sequences are responsible for targeting the assembled snoRNPs to a specific target (20). Recently snoRNAs have been detected in biological fluids and have been suggested to be useful as biomarkers (21, 22). Whether they possess functional significance in biofluids remains to be determined.

## lncRNAs AS POTENTIAL BIOMARKERS

LncRNAs include a broad and heterogeneous family of non-coding RNAs defined by their size being over 200 nt long. LncRNAs are involved in a diverse array of biological processes. This family is the largest family of ncRNAs including the largest portion of the mammalian non-coding transcriptome. The main function of lncRNAs is regulation of gene expression. Thus, lncRNAs mediate epigenetic modifications of DNA by recruiting chromatin remodeling complexes to target genes and therefore controlling their temporal and spatial expression (23). Among the known functions of lncRNAs, one involves regulating chromatin accessibility through histone modification enzymes and RNA polymerases. Physiological processes such as X chromosome inactivation occurs by the X-inactivation

specific transcript *(XIST)* lncRNA which recruits the polycomb complex to silence the X chromosome from which it is transcribed. *TSIX,* another lncRNA, is transcribed from the opposite strand to *XIST* and regulates *XIST* levels during X-chromosome inactivation (24). In addition, multiple lncRNAs are expressed by imprinted loci, where they contribute in genetic imprinting (25). A distinct family of lncRNAs are lincRNAs, which are transcribed from intergenic regions. LincRNAs control transcription, such as the p53 regulated lincRNA, lincRNA p21, which is located near the p21 gene and suppresses transcription when p53 is activated upon DNA damage (26). LincRNAs do not only regulate expression of neighboring genes but also act on distant ones. Another family of lncRNAs is this including lncRNAs transcribed from ultraconserved regions (UCRs). UCRs are conserved between vertebrates and are thought to date from a very early period in evolution. Some UCRs overlap with coding exons, although it is believed that more than half of the UCRs do not encode proteins. The UCRs that are transcribed are termed T-UTRs (27). The length of UCRs ranges from 200 to 80 nucleotides while T-UCRs have an unspliced length of up to 2kb. Their function remains unknown and the expression pattern has not been determined in disease conditions to allow them to serve as biomarkers.

**Table 1** ncRNAs, their function and use as biomarkers

| Family of ncRNAs | Size (nucleotides) | Potential function | Used as biomarker |
|---|---|---|---|
| **miRNAs** | 19–24 | Regulation of mRNA expression | Yes, widely |
| **piRNAs** | 26–31 | Repression of transposons and DNA methylation in germ cells | Yes, limited |
| **tiRNAs** | 17–18 | Regulation of transcription | no |
| **snoRNAs** | 60–300 | Regulation of rRNAs | Yes, limited |
| **PASRs** | 22–200 | Unknown | no |
| **TSSa-RNAs** | 20–90 | Transcriptional regulation | no |
| **PROMPTs** | <200 | Transcriptional regulation | no |
| **lincRNAs** | >200 | Chromatin regulation | Yes, limited |
| **T-UCRs** | 200-780 | Regulation of miRNAs | no |
| **Other lncRNAs** | >200 | Transcriptional and epigenetic regulation of gene expression | Yes, limited |

## OTHER ncRNAs AND THEIR POTENTIAL AS BIOMARKERS

Different ncRNAs have been shown to associate with transcriptional initiation sites such as the promoter-associated small RNAs (PASRs), promoter upstream transcripts (PROMPTs), the Transcription Start Site-associated RNAs (TSSRNAs) and transcription initiation RNAs (tiRNAs) (28,29). The biological role of these ncRNAs is not well characterized but it is believed that they also regulate transcription. Another family of ncRNAs is this of telomeric repeat-containing RNAs (TERRAs), which are transcribed from telomeres (30). TERRAs regulate telomerase function and secure maintenance of heterochromatin integrity (31).

## CONCLUSIONS

Since the initial characterization of non-coding RNAs and their identification as determinants on human disease pathogenesis, a new area in the field of biomarkers has emerged. Thus, our knowledge on the differential expression of different families of ncRNAs as well as their contribution in tissue homeostasis will open a new field of biomarkers that could be measured both in tissues and in biological fluids, and support disease diagnosis and prediction. A bottleneck at the moment in the field is the availability of automated rapid detection methods of ncRNAs and the identification of the most important ones that will serve as biomarkers. A list of different types of ncRNAs, their size, function and whether are currently used as biomarkers is presented in Table 1.

## REFERENCES

1. Esteller, M. 2011. Non-coding RNAs in human disease. Nat Rev Genet 12: 861-874.

2. Calin, G. A., C. D. Dumitru, M. Shimizu, R. Bichi, S. Zupo, E. Noch, H. Aldler, S. Rattan, M. Keating, K. Rai, L. Rassenti, T. Kipps, M. Negrini, F. Bullrich, and C. M. Croce. 2002. Frequent deletions and down-regulation of micro- RNA genes miR15 and miR16 at 13q14 in chronic lymphocytic leukemia. Proc Natl Acad Sci U S A 99: 15524-15529.

3. He, L., and G. J. Hannon. 2004. MicroRNAs: small RNAs with a big role in gene regulation. Nat Rev Genet 5: 522-531.

4. Esquela-Kerscher, A., and F. J. Slack. 2006. Oncomirs - microRNAs with a role in cancer. Nat Rev Cancer 6: 259-269.

5. Gupta, P., S. Bhattacharjee, A. R. Sharma, G. Sharma, S. S. Lee, and C. Chakraborty. 2017. miRNAs in Alzheimer Disease - A Therapeutic Perspective. Curr Alzheimer Res 14: 1198-1206.

6. Navickas, R., D. Gal, A. Laucevicius, A. Taparauskaite, M. Zdanyte, and P. Holvoet. 2016. Identifying circulating microRNAs as biomarkers of cardiovascular disease: a systematic review. Cardiovasc Res 111: 322-337.

7. Backes, C., E. Meese, and A. Keller. 2016. Specific miRNA Disease Biomarkers in Blood, Serum and Plasma: Challenges and Prospects. Mol Diagn Ther 20: 509-518.

8. Robbins, P. D., and A. E. Morelli. 2014. Regulation of immune responses by extracellular vesicles. Nat Rev Immunol 14: 195-208.

9. Trzybulska, D., J. Bobjer, A. Giwercman, and C. Tsatsanis. 2017. Serum microRNAs in male subfertility-biomarkers and a potential pathogenetic link to metabolic syndrome. J Assist Reprod Genet 34: 1277-1282.

10. Tsatsanis, C., J. Bobjer, H. Rastkhani, E. Dermitzaki, M. Katrinaki, A. N. Margioris, Y. L. Giwercman, and A. Giwercman. 2015. Serum miR-155 as a potential biomarker of male fertility. Hum Reprod 30: 853-860.

11. Androulidaki, A., D. Iliopoulos, A. Arranz, C. Doxaki, S. Schworer, V. Zacharioudaki, A. N. Margioris, P. N. Tsichlis, and C. Tsatsanis. 2009. The kinase Akt1 controls macrophage response to lipopolysaccharide by regulating microRNAs. Immunity 31: 220-231.

12. Vergadi, E., K. Vaporidi, E. E. Theodorakis, C. Doxaki, E. Lagoudaki, E. Ieronymaki, V. I. Alexaki, M. Helms, E. Kondili, B. Soennichsen, E. N. Stathopoulos, A. N. Margioris, D. Georgopoulos, and C. Tsatsanis. 2014. Akt2 deficiency protects from acute lung injury via alternative macrophage activation and miR-146a induction in mice. J Immunol 192: 394-406.

13. Doxaki, C., S. C. Kampranis, A. G. Eliopoulos, C. Spilianakis, and C. Tsatsanis. 2015. Coordinated Regulation of miR-155 and miR-146a Genes during Induction of Endotoxin Tolerance in Macrophages. J Immunol 195: 5750-5761.

14. Trzybulska, D., D. Eckersten, A. Giwercman, A. Christensson, and C. Tsatsanis. 2018. Alterations in Serum MicroRNA Profile During Hemodialysis - Potential Biological Implications. Cell Physiol Biochem 46: 793-801.

15. Aravin, A. A., R. Sachidanandam, A. Girard, K. Fejes-Toth, and G. J. Hannon. 2007. Developmentally regulated piRNA clusters implicate MILI in transposon control. Science 316: 744-747.

16. Carmell, M. A., A. Girard, H. J. van de Kant, D. Bourc'his, T. H. Bestor, D. G. de Rooij, and G. J. Hannon. 2007. MIWI2 is essential for spermatogenesis and repression of transposons in the mouse male germline. Dev Cell 12: 503-514.

17. Camprubi, C., R. A. Cigliano, A. Salas-Huetos, N. Garrido, and J. Blanco. 2017. What the human sperm methylome tells us. Epigenomics 9: 1299-1315.

18. Yin, K. J., M. Hamblin, and Y. E. Chen. 2014. Non-coding RNAs in cerebral endothelial pathophysiology: emerging roles in stroke. Neurochem Int 77: 9-16.

19. Hale, B. J., A. F. Keating, C. X. Yang, and J. W. Ross. 2015. Small RNAs: Their Possible Roles in Reproductive Failure. Adv Exp Med Biol 868: 49-79.

20. Kiss-Laszlo, Z., Y. Henry, J. P. Bachellerie, M. Caizergues-Ferrer, and T. Kiss. 1996. Site-specific ribose methylation of preribosomal RNA: a novel function for small nucleolar RNAs. Cell 85: 1077-1088.

21. Steinbusch, M. M., Y. Fang, P. I. Milner, P. D. Clegg, D. A. Young, T. J. Welting, and M. J. Peffers. 2017. Serum snoRNAs as biomarkers for joint ageing and post traumatic osteoarthritis. Sci Rep 7: 43558.

22. Umu, S. U., H. Langseth, C. Bucher-Johannessen, B. Fromm, A. Keller, E. Meese, M. Lauritzen, M. Leithaug, R. Lyle, and T. B. Rounge. 2018. A comprehensive profile of circulating RNAs in human serum. RNA Biol 15: 242-250.

23. Gupta, R. A., N. Shah, K. C. Wang, J. Kim, H. M. Horlings, D. J. Wong, M. C. Tsai, T. Hung, P. Argani, J. L. Rinn, Y. Wang, P. Brzoska, B. Kong, R. Li, R. B. West, M. J. van de Vijver, S. Sukumar, and H. Y. Chang. 2010. Long non-coding RNA HOTAIR reprograms chromatin state to promote cancer metastasis. Nature 464: 1071-1076.

24. Navarro, P., D. R. Page, P. Avner, and C. Rougeulle. 2006. Tsix-mediated epigenetic switch of a CTCF-flanked region of the Xist promoter determines the Xist transcription program. Genes Dev 20: 2787-2792.

25. Guttman, M., I. Amit, M. Garber, C. French, M. F. Lin, D. Feldser, M. Huarte, O. Zuk, B. W. Carey, J. P. Cassady, M. N. Cabili, R. Jaenisch, T. S. Mikkelsen, T. Jacks, N. Hacohen, B. E. Bernstein, M. Kellis, A. Regev, J. L. Rinn, and E. S. Lander. 2009. Chromatin signature reveals over a thousand highly conserved large non-coding RNAs in mammals. Nature 458: 223-227.

26. Huarte, M., M. Guttman, D. Feldser, M. Garber, M. J. Koziol, D. Kenzelmann-Broz, A. M. Khalil, O. Zuk, I. Amit, M. Rabani, L. D. Attardi, A. Regev, E. S. Lander, T. Jacks, and J. L. Rinn. 2010. A large intergenic noncoding RNA induced by p53 mediates global gene repression in the p53 response. Cell 142: 409-419.

27. Lujambio, A., A. Portela, J. Liz, S. A. Melo, S. Rossi, R. Spizzo, C. M. Croce, G. A. Calin, and M. Esteller. 2010. CpG island hypermethylation-associated silencing of non-coding RNAs transcribed from ultraconserved regions in human cancer. Oncogene 29: 6390-6401.

28. Kapranov, P., J. Cheng, S. Dike, D. A. Nix, R. Duttagupta, A. T. Willingham, P. F. Stadler, J. Hertel, J. Hackermuller, I. L. Hofacker, I. Bell, E. Cheung, J. Drenkow, E. Dumais, S. Patel, G. Helt, M. Ganesh, S. Ghosh, A. Piccolboni, V. Sementchenko, H. Tammana, and T. R. Gingeras. 2007. RNA maps reveal new RNA classes and a possible function for pervasive transcription. Science 316: 1484-1488.

29. Preker, P., J. Nielsen, S. Kammler, S. Lykke-Andersen, M. S. Christensen, C. K. Mapendano, M. H. Schierup, and T. H. Jensen. 2008. RNA exosome depletion reveals transcription upstream of active human promoters. Science 322: 1851-1854.

30. Montero, J. J., I. Lopez-Silanes, D. Megias, F. F. M, A. Castells-Garcia, and M. A. Blasco. 2018. TERRA recruitment of polycomb to telomeres is essential for histone trymethylation marks at telomeric heterochromatin. Nat Commun 9: 1548.

31. Koch, L. 2017. Non-coding RNA: A protective role for TERRA at telomeres. Nat Rev Genet 18: 453.

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# NGS for metabolic disease diagnosis

Dèlia Yubero[1], Rafael Artuch[2]

[1] *Department of Genetics and Molecular Medicine, Institut de Recerca Sant Joan de Déu, Barcelona, Spain*

[2] *Department of Clinical Biochemistry, Institut de Recerca Sant Joan de Déu, and CIBER de Enfermedades Raras (CIBERER), Barcelona, Spain*

INFO

***Corresponding author:***
Dèlia Yubero
Department of Genetics
and Molecular Medicine
Institut de Recerca Sant Joan de Déu
Barcelona, Spain.
Phone: + 34936009451
E-mail: dyubero@sjdhospitalbarcelona.org

***Key words:***
inborn errors of metabolism,
next generation sequencing

LETTER TO THE EDITOR

Inborn errors of metabolism (IEM) comprise heterogeneous and rare genetic diseases with a variety of overlapping or unspecific clinical phenotypes. Multiple proteins with enzymatic, transporter, regulatory and other functions participate in the complexity of metabolic pathways.

The breakdown of the normal function of some of these proteins may impair the metabolic state of an organism. These disruptions can generally be assessed biochemically through the detection of metabolites in different biological fluids.

However, the specificity and sensitivity of some of these biomarkers are not always high. IEM are, generally, severe diseases, and the accurate identification of the molecular basis of these diseases is important for appropriate patient treatment and genetic counselling.

Thus, even though the establishment of an IEM diagnosis is supported by clinical suspicion and biochemical investigations, genetic investigations also play a significant role. Genetic diagnosis in clinical practice has substantially changed in the past decade. The incorporation of next generation sequencing (NGS) technologies allows researchers, depending on the

selected strategy, to obtain the molecular sequence of the desired genes simultaneously, offering high-quality data. The utility of NGS in the clinical field has been widely demonstrated in different groups of diseases (1,2,3), but the actual debate encompasses two main points: the reinterpretation of the classical diagnostic algorithms and the chosen NGS strategy.

Today, regarding NGS strategies, targeted gene panels have been progressively replaced with larger panels, including all known disease-associated genes, or directly by whole exome sequencing (WES) (4).

In terms of rare genetic diseases, the identification of the genetic basis of disease is reached in 20 to 40% of patients using WES (5). Some of the reasons why up to 70% of patients might remain unsolved are explained by methodological issues like incomplete coverage of the exome and genetic mutations elusive to the technology itself. However, in most of the cases, the disease-causing variant is within the WES data, but there is insufficient evidence to support a definitive diagnosis (5).

Whole genome sequencing (WGS) is the ideal framework, in which a unique approach provides the most complete knowledge of an individual genetics, offering the possibility to analyze and interpret the genomic data along with the advances of scientific learning (6).

In a recent work, we demonstrate the importance of biomarkers as a key clue for genetic diagnosis achievement (7). In spite of this, the most common picture in IEM is represented by a patient with unspecific clinical features and also unspecific biomarkers. Although accurate characterization of clinical, biochemical and pathological patterns of patients are immensely valuable to understand genetic findings, NGS technology can be a step forward in terms of diagnostic issues. Some IEM are straight ahead to the causative gene, but others are not so easy to solve.

One of the most evident examples are CoQ deficiency syndromes, which are defects of the energy metabolism system. Several proteins with unclear molecular functions facilitate CoQ biosynthesis through unknown mechanisms, and multiple steps in this pathway are catalyzed by currently unidentified proteins (8).

This intricate metabolic process implies unidentified enzymes and, there are several genes that remain to be identified as essential for CoQ biosynthesis regulation. Furthermore, negative findings in COQ genes do not completely discard the possibility of a CoQ primary deficiency.

Thereby, biochemical measurements maintain a significant role in the diagnostic strategy. The heterogeneity of clinical and biochemical patterns in this specific mitochondrial disease reinforces the idea of using widespread NGS strategies like huge genic panels or whole exome sequencing to reach molecular diagnosis.

In conclusion, analysis and comprehension of genomic data must be closely connected to all the hints that classical biomarkers can offer, which is crucial information to guide the interpretation of an individual's exome or genome.

## REFERENCES

1. Wong LJ. Challenges of bringing next generation sequencing technologies to clinical molecular diagnostic laboratories. Neurotherapeutics 2013;10:262-272

2. DaRe JT, Vasta V, Penn J, et al. Targeted exome sequencing for mitochondrial disorders reveals high genetic heterogeneity. BMC Med Genet 2013;14:118

3. Ghosh A, Schlecht H, Heptinstall LE, et al. Diagnosing childhood-onset inborn errors of metabolism by next-generation sequencing. Arch Dis Child 2017;102(11):1019-1029

4. LaDuca H, Farwell KD, Vuong H, et al. Exome sequencing covers >98% of mutations identified on targeted next generation sequencing panels. PLoS One 2017;12(2):e0170843

5. Sawyer SL, Hartley T, Dyment DA, et al. Utility of whole-exome sequencing for those near the end of the

diagnostic odyssey: time to address gaps in care. Clin Genet 2016;89(3):275-284

6. Petersen BS, Fredrich B, Hoeppner MP, et al. Opportunities and challenges of whole-genome and -exome sequencing. BMC Genet 2017;18(1):14

7. Yubero D, Brandi N, Ormazabal A, et al. Targeted Next Generation Sequencing in Patients with Inborn Errors of Metabolism. PLoS One. 2016;11(5):e0156359

8. Stefely JA, Pagliarini DJ. Biochemistry of Mitochondrial Coenzyme Q Biosynthesis. Trends Biochem Sci 2017;42(10):824-843

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# Reflections on the mentor-mentee relationship: a symbiosis

Josep Miquel Bauça[1,2]

[1] *Department of Laboratory Medicine, Hospital Universitari Son Espases, Palma, Balearic Islands, Spain*

[2] *Institut d'Investigació Sanitària de les Illes Balears (IdISBa), Spain*

INFO

***Corresponding author:***
Josep Miquel Bauça
Ctra. de Valldemossa, 79
07010 Palma, Mallorca
Spain
Phone: +34 871205876
E-mail: pepmiquel@gmail.com

***Key words:***
clinical biochemistry,
teaching, research

***Disclosure:***
The author declares no conflict of interest.

LETTER

Residents in laboratory medicine need to develop the proper skills to become true clinical scientists for the mid-21st century. A key figure towards expertise is the mentor, who is responsible for the motivation and guidance of the young scientist from the moment of their landing in the laboratory.

It is essential for the mentor to assure the expertise of the mentee in the different areas of wisdom in laboratory medicine, which encompass laboratory organization and management, analytical techniques (instrumentation and methodology) and clinical outcomes (pathophysiology, test usefulness and appropriateness and result interpretation) (1). Towards this purpose, not only should the mentor guide the young scientist along the residency path, but also check and proof that the required concepts, aptitudes and abilities were properly acquired and integrated.

Every year, new analytical tools and devices are developed, and more sensitive and accurate biomarkers are brought from the bench to the clinic, so it is of utmost importance not only for the young clinical scientists but also for their senior mentors to keep up to date. Sail or sink.

The otherwise called 'advisors', 'coaches' or 'counsellors' also need to motivate and encourage their mentees to be involved and master the three basic pillars of laboratory medicine: healthcare, research and teaching. Each of them is absolutely essential, and to outstand in our medical field, none of them should be forgotten (Figure 1). With no doubt, we all agree that patient care and safety are our ultimate goal and the reason we (laboratory professionals) wake up every day -earlier and more accurate diagnostic strategies, better prognostic tools and improved means to check the effectiveness of treatments and prediction of relapse. However, no excellence may be reached if not involved in scientific research and the transmission of knowledge to others.

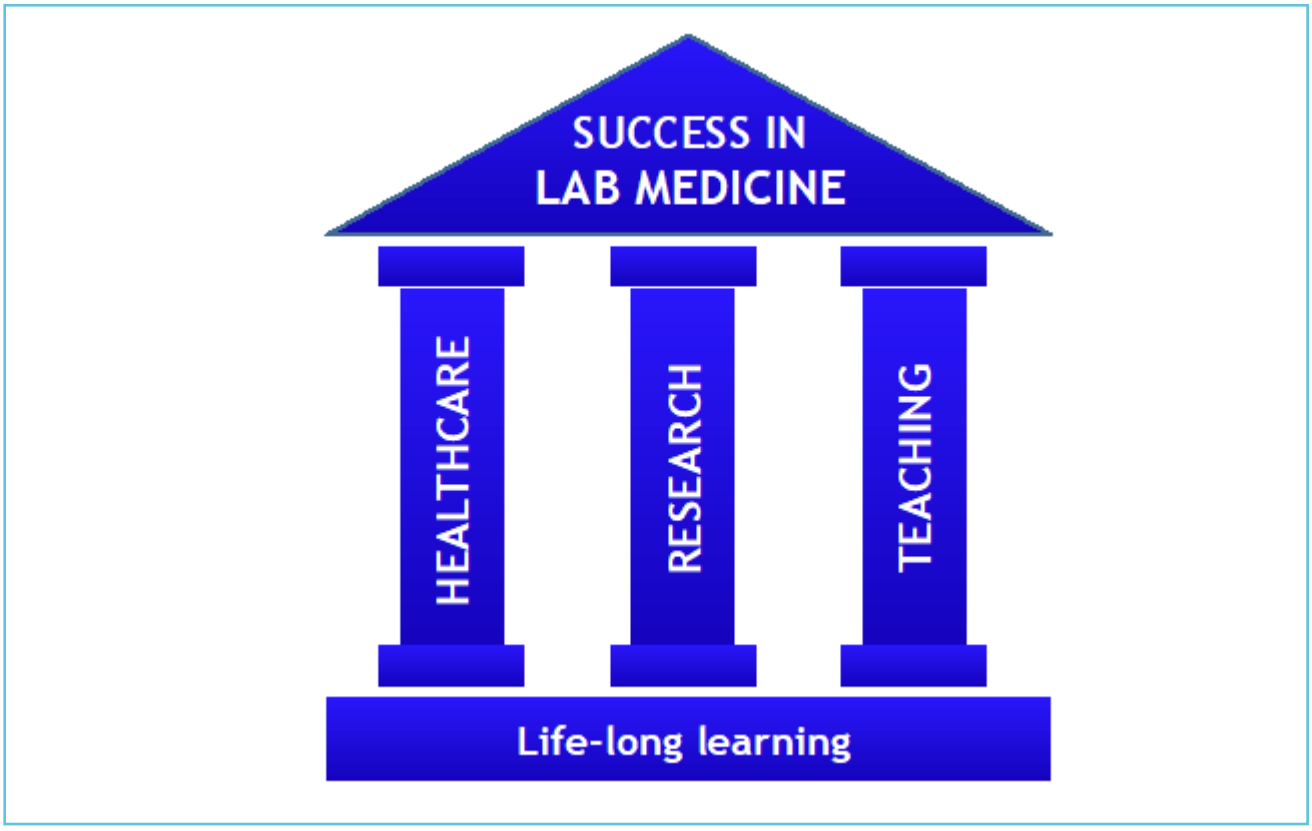
Besides the need of technological development in our field, the active participation of a young scientist in research projects and scientific discussions makes a resident more independent and gives them a powerful and critical thinking.

Those who do not participate in research themselves are forced to believe what others say or tell them. Investigation may also help in-house method development and translate into interesting economic benefits for the whole institution itself. Nevertheless, the taking-off and specialization of a young scientist in research are usually of extreme toughness, so the aid of the mentor is highly appreciated.

And teaching. By preparing lectures to a big audience, giving a presentation to our closest colleagues or simply by explaining basic concepts to students, a laboratory professional strengthens their knowledge on a specific topic.

As we prepare those explanations and anticipate possible questions, further concepts appear, and it is easier to secure them in our minds.

**Figure 1** Pillars in Laboratory Medicine



Knowledge, advice and benefits do not flow only one-way from the (senior) mentor to the (young) mentee, but the profits do also travel the other way around. Teamwork. Mutual growth. A proper mentorship represents a full commitment and creates an environment of trust and enrichment; a climate of collaboration between two professionals with common interests; a long-term biological interaction between two living organisms. An explicit and respectful alliance.

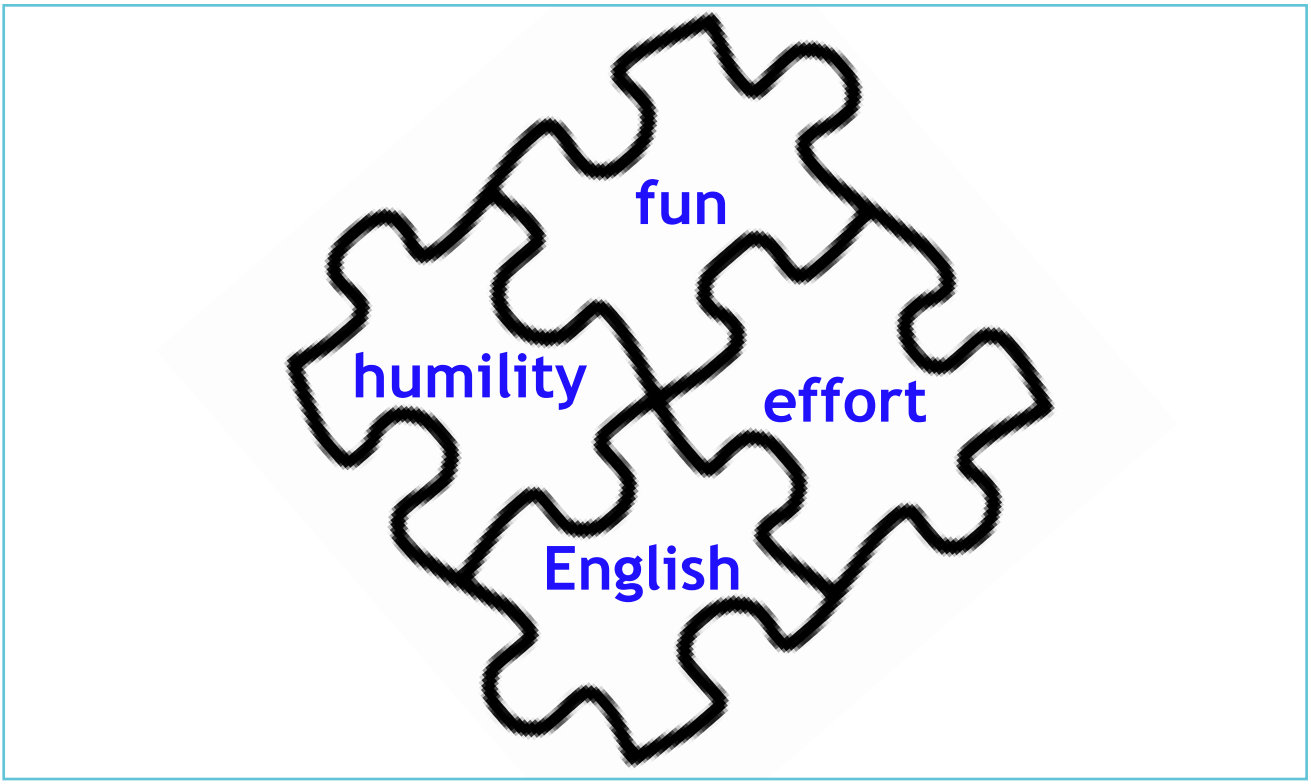
Thanks to an altruistic, generous and patient personality, the mentor learns from the mentee's questions and novel ideas, and is intellectually stimulated as a result of the exposure to new information or to the relearning of past material; rejuvenation (2).

The engagement and participation of both senior and junior professionals in academic projects is also of substantial interest for the institution. The preparation of courses, the publication of research findings, the introduction of newly-described analytical procedures and biomarkers in the laboratory and the participation in clinically-focused meetings or committees, among other, helps to create this climate of well-being and enthusiasm for both. A great example of such win-win situation may be found in a previous manuscript on this journal (3). The mentor shapes the mentee, who in turn does shape the mentor.

This two-way street demands attentive listening, defying the mentee with increasingly complex challenges, giving them autonomy, let them take risks and face both wins and failures (4,5). This atmosphere requires a fluid communication with emotional support and psychologic encouragement.

In addition to personal fulfillment, a publication for the mentee (whether article, book chapter, poster or oral communication in a congress) is a publication for the mentor; an award for the

**Figure 2** Requirements for good residency training



mentee turns into a recognition for the people who made that possible and a grant or internship for them turns into a considerable opportunity for all the working group.

Besides the guidance by a mentor, additional key contributions need to be made by the mentee him- or herself. According to Dr. A. Buño (Hospital La Paz, Spain), a successful residency and career in laboratory medicine requires fun, effort and humility.

We could also add that proficiency in the English language for non-English speakers means a great leap for them (Figure 2).

Fun is, by far, the most important. Happy, proactive and passionate residents (not surprisingly) get luckier. Inspiration comes with fun. After all, a laboratory professional spends large amounts of time at work, so it is great advice to have as much fun as possible there. Effort: although it does not guarantee success, it has proven to be a great source of confidence. A smooth sea never made a skilled sailor, so no effort translates into no gain.

And in regard to humility, scientists need to identify their limitations, value group work as well as individual collaborations, help others and not be afraid of making mistakes. Humans are error-prone, so residents are too. It is humility itself what enables the learning process. If own failures are not recognized, progress and growth are inconceivable.

Embarking in a mentorship program will (surely) provide greater confidence and career satisfaction for the mentor, along with stronger connections within the clinical biochemistry community. And for the mentee, success may be reached in any part of the world by integrating all the above-mentioned abilities, effort, enthusiasm, and with the irreplaceable feedback and advice from a trusted mentor. It just requires having fun.

## REFERENCES:

1. Greaves R and Smith JM. The IFCC Curriculum. Rev 0 – 2017.

2. Henry-Noel N, Bishop M, Gwede CK, Petkova E, Szumacher E. Mentorship in Medicine and Other Health Professions. J Cancer Educ 2018 Apr 24.

3. IFCC Task Force for Young Scientists presents: a mentorship interview. IFCC eNews June 2018.

4. Craig PA. Lessons from my undergraduate research students. J Biol Chem 2018 [Epub ahead of print].

5. Toklu HZ, Fuller JC. Mentor-mentee Relationship: a Win-Win Contract in Graduate Medical Education. Cureus 9(12):e1908

The Journal of the International Federation of Clinical Chemistry and Laboratory Medicine

eJIFCC



# Inter-laboratory exchange of knowledge and technology around our Sea – a lab surfing project

Guilaine Boursier[1,2]

[1] *IFCC Task Force for Young Scientists*

[2] *CHU Montpellier, Univ Montpellier, Department of Genetics, Montpellier, France*

INFO

***Corresponding author:***
Guilaine Boursier
Department of Genetics
CHU Montpellier, University of Montpellier
Montpellier
France
E-mail: guilaineboursier@yahoo.fr

***Key words:***
networking, lab-surfing

LETTER

There is a need for stringent relationships between young scientists in laboratory medicine from all over the world and networking is the key to ensure a good communication and to encourage participation of young scientists.

In this aim, the task force for young scientists developed some ways of communication to extend the young scientists network and uniting young scientists globally.

Our community includes the five core members ensuring geographical representation, one corporate member, corresponding members nominated by thirty-six different National Societies and also more than thousand members through our different media. Thanks to our regular workshops for young scientists organized worldwide within the framework of IFCC&LM international or regional meetings, the community grows every year.

Different kinds of social networking media are easily accessible on the web. More than six hundred people follow us on our Facebook page, Twitter account and our LinkedIn group, where we share our activities (@ifccYOUNG). We communicate through our webpage (http://www.ifcc.org/task-force-young-scientists-web-pages/) and mailing list which is a Google group

including more than 280 members (ifcc-task-force-ys@googlegroups.com).

All this ways of communication are devoted to establish formal and informal networks to facilitate the communication between young scientists who are involved in laboratory medicine.

Our social medias facilitate regular exchange of information dealing with activities of the task force, moreover young scientists can submit interrogations to the community when they are faced to a problem in their clinical practice.

Our best way of inter-laboratory exchange is the Lab-surfing project initiated in 2016. A successful evening event dedicated to young scientists was conducted at EuroMedLab Paris congress in 2015. Strong relationships between young delegates have emerged.

That's how the two founders of the Lab-surfing project, Marie Lenski (France) and Santiago Fares Taie (Argentina) have met. They both like travelling and discovering foreign countries by themselves, meeting new people. They were interested in connecting young scientists and facilitating opportunities for them to share their experiences.

The Lab-surfing website (https://www.lab-surfing.com/) was created the next year with the support of IFCC&LM through the task force for young scientists. Today, more than six hundred users are registered at the website.

For those interested in networking with young scientists from foreign countries, once registered, Lab-surfing is a tool where you can find a contact from the country of interest with only two clicks.You will find some information about the contact and can reach him directly by email. Last year, a discussion forum was created for improving fast and easy communication about the fields of laboratory medicine.

Lab-surfing project encourage young scientists to exchange knowledge and to contribute to advances in our field of work. Our community aims at helping you to face the ongoing challenges in laboratory medicine. Join us!

**Publisher:** IFCC Communications and Publications Division (IFCC-CPD)



The eJIFCC is a member of the **Committee on Publication Ethics (COPE).**

The eJIFCC (Journal of the International Federation of Clinical Chemistry) is an electronic journal with frequent updates on its home page. Our articles, debates, reviews and editorials are addressed to clinical laboratorians. Besides offering original scientific thought in our featured columns, we provide pointers to quality resources on the World Wide Web.



**Produced by:** Insoft Digital Web Solutions **www.insoftdigital.com**

**Published by:** IFCC International Federation of Clinical Chemistry and Laboratory Medicine **www.ifcc.org**