

November 2020

ISSN 1650-3414

Volume 31 Number 4

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](mailto:ejifcc@ifcc.org)

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



## In this issue

<b>Foreword: Ethics in laboratory medicine</b> Guest editor: Nilda E. Fink	<b>260</b>
<b>Codes of ethics for laboratory medicine: definition, structure and procedures – a narrative review based on existing national codes</b> Richard X. Davey	<b>262</b>
<b>Ethics in laboratory medicine: perspectives and challenges in resource limited settings</b> Sudip Kumar Datta	<b>274</b>
<b>Research ethics committees in laboratory medicine</b> Lamis Beshir	<b>282</b>
<b>Conflicts of interest and an approach to managing them</b> Nilda E. Fink	<b>292</b>
<b>A framework to ethically approach incidental findings in genetic research</b> Lamis Beshir	<b>302</b>
<b>Ethics and the electronic health record: description of an integrating system of electronic health records in Argentina and a proposal to shift towards a patient-centered conception</b> Julián Verona	<b>310</b>
<b>How does the MedTech Europe Code of Ethical Business Practice affect the activities of professional societies in laboratory medicine?</b> Valdas Banys	<b>320</b>

## In this issue

### **The clinical laboratory: a key player in diagnosis and management of COVID-19**

Sojit Tomo, Sreenivasulu Karli, Karthick Dharmalingam,  
Dharmveer Yadav, Praveen Sharma

---

**326**

### **Broadening the list of differential diagnosis for acute abdomen**

**– a case report from Nepal**

Vivek Pant, Keyoor Gautam, Devish Pyakurel, Aabha Shrestha,  
Santosh Pradhan, Neeraj Joshi

---

**347**

### **Galactorrhoea and hyperprolactinaemia in a non-pregnant female with burns**

Marli van Heerden, Dineo Mabuza

---

**354**

### **Improved COVID-19 testing by extraction-free SARS-CoV-2 RT-PCR**

Khelil Mohamed Mokhtar

---

**362**

# Foreword: Ethics in laboratory medicine

Guest editor: Nilda E. Fink<sup>1,2</sup>

<sup>1</sup> On behalf of the IFCC Task Force on Ethics (TF-E)

<sup>2</sup> PROES Program, Biochemical Foundation of Argentina

---

## ARTICLE INFO

**Corresponding author:**

Nilda E. Fink  
Chair, IFCC Task Force on Ethics (TF-E)  
Director, Programa PROES  
Fundación Bioquímica Argentina  
Viamonte 1167  
Buenos Aires C 1053  
Argentina  
E-mail: [nilda.fink@yahoo.com.ar](mailto:nilda.fink@yahoo.com.ar)

**Key words:**

clinical laboratory, laboratory medicine, ethics, bioethical principles, ethics code, conflict of interest, disclosure, management, patient-centered electronic health record, incidental findings, research ethics, research ethics committees, resource limited settings, biological samples, autonomy, informed consent, justice, beneficence, non-maleficence, MedTech Europe Code, continuous professional development

---

## FOREWORD

The field of ethics involves concepts and rules of right and wrong behavior. Bioethics is defined as a branch of applied ethics that studies the philosophical, social, and legal issues arising in medicine and life sciences. Presently it is mandatory for different areas of Medicine to comply with ethical standards, and the field of Laboratory Medicine is no exception. Notwithstanding, there is variability in ethics education within health professions (1).

Although there are recent publications on the topic of ethics (2), Members of the IFCC Task Force on Ethics (TF-E) further contribute to the goal of ethics education in the field of Laboratory Medicine in collaboration with the eJIFCC in the form of this dedicated thematic issue. This special issue of the eJIFCC presents a series of manuscripts that summarize relevant aspects on Ethics. Seven manuscripts are included, four of them are updating on classical ethical topics, two refer to more recent challenges in Ethics and finally, but equally important, an opinion paper.

In the first of these four articles, Davey presents an analysis of Codes of ethics for laboratory medicine including aspects such as its definition, structure and procedures. As highlighted in the title, it is a narrative

review based on existing national codes that complement his previous work available on the IFCC website (3).

Currently, diagnosis and management of patients is very much dependent on laboratory diagnostics, as such utility of data obtained from patients mandate ethical guideline-dependent actions. Ethical standards and practices vary and are resource dependent. Datta discusses ethical challenges for Laboratory Medicine in resource limited settings.

Beshir has compiled an article for better comprehension of Research Ethics Committees (RECs) in Laboratory Medicine. International ethical guidelines and declarations ensure the autonomy, dignity and well-being of research subjects, as well as integrity and credibility of research results. Here, the author defines the RECs in laboratory medicine and described their roles based on the examination of the requirements of ethical research.

Conflicts of interest (COIs) exist frequently in medicine and science. Physicians in patient care, health professionals in pharmaceutical and biomedical industry, in management positions, in teaching or in research, must apply rigid ethical principles. It is likely that COIs prop up in numerous settings. All relevant actions must comply with the essential principles of Bioethics. In the article by Fink, several aspects on conflict of interest, in terms of its definition, classification, applications, management and other challenges are described.

As we mentioned previously, another two articles deal with challenging topics from the ethical point of view. One of them is the second article in this issue penned by Beshir and is related to incidental findings (IFs). With the advancement in areas of genetics and genomics, special and additional ethical considerations should be made as genetic research can reveal

information about susceptibility of an individual to disease. The duty of researchers to disclose IFs to participants under ethical guidelines is discussed and an approach to its disclosure is recommended. In the article written by Verona, Integrating System of Electronic Health Record applied at city level in a Latin-American country is described. Detailed ethical considerations taken into account were analyzed as patient-centered conception was employed.

Finally, Banys presents in an opinion paper, in the form of an answer to a question referred to the MedTech Europe Code of Ethical Business Practice created by the medical technology industry, and how it influences the activities of professional societies in Laboratory Medicine. He described the experience of a National Society, the Lithuanian Society of Laboratory Medicine (LLMD). It addresses the importance of an ethical use of resources and fair management of educational grants, public disclosure of provided educational grants, compliance of conferences with the Conference Vetting System and other allocation of resources as examples.

The TF-E with this thematic issue, seeks to promote Ethics teaching, to catalyze discussion between stakeholders, and perhaps may also prove useful for improving guidelines and documents at local or national levels.

## REFERENCES

1. Bruns DE, Burtis CA, Gronowski AM, McQueen MJ, Newman A, Jonsson JJ, IFCC Task Force on Ethics. Variability of ethics education in laboratory medicine training programs: results of an international survey. *Clin Chim Acta*. 2015;442:115-8.
2. Gronowski AM, Budelier MM, Campbell SM. Ethics for Laboratory Medicine. *Clin Chem*. 2019;65(12):1497-1507.
3. Davey R, IFCC Task Force on Ethics. A survey of extant Ethics Policies. Available at [https://www.ifcc.org/media/478424/tf-e\\_comparison\\_of\\_codes-2020-04-30.pdf](https://www.ifcc.org/media/478424/tf-e_comparison_of_codes-2020-04-30.pdf). Accessed, Oct 2020.

# Codes of ethics for laboratory medicine: definition, structure and procedures – a narrative review based on existing national codes

Richard X. Davey

On behalf of the IFCC Task Force on Ethics (TF-E)

---

## ARTICLE INFO

### **Corresponding author:**

Richard X. Davey, FRCPA  
PO Box 310  
Shoreham, 3916  
Australia  
E-mail: [richardd18@bigpond.com](mailto:richardd18@bigpond.com)

### **Key words:**

clinical laboratory, ethics code

---

## ABSTRACT

### **Background**

It behoves every national society of clinical laboratory medicine to have a well formulated and publicly accessible policy concerning the morally acceptable way in which its members should practise their profession; such a policy is published as a Code of Ethics.

This Code assists its members in the performance of their duties in relation to the patients they share with other clinicians, within their own particular professional environment and, at large, to the rest of their national society.

### **Methods and result**

The International Federation of Clinical Chemistry and Laboratory Medicine's (IFCC) Task Force on Ethics here examines a curated selection of extant Codes and provides guidance at the level of definition, structure and procedures to assist national societies

and their clinical chemistry and laboratory medicine professionals in the task of crafting their own Ethics Code.



The leading aims of the Task Force on Ethics of the IFCC (TF – E), the International Federation of Clinical Chemistry and Laboratory Medicine, are:

- To increase awareness among Laboratory Medicine Professionals of ethical issues, *whence*
- To encourage the practice of Laboratory Medicine to the highest ethical standards *and to assist in the process,*
- To develop guidance documents for member societies on ethics related issues.

Whilst the TF – E accepts that it cannot produce documents for individual member societies at the national level, such guidance documents may be seen as a part of a “tool kit” with which such member societies can construct a Code of Ethics that is fit for purpose within their individual jurisdiction whilst at the same time preserving the essentials accepted world-wide as vital to such codes.

This work was envisaged by the foundation TF – E group nearly 20 years ago, and is now offered for use. The prior input from the initial TF members led by the then chairman, Professor Leslie Burnett, and subsequent chairholders and members is acknowledged here and in pertinent references.

## HISTORICALLY

TF – E members have previously noted [1] that the evolution of biologically focussed ethics over the years is well documented and includes

- the Nuremberg Code from 1947 [2],
- the Declaration of Geneva from 1948 [3],

- the Declaration of Helsinki from 1964 [4], and
- the Belmont report from 1978 [5].

The need for these documents was driven by developments in medical research, initially during and then after the twentieth century’s “World War 2”, but concepts in the Declaration of Geneva and the Belmont report are also applicable to the practice of clinical medicine.

The Belmont Report [5] is one key work concerning ethics and healthcare research. Created in 1978, by the U.S.A. National Commission for the Protection of Human Subjects of Biomedical and Behavioural Research, it outlines a number of ethical principles and guidelines for the protection of human subjects. It identifies the following three core principles:

1. Respect for persons: The acknowledgement of human autonomy but, complementarily, the need for protection of those with diminished autonomy.
2. Beneficence: The duty to act in the best interests of patients or research subjects, the goal being to maximise benefits and minimise harm, the latter sometimes Latinised as *non-maleficence*.
3. Justice: The obligation to treat all patients equally and to distribute, by allocating fairly, what is rightly due in terms of benefits, risks and cost.

These principles can be applied to both research and clinical settings. They must be applied equally to clarify the ethical issues in clinical chemistry and laboratory medicine.

## THE SCENARIO ENVISAGED

The scenario to be addressed has altered little since Burnett wrote in 2007 [6], though further specific demands may have appeared. Burnett is paraphrased and extended.

Laboratory Medicine organizations and their professional members have a goal and responsibility to benefit the health and wellbeing of the patients and communities they serve. This test of their professional responsibility demands that they do not simply perform tests and use technology uncritically. They cannot be isolated from the impact of their work on society.

Ethics has the potential to make demands of all laboratorians, whichever discipline they work within, and at no less than the three different levels described below.

Firstly, personal ethics describes the pertinent, personal set of moral beliefs which governs how each of us lives our life. One's personal moral code will probably stand on and spring from a universally acknowledged minimal framework, and it thus may readily resemble other humans' efforts thereat, but it is also vital to acknowledge that each human is a unique individual and must be respected as such. The extent to which the individual's personal code is driven by community consensus, religion, personal study and reflexion, or some combination thereof, is the individual's choice. Apart from the interplay at the level of respecting autonomy and ensuring beneficent outcomes from the individual's personal professional activity, this aspect of one's conformance with ethics is not *sui generis* within the scope of this review. It is the responsibility of the individual.

Secondly, one's professional ethics describe the set of standards we each personally seek to apply in our working environment and organizations. Some of our professional ethics are governed by scientific protocols and standards and relate to the way in which we operate our laboratories, while others relate to the way in which we conduct ourselves to promote the good standing and advancement of our profession. Here we are aiming to most beneficially serve the needs of both our patients and our peers.

Thirdly, the ethics of our profession is not the same thing as one's own professional ethics. It goes further: to our work as a body of professional practitioners, working together as a profession where we must consider what together we should do to meet our societal obligations in Clinical Chemistry and Laboratory Medicine at large, in short, the needs of the people. In practice, however, professional ethics and the ethics of the profession cannot be dealt with separately since we are the practitioners. The profession is us. What we do as individual craftsmen is what is done by the profession; it is thus seen by society.

In constructing an Ethics Policy that is fit for purpose within their individual jurisdiction national societies will thus formulate their own unique document, integrating as they do, the demands cited above.

### **Terminology, a footnote in text**

It seems necessary to address explicitly a potentially confounding conundrum, thereby to avoid confusion. Thus, although the practice of Clinical Chemistry and Laboratory Medicine is driven by science and should vary little across the world, related terminology does vary at national level.

Clinical Chemistry and Laboratory Medicine may be described as Biochemistry, Clinical Biochemistry, Chemical Pathology and by still other titles. Similarly, the term "Laboratory Medicine Professionals" both encompasses an array of terms that describe the practitioners and also incorporates all levels of expertise within the profession. Practice concerning who may do what within a laboratory hierarchy differs between different jurisdictions.

In some countries both technologists (meaning by this term people without university degrees) and scientists (people with such degrees) may work as laboratory practitioners, but in others



only pertinent degree holding scientists qualify for employment. In some countries only people who initially trained as medical practitioners and who have gone on then to gain post-graduate qualifications as Laboratory Physicians or Pathologists may lead or direct laboratories, but in others such a level within the laboratory's hierarchy may also be open to scientists or, further, to other initially scientifically trained people such as pharmacists.

The term “practitioner” may be a convenient general description for the practicing laboratory professional that can be deployed across the board. It necessarily also permits levels of expertise and responsibility to be categorised within the body of practitioners by a suitable set of titles.

The underlying need in drafting Ethics Codes is to be consistent with the given jurisdiction's legal requirements for the qualifications and experience required by, and the description of, the given practitioner at the given level of expertise. The requirement to practice Laboratory Medicine to the highest achievable ethical standards equally challenges practitioners at all levels of expertise.

## THE CURRENT SCENE

Only a minority of pertinent national societies have a published Ethics Code as of 2019. It is the hope of the TF-E that this instrument may help many more to craft, and to publish, theirs.

### ISO

Why ISO? ISO, the International Organization for Standardization, based in Geneva, Switzerland, is an independent, non-governmental international organization with a membership of 164 national standards bodies [7]. In its words, it “develops voluntary, consensus-based, International Standards, documents that provide requirements, specifications, guidelines or char-

acteristics that can be used consistently to ensure that materials, products, processes and services are fit for their purpose.”

In particular, the ISO standard 15189:2012, *Medical laboratories – Requirements for quality and competence*, specifies requirements for quality and competence in medical laboratories [8]. It “can be used by medical laboratories in developing their quality management systems and assessing their own competence. It can also be used for confirming or recognizing the competence of medical laboratories by laboratory customers, regulating authorities and accreditation bodies”, and routinely is so used. Its comments on ethics are therefore potentially essential input into the process of crafting a national Laboratory Ethics Code. In prior editions cited as an appendix to the Standard, the inclusion in 2012 of the ethics material in to the text of the Standard itself raises its level of “importance”.

ISO 15189's section 4, Management requirements, at 4.1.1.3 thus specifically requires that “Laboratory management shall have arrangements in place to ensure the following:

- a. there is no involvement in any activities that would diminish confidence in the laboratory's competence, impartiality, judgement or operational integrity;
- b. management and personnel are free from any undue commercial, financial, or other pressures and influences that may adversely affect the quality of their work;
- c. where potential conflicts in competing interests may exist, they shall be openly and appropriately declared;
- d. there are appropriate procedures to ensure that staff treat human samples, tissues or remains according to relevant legal requirements;
- e. confidentiality of information is maintained.”

Given that this listing is framed as advice to management for the purpose of ordering a laboratory's activity, its sequence is understandable and it goes to many of the questions that need a directive, however as a model for framing a Society's own Ethics Code, its prioritising the avoidance of evil ahead of actively doing good may not be the better order (of those two) to choose.

### Extant society policies

In general, there are two different approaches that have been adopted in writing such policies by National Societies. Both focus on the duties involved in acting ethically well.

One approach categorises the task by the focussed target of duty, thus almost invariably:

- patient,
- professional peer, and
- pertinent population or wider society,

usually, though not necessarily, in that order.

The other categorises the task by form of activity, and here the products are rather more variable, defying tabulated comparison. In each case many of the extant national society policies seen have been examined. Three illustrative

policies were selected as exemplars for comparison of each of the two methods.

### Policies codified by focus of duty as the segregator

Here three typical codes have been selected, one from each of the U.S.A. [9], Poland [10], and Australia [11], and cross tabulated; the Polish code was originally published in Polish. Each code is at least a decade old in 2019. There is a range of linguistic prolixity and depth of detail addressed, although the textual cross dependence is obvious. Whether the subsequent users have improved the prior published text is a decision for the reader.

These examples might be considered to contain the essentials accepted world-wide as vital to such policies, the elements *sine que non*, but of course individual Societies must be free to add elements that their own circumstances, or their jurisdiction's law, or both, demand, and equally, are free to choose the style of drafting that suits them. Similarly, they should not be afraid to utilise pre-existing text if it appears to be as close to an acceptable statement of the matter addressed as can be achieved. (Table 1)

**Table 1** Three extant clinical laboratory ethics codes, textually compared, using the focus of duty as the task segregator

	USA, (American) Society for Clinical Laboratory Science [9]	Poland National Chamber of Medical Laboratory Specialists [10]	Australasian Association of Clinical Biochemists [11]
<b>Year</b>	< 2009	13 January 2006	< 2010
<b>PREAMBLE</b>	The Code of Ethics of the American Society for Clinical Laboratory Science sets forth the principles and standards by which Medical Laboratory	The Code of Ethics of the Laboratory Diagnostician is a set of basic ethical norms which should guide each representative of this profession and is the	The Code of Ethics of the Australasian Association of Clinical Biochemists (AACB) sets forth the principles and standards by which clinical laboratory

	Professionals and students admitted to professional education programs practice their profession.	basis for the personal and professional formation of a laboratory diagnostician.	practitioners practice their profession.
<b>1. Duty to the Patient</b> <i>Focus</i>	<p>Medical Laboratory Professionals' primary duty is to the patient, placing the welfare of the patient above their own needs and desires and ensuring that each patient receives the highest quality of care according to current standards of practice.</p> <p>High quality laboratory services are safe, effective, efficient, timely, equitable, and patient-centered.</p> <p>Medical Laboratory Professionals work with all patients and all patient samples without regard to disease state, ethnicity, race (<i>sic</i>), religion, or sexual orientation.</p> <p>Medical Laboratory Professionals prevent and avoid conflicts of interest that undermine the best interests of patients.</p>	<p>The laboratory diagnostician, ... following the principles of reliability, honesty, impartiality ... should perform his professional activities with respect for the human person.</p> <p>... performs his professional activities with the utmost care and the awareness that the results of his work are used to protect human health and life.</p>	<p>Clinical laboratory practitioners are accountable for the quality and integrity of the laboratory services they provide.</p> <p>This obligation includes maintaining individual competence in judgement and performance and striving to safeguard the patient from incompetent or illegal practice by others.</p>
<i>Method</i>	<p>Medical Laboratory Professionals are accountable for the quality and integrity of the laboratory services they provide. This obligation includes maintaining the highest level of individual competence as patient needs change the limits of their level of practice.</p>	<p>A laboratory diagnostician, applying all his knowledge, skills and experience, strives to obtain reliable results of research and interprets them for the needs of practical medicine and science.</p> <p>A laboratory diagnostician in relations with other laboratory diagnosticians,</p>	<p>Clinical laboratory practitioners maintain high standards of practice. They exercise sound judgment in establishing, performing and evaluating laboratory testing.</p>

	<p>Medical Laboratory Professionals exercise sound judgment in all aspects of laboratory services they provide. Furthermore, Medical Laboratory Professionals safeguard patients from others' incompetent or illegal practice through identification and appropriate reporting of instances where the integrity and high quality of laboratory services have been breached.</p>	<p>in the case of noticing mistakes in their conduct, should pay due care first to the person concerned, or consult his supervisor. [But ...] A laboratory diagnostician in the presence of a patient does not assess the work of other diagnosticians, doctors and specialists involved in the treatment process.</p>	
<b>Practice</b>	<p>Medical Laboratory Professionals maintain strict confidentiality of patient information and test results. They safeguard the dignity and privacy of patients and provide accurate information to patients and other health care professionals. Medical Laboratory Professionals respect patients' rights to make decisions regarding their own medical care.</p>	<p>The laboratory diagnostician ... is obliged to keep secret everything he learned about the patient in connection with the conducted tests. ... The test results belong to the person they concern and can be made available only to that person or with his consent to other persons or institutions. He is also thus obliged to provide information from medical records to [nominated] third parties.</p>	<p>Clinical laboratory practitioners maintain strict confidentiality of patient information and test results and thereby safeguard the dignity and privacy of patients and any samples removed from them. They provide accurate reports about patients' results to other health care practitioners.</p>
<p><b>2. Duty to Colleagues and the Profession</b> <i>Focus</i></p>	<p>Medical Laboratory Professionals uphold the dignity and respect of the profession and maintain a reputation of honesty, integrity, competence, and reliability.</p>	<p>The laboratory diagnostician is obliged to build the ethos of his profession, to its promotion and development. Bearing in mind the importance of the</p>	<p>Clinical laboratory practitioners uphold and maintain the dignity and respect of our profession and strive to maintain a reputation of honesty, integrity and reliability.</p>

		<p>profession, the laboratory diagnostician performs his professional duties with a sense of responsibility for shaping impeccable attitudes in the professional environment of which he is an integral part.</p>	
<b>Method</b>	<p>Medical Laboratory Professionals ... contribute to the advancement of the profession by improving and disseminating the body of knowledge, adopting scientific advances that benefit the patient, maintaining high standards of practice and education, and seeking fair socioeconomic working conditions for members of the profession.</p> <p>... accept the responsibility to establish the qualifications for entry to the profession, to implement those qualifications through participation in licensing and certification programs, [and] to uphold those qualifications in hiring practices ...</p>	<p>Taking into account the dynamic development of laboratory medical diagnostics, the laboratory diagnostician should constantly expand his professional knowledge and improve his professional qualifications.</p>	<p>Clinical laboratory practitioners ... contribute to the advancement of the profession by improving the body of knowledge, adopting scientific advances that benefit the patient, maintaining high standards of practice and education, and seeking fair socio-economic working conditions for members of the profession.</p>
<b>Practice</b>	<p>Medical Laboratory Professionals establish cooperative, honest, and respectful working relationships within the clinical laboratory and</p>	<p>The laboratory diagnostician should share his knowledge with co-workers. [and] ... is obliged to motivate them to develop and facilitate</p>	<p>Clinical laboratory practitioners ... actively strive to establish cooperative and respectful working relationships with other health care</p>

	with all members of the healthcare team with the primary objective of ensuring a high standard of care for the patients they serve.	the improvement of qualifications. The Laboratory Diagnostician, as a teacher of the profession, should act as an example worth imitating and make every effort to ensure that the knowledge conveyed by him is up-to-date and corresponds to the principles of the profession.	practitioners with the primary objective of ensuring a high standard of care for the patients they serve. ... demonstrate honesty and integrity in business dealings with manufacturers, suppliers, competitors and customers.
<b>3. Duty to Society</b> <i>Focus</i>	As practitioners of an autonomous profession, Medical Laboratory Professionals have the responsibility to contribute from their sphere of professional competence to the general wellbeing of society.	<i>The laboratory diagnostician for society ...</i> should follow general standards of social coexistence, ...	As members of an autonomous profession, clinical laboratory practitioners have the responsibility to contribute from their sphere of professional competence to the general wellbeing of the community.
<i>Method</i>	Medical Laboratory Professionals comply with relevant laws and regulations pertaining to the practice of Clinical Laboratory Science and actively seek, to change those laws and regulations that do not meet the high standards of care and practice.	In relation to the patient, his family and the surroundings, the laboratory diagnostician pays due respect to, and observes the principles of, personal culture.	Clinical laboratory practitioners comply with relevant laws and regulations pertaining to the practice of clinical laboratory science and actively seek, within the dictates of their consciences, to change those which do not meet the high standards of care and practice to which the profession is committed.
<i>Practice</i>	Medical Laboratory Professionals serve as patient advocates. They apply their expertise to	The diagnostician performs laboratory tests with a view to obtaining a reliable result and cannot make	Clinical laboratory practitioners ensure scientifically appropriate, accurate and cost-effective

	improve patient healthcare outcomes by eliminating barriers to access to laboratory services and promoting equitable distribution of healthcare resources.	the service provided by him dependent on other circumstances including additional gratuities ... from people and institutions in any way interested in them.	application of health-care pathology service funding, guarding against waste, particularly clinical futility, inefficiency and needless investigative duplication.
--	--	--	--

### **Policies categorising the task by form of activity**

Here, the three selected codes were all originally published in English, those of the English Royal College of Pathologists [12], (which is a Code of Practice, incorporating ethical advice), the Canadian Society for Medical Laboratory Science [13], and from Australia, its Royal College of Pathologists of Australasia [14]. Tabulation was attempted, on the model above, but is patently impracticable. Each code lists many elements in common with the other two, and all also in common with matters dealt with in the first examined format, but each also cites many elements that are not readily discernible elsewhere; moreover, there is no obviously discernible pattern.

One important detail the Pathologists' Colleges specifically mention does also deserve specific consideration. The array of testing that has become available in recent years is vast by comparison with the menu laboratories offered 70 years ago, and inevitably as new, more precise and accurate, tests are offered there is a duty of care on the part of the laboratorian *vis à vis* the laboratory's clinician clientele to educate them about newly offered tests, thus to ensure that patients are best served by both.

The Australasian College has had a specific policy document addressing this need since 2004 [15], thus:

"Policy 3/2004: Ethical responsibility of pathologists in relation to test utility.

Specific Scenarios ...

*The test requested is inappropriate, not indicated or unnecessary:*

The pathologist may elect not to proceed with the test, in which case they may choose to contact the referrer personally or to include a qualifying note on the report ... [and] ... The medical practitioner may benefit from education on what would be a more appropriate test considering the clinical context."

In general, the Canadian Code, which is also supported by a Guidance Document [16], notes explicitly that the "...ethical principles contained herein are not listed in order of importance, but rather, should be considered in relation to each other during their application within situations involving ethical dilemmas." Specifically, however it does also mimic in text the exact tripartite focus seen above, thus (with numbering inserted).

"MLPs [medical laboratory professionals] shall practise ... for:

1. the protection and integrity of patients ...,
2. colleagues, health care providers, [and]
3. society, the environment and one's self."

### **CONCLUSIONS**

On balance, it seems that using the target of care as the primary sorting category when constructing an Ethics Code probably works best at a practical level. It also resonates with the

Belmont categorisations and may well have arisen therefrom; thus:

1. Respect for persons,  
*thus, the laboratorian's primary duty is to the patient*
2. Beneficence,  
*thus, the laboratorian will uphold the dignity and respect of the profession and maintain a reputation of honesty, integrity, competence, and reliability ..., and*
3. Justice  
*thus, practitioners have the responsibility to contribute from their sphere of professional competence to the general wellbeing of the community.*

To reiterate, whilst the Task Force cannot write documents for individual National Member Societies at the national level, it hopes that this guidance document, published also online on the IFCC website, will become a useful instrument in their “tool kit”. With it such member societies can construct an Ethics Code that is fit for purpose within their individual jurisdiction whilst at the same time preserving the essentials accepted worldwide as vital to such policies.



### **Acknowledgements**

RXD thanks the IFCC Ethics Taskforce Chair, Dr Nilda Fink, and our fellow TF – E members for their insightful and helpful review of the manuscript which has unequivocally improved it.

### **Disclosure**

The author, RXD, reports that he neither has, nor ever has had, any conflict of interest in relation to the research for and writing of this review. He received no funding from any source

for it, though the IFCC does reimburse him part of the cost of attending meetings of the IFCC's Ethics Task Force.



### **REFERENCES**

1. Sekadde-Kigonda CB, Higgins T, Bruns DE, Gronowski AM. Ethical considerations in clinical chemistry and laboratory medicine. Available at [http://www.ifcc.org/media/477698/07ifcc\\_tf\\_ethics\\_in\\_lab\\_medicine.pdf](http://www.ifcc.org/media/477698/07ifcc_tf_ethics_in_lab_medicine.pdf). Accessed, July 2019.
2. Nuremberg Code, The (1947). Textasevolved. Available at <https://history.nih.gov/research/downloads/nuremberg.pdf>. Accessed, July 2019.
3. World Medical Association (WMA). Declaration of Geneva. 1948, as amended by the 68th WMA General Assembly, October 2017. <https://www.wma.net/policies-post/wma-declaration-of-geneva/> Accessed, July 2019.
4. World Medical Association. World Medical Association Declaration of Helsinki: ethical principles for medical research involving human subjects. 2008. <https://www.wma.net/wp-content/uploads/2018/07/DoH-Oct2008.pdf>. Accessed, July 2019.
5. The National Commission for the Protection of Human Subjects of Biomedical and Behavioral Research. Ethical Principles and Guidelines for the Protection of Human Subjects of Research. <http://www.hhs.gov/ohrp/human-subjects/guidance/belmont.html>. Accessed, July 2019.
6. Burnett L, McQueen MJ, Jonsson JJ, Torricelli F. IFCC Position Paper: Report of the IFCC Taskforce on Ethics: Introduction and framework. *Clinical Chemistry and Laboratory Medicine* 2007; 45: 1098-104.
7. The International Organization for Standardization, ISO. <https://www.iso.org/standards.html>
8. The International Organization for Standardization, ISO. ISO 15189:2012, Medical laboratories -- Requirements for quality and competence. <https://www.iso.org/standard/56115.html>. Accessed, July 2019.
9. American Society for Clinical Laboratory Science. Code of Ethics. <https://www.ascls.org/about-us/code-of-ethics> Accessed, July 2019.
10. Poland National Chamber of Medical Laboratory Specialists. Code of Ethics. Krajowa Izba Diagnostów Laboratoryjnych, Kodeks Etyki Diagnosty Laboratoryjnego. <http://kidl.org.pl>. Accessed, July 2019.



11. Australasian Association of Clinical Biochemists. Code of Ethics. <https://www.aacb.asn.au/documents/item/49> Accessed, July 2019.

12. The Royal College of Pathologists. Code of practice for clinical biochemists/chemical pathologists and clinical biochemistry services. [www.rcpath.org](http://www.rcpath.org) Accessed, July 2019.

13. The Canadian Society for Medical Laboratory Science. Code of Ethics. <https://www.csmls.org/About-Us/Our-Members/Code-of-Ethics.aspx>. Accessed, July 2019.

14. The Royal College of Pathologists of Australasia. Code of Ethics. <https://www.rcpa.edu.au/getattachment/>

[6ed66de4-222a-44e4-9545-95a0f014ba0b/Code-of-Ethics.aspx](https://www.rcpa.edu.au/getattachment/6ed66de4-222a-44e4-9545-95a0f014ba0b/Code-of-Ethics.aspx). Accessed, July 2019.

15. The Royal College of Pathologists of Australasia. Policy 3/2004: Ethical responsibility of pathologists in relation to test utility. <https://www.rcpa.edu.au/getattachment/a3851604-2585-4a04-ab49-82a30b377b47/Ethical-Responsibility-Pathologists-Test-Utility.aspx> Accessed, July 2019.

16. The Canadian Society for Medical Laboratory Science. Code of Ethics. Guidance Document. [https://www.csmls.org/csmls/media/documents/Governance/Code\\_of\\_Ethics\\_Guidance\\_Document.pdf](https://www.csmls.org/csmls/media/documents/Governance/Code_of_Ethics_Guidance_Document.pdf). Accessed, July 2019.

# Ethics in laboratory medicine: perspectives and challenges in resource limited settings

Sudip Kumar Datta<sup>1,2</sup>

<sup>1</sup> Department of Lab Medicine, All India Institute of Medical Sciences, New Delhi, India

<sup>2</sup> On behalf of the IFCC Task Force on Ethics (TF-E)

---

## ARTICLE INFO

### **Corresponding author:**

Sudip Kumar Datta  
Assoc. Professor  
Dept. of Lab Medicine  
All India Institute of Medical Sciences  
New Delhi  
India  
E-mail: [dr.sudipdatta@gmail.com](mailto:dr.sudipdatta@gmail.com)

### **Key words:**

ethics, laboratory medicine,  
resource-limited, challenges,  
perspectives

---

## ABSTRACT

Currently diagnosis and management of patients in Clinical Practice is very much dependent on laboratory diagnostics. Laboratory Medicine, like any other branch of Medicine, is therefore, mandated with ethical usage of materials and data obtained from patients. Several countries, professional societies and the have developed policies and guidance materials on ethical issues related to laboratory medicine. However, ethical standards and practices vary between different cultures, geographies, legal architecture and according to available resources. In this article, we try to understand the challenges presented in terms of Ethics, where there are constraints of resources.

## INTRODUCTION

Like in any branch of medicine, which involves taking decisions about the wellbeing of individual patients as well as catering to the overall wellbeing of the society through continued learning through scientific observation and interventions on patients, Laboratory Medicine is also mandated with ethical usage of patient data and other materials for the optimum utilization of the same for benefit of the individual and the society. "Decisions about diagnosis, prognosis and treatment are frequently based on results and interpretations of laboratory tests. Irreversible harm may be caused by erroneous tests."

Medical Bio-Ethics developed and has evolved over the years starting from the Nuremberg Code in 1947 (1), the Declaration of Geneva in 1948 (2), through the Declaration of Helsinki in 1964 (3) to the Belmont report in 1978 (4). These documents mostly focus on medical research, however, the concepts in the Declaration of Geneva and the Belmont report are also applicable to the practice of clinical medicine. This is because, clinical medicine and medical research are complimentary to each other. This is especially true for laboratory tests, which are developed as a research tool one day goes on to become a diagnostic parameter very fast.

The core ethical principles of all these documents include: (i) Respect for persons, i.e. Acknowledgement of autonomy and protection of those with diminished autonomy; (ii) Beneficence, i.e. the duty to act in the best interests of patients or research subjects with the goal of maximizing benefits and minimizing harm (nonmaleficence); and (iii) Justice, i.e. the duty or obligation to treat patients equally and to distribute, by allocating fairly, what is rightly due in terms of benefits, risks and cost.

Like any other branch of Medicine, Laboratory medicine is obliged to adhere to high ethical standards. Many countries and professional

societies have developed policies and guidance materials on ethical issues related to laboratory medicine. The International Organization for Standardization (ISO) has created ISO 15189:2012 "Medical laboratories – Requirements for quality and competence" (5). Section 4.1.1.3 of the document summarizes the ethical conduct expected in laboratories. The core principles outlined in the document mention that (i) there should not be involvement in any activities that would diminish confidence in the laboratory's competence, impartiality, judgment or operational integrity; (ii) management and personnel are free from any undue commercial, financial, or other pressure and influences that may adversely affect the quality of work; (iii) where potential conflicts in competing interests exist, they shall be openly and appropriately declared; (iv) there are appropriate procedures to ensure that staff treat human samples, tissues or remains according to relevant legal requirements; (v) confidentiality of information is maintained.

Despite the importance of ethics in laboratory medicine, there is variability in education and training focused on laboratory ethics. Formal teaching of ethics is absent from many clinical chemistry and laboratory medicine training programs. Recognising this, need for training tools, especially, online ones to facilitate training of laboratory professionals with the convenience of location and timings, International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) has recently constituted a task force on ethics (TF-E) to streamline these documents and spread the ideas on ethics (6). The TF-E has come up with a toolkit for this purpose which serves as a repository of documents developed worldwide in the realm of laboratory ethics (7).

However, it is important to understand that ethical guidelines cannot be uniform over different cultures, geographies and legal architecture. Besides, adherence to the set principles differ

based on available resources and social practices. This article tries to focus on the challenges in setting guidelines and implementation of the same in resource limited settings.

### **CODES OF ETHICS AND ITS RELEVANCE IN LABORATORY MEDICINE**

A Code of Ethics may be described as an expression of basic values - the principles and standards by which we should conduct ourselves. Numerous laboratory professional organizations have developed codes of ethics, with common principles of conduct which act as guidelines to professional members of those organizations. The International Federation of Biomedical Laboratory Science (8) advises to maintain strict confidentiality of patient information and test results, safeguard the dignity and privacy of patients and above all be accountable for the quality and integrity of clinical laboratory services being provided.

On similar lines the American Society of Clinical Pathologists advise laboratory professionals to treat patients and colleagues with respect, care and thoughtfulness; perform duties in an accurate, precise, timely and responsible manner; and safeguard patient information as confidential, within the limits of the law.

As can be observed from above, most organizations and codes of ethics focus on several points while prescribing a guideline for Laboratory Medicine professionals. There are several areas in Laboratory Medicine practice where the formulation and implementation of ethical guidelines present challenges (9). These include: (i) Consent from patients including consent for unforeseen complications, usage of Leftover samples and biobanking; (ii) Considerations in genetic testing; (iii) Reporting implications in Incidental findings; (iv) Error disclosure; (v) Role of laboratories in Test utilization; (vi) Direct to consumer testing and (vii) Emerging diseases

setting. All of the above considerations relevant to Laboratory Medicine has been addressed elegantly in a recent article by Gronowski et al (10). In this paper we would focus on the challenges faced in resource limited settings.

Resource allocation is not uniform all over the world. Especially in developing countries, healthcare facilities have to work with several constraints. These may range from inadequate manpower, lack of training, less availability of latest equipment or methods, lack of adequate facilities for staff, and an ever-increasing load of patients. In these above-mentioned scenarios it is often difficult to conform to the highest standards of ethics.

#### ***I. Consent***

Most often the laboratories receive patient samples for testing. In such a setting obtaining consent for such exercise is the responsibility of the treating physician. In the hospital setting this is often 'implied', especially when the patient is admitted and sometimes not in a position to give consent. Hence, it is often a practice to take a blanket consent for such diagnostic tests which do not add significant risk to the patient. However, it is a good practice to take consents for such diagnostic procedures which might be adding significant risk to patient's life.

In resource limited settings which includes lack of manpower and time, the ideas of beneficence and non-maleficence should prevail. More importantly, in certain parts of the world, literacy and language issues may be a significant problem. Hence, implementation of a uniform ethics code presents a challenge. The problem sometimes escalates due to some unforeseen complications arising out of some diagnostic procedures and must be accounted for in the informed consent process. The Laboratory, thus, should be able to always abide by the ethical principles of

respect for persons, beneficence, even on a case to case basis.

In resource limited settings, handling of left-over samples becomes yet another challenge. Laboratories often facilitate add-on tests on these leftover samples to minimise turn-around time (TAT). However, the informed consent process must include provisions for the same and abide by the guiding principles of ethical codes. When using leftover samples for research, risk can be minimized by removing patient identifiers. Personal identifiers may be removed and replaced with a code i.e. deidentified or anonymized, i.e. identifiable information, if collected, identifiers are not retained and cannot be retrieved.

Biobanking, defined as a resource that holds human biological samples and/or data to facilitate research over time is also coming up in the developing countries in recent years, especially in the settings of emerging and exotic diseases. Often these are associated with storage of left-over samples in resource constrained parts of the world. However, the process of informed consent should abide by the general principles of ethics. In normal circumstances mostly two options are explored for such initiatives regarding consent: (i) recontacting patients and get consent for each new research study, which is logistically difficult, time-consuming, and expensive, and hence often practically not feasible in resource constrained settings; or (ii) allow patients to give a broad consent that allows for future use of the samples. However, the more general the consent becomes, the less informed it gets.

## II. Genetic testing

In principle, the 'right to autonomy' should allow people to decide whether genetic testing is to be performed or not. However, different governments have different policies regarding

'newborn screening', which is performed automatically, without physician orders. Once a disease or risk for disease is detected, patients and physicians face a dilemma whether to disclose the results of the tests to other family members who we now know to have increased risk. This may help individuals and the society as a whole device better preventive and therapeutic strategies and hence the principle of beneficence overrides the individual's right to autonomy. If the disease detected are treatable, the benefit to the public outweighs the autonomy of the individual.

## III. Incidental findings

These are results that have potential health or reproductive importance and are unintentionally discovered while processing for other tests. Incidental findings may be carefully evaluated of the benefits against the potential risks and may involve evaluating the result's accuracy, significance to health, and clinical actionability. In the resource limited settings in developing countries it often has other ramifications like cost of treatment and potential benefit of such treatment. Moreover, societal benefits must also be considered simultaneously before ruling in favour of patient's autonomy.

## IV. Error disclosure

Disclosure of errors in Laboratory Medicine setting comes with unique challenges related to error reporting because the laboratories usually have no relationship with the affected patient. Hence the disclosure has to happen through the treating counterparts. Several barriers to disclosing error exist viz. unclear definitions of error, fear that patients may not understand the error, worry that clinicians may not be able to properly explain the error, and disclosure of error that was actually committed by someone else. The process of disclosing medical errors is

gradually becoming formalized into the health care process.

Clinical laboratories should have policies and procedures for detecting errors that affect patient care and for informing both providers and patients. But several apprehensions exist among professionals inhibiting them to participate in the process. The most important amongst them is the fear of retribution from their colleagues, peers, supervisors, treating physicians and above all their patients. Sometimes there is inadequate understanding of the consequences of the error leads to the belief that eventual outcome would have been the same. The fear of being penalised because of disclosure is another barrier hence often people risk being caught. Fear of improperly conveyed error disclosure by physician colleagues also act as a barrier of appropriate error disclosure. Hence, appropriate mechanisms for error disclosure and mechanisms to protect the lab professional may increase the effectivity of error disclosure.

#### **V. Test utilization**

One of the major problems in resource limited settings is inappropriate test utilization. In developing countries like India healthcare system is run parallelly through government and private mechanisms. In the private setting patients generally pay from out of their pocket to meet the expenses incurred during their treatment etc. Only a small percentage of patients are covered under insurances. On the other hand in the government facilities the services are either free or at a subsidized rate. However, the waiting periods in those systems are long and often the ancillary facilities are inadequate, hence not preferred by people who can afford. Hence, inappropriate test utilization is a problem in both the scenario: in the private set-up unethical practices for profiteering might be discouraged by the labs; in government set up inappropriate test utilization should be discouraged as it leads

to mis-utilisation of public money. Laboratorians should advocate for proper test utilization and communicate with physicians when they feel testing has been ordered inappropriately.

Inappropriate laboratory testing can potentially lead also to false-positive results that can lead to unnecessary testing and intervention or even misdiagnosis, and increased costs for the patient and society as a whole. A lot of factors lead to poor test utilization: large and growing number of tests, lack of proper physician training, difficult to use direct order entry and electronic medical record systems, and demand from patients themselves as exposure to internet information leads patients to demand certain tests.

#### **VI. Direct-to-Consumer (DTC) testing**

DTC laboratory testing is growing rapidly all over the world along with the developing world. DTC allows consumers to order their own laboratory tests providing greater autonomy in some cases, is more accessible than going through standard healthcare providers and may be less expensive, which is also a source of justice for patients with limited financial means. However, it has several limitations; consumers are less likely to properly interpret their own laboratory tests and may find erroneous information without expert guidance. This becomes even more evident in low-prevalence disease which increases the chances for false positive results. Hence, although not directly under the purview of the laboratories ethically laboratories are bound to provide support to their customers.

#### **VII. Emerging disease setting**

The emergence of COVID-19 and some other novel diseases in recent years have presented a new challenge to ethical principles. A lot of questions have come up in these unusual circumstances like how the decisions are taken to ascertain which risks are acceptable for laboratory workers? Who decides what risks to

patients are acceptable to protect laboratory workers or to protect other patients? But most importantly in resource limited settings where there is shortage of appropriate personal protective equipment (PPE) creating awareness among lab staff about the level of PPE required for each lab activity. Importantly, there should be initiatives to spread awareness among staff to mitigate their apprehensions.

On the other hand laboratories should ensure access to laboratory testing for all patients who require testing. However, the low capacity of resource limited settings in analyzing samples through appropriate testing methods often leads to unethical practices. However, development of a policy in sync with legal requirements and local needs often addresses the issue.

### **OVERCOMING CHALLENGES**

Overcoming the above-mentioned challenges in Laboratory Medicine in resource limited settings is difficult, but not impossible. The most important step to ensuring ethical standards and practices in the laboratory must be recognised as a shared responsibility between all the laboratory staff. It is important that the roles of each of them are defined and all are made to understand the accountability associated with their jobs. This would be possible through repeated training of the staff at all levels.

#### ***Ethical issues in the pre-analytical phase***

The responsibility of the laboratory starts with proper identification of the patient or subject, collection of the appropriate sample using the appropriate technique, appropriate identification and labelling of the sample so that the right tests are performed and appropriate handling of the specimen until testing is performed. In the whole process respect for the persons must be maintained through obtaining proper consent: informed, implied. Besides, the right to

refuse to be tested, should be respected unless there are legal obligations, as has happened during the COVID-19 pandemic. Most importantly, confidentiality must be maintained at every step of the process including specimen transportation and data entry. Finally, the tests should benefit the patient based on the best medical evidence and should be done using universal precautions to protect the patient and the healthcare worker. And all these should be available at a reasonable cost to ensure access to the population as a whole.

#### ***Ethical issues in the analytical phase***

The most important issue during the analytical phase is to provide the best possible analytical results through good laboratory practice and maintenance of rigorous quality assurance program which becomes a challenge in resource limited settings. However, the guiding principle should be: “a wrong result is worse than no result”. Besides, all patient samples need to be treated equally. Discrimination based on gender, age, racial origin, or even socio-economic status is an injustice. However, specimens designated as STAT or priority must be analysed promptly to meet the medical need.

Another aspect of good laboratory practice (GLP), often ignored in the setting of resource-poor settings, is the refusal to analyse or report a result when there is evidence of: improper patient preparation, poor sample integrity, incorrect or poor labelling and other deficiencies that may compromise the test result. Lab staff are sometimes persuaded to accept ‘otherwise unacceptable samples’ due to patients coming in from remote areas with limited access to similar healthcare facilities in their vicinity. But this should be avoided.

Acceptability criteria of samples that are classified as “difficult to obtain” (such as cerebrospinal fluid) may be relaxed sometimes based on

clinical judgement; however the responsibility of the same is to be defined on persons with experience and laboratory should develop an appropriate policy on analysis. Besides, documentation of the specimens when specimen integrity or identification is compromised may be developed as a policy.

### **Ethical issues in the post analytical phase**

The post analytical phase poses certain other challenges in resource limited settings and includes reporting and interpretation of results, residual specimen storage, and data access.

Even in resource limited settings with limited manpower, identification of authorized personnel allowed to access medical records such as doctors, patients, and laboratory staff should be documented. This would ensure quality of reports as well as confidentiality of results. However, these policies should abide by legal requirements, insurance rules, and government regulations. Disclosure of errors if any needs to be notified as soon as they are identified, and test results should be corrected as soon as possible.

Maintaining confidentiality presents the biggest challenge in this phase. It is important to keep all client/patient information secure and restrict access to testing areas. In resource limited settings where records are not maintained electronically with appropriate access control, all physical documents need to be secured. Repeated reinforcements by training for maintenance of ethical standards need to be done because people often violate ethics not because they mean to, but because they are careless; sometimes even acting with good intentions.

The major managerial issues which needs to be addressed in all settings and is equally applicable in resource-poor settings is Conflict of Interest issues involved in procurement etc. of the lab. All work or travel-related payments

from a diagnostic or pharmaceutical company, or receipt of fees as a consultant, member of an advisory board, lecturer, speaker, or expert witness; grants, either financial or reagents, received from governmental sources, foundations, non-profit granting agencies, diagnostics or pharmaceutical companies come under the purview of the same and need to be disclosed during management review meetings.

### **CONCLUSION**

It can be well appreciated that ensuring adherence to ethical standards is a challenge in the resource limited settings. The challenges vary from place to place and the solutions need to be tailored to practical situations. Addressing these issues in the form of policy at the level of the country, local administration or even at the hospital/laboratory level may help in providing a guideline improving ethical practices. Framework for addressing ethical issues encountered in the practice of laboratory medicine need to be addressed and training of staff in this regard needs to be undertaken to ensure compliance to ethical requirements. We must constantly remind ourselves of the code of conducts and ensure we do the right thing because ethical issues are often hard to deal with because they create dilemmas.

### **REFERENCES**

1. The Nuremberg Code [Internet]. [cited 2020 Jun 27]. Available from: <http://www.cirp.org/library/ethics/nuremberg/>
2. WMA - The World Medical Association-Declaration of Geneva [Internet]. [cited 2020 Jun 27]. Available from: <https://www.wma.net/what-we-do/medical-ethics/declaration-of-geneva/>
3. WMA - The World Medical Association-Declaration of Helsinki [Internet]. [cited 2020 Jun 27]. Available from: <https://www.wma.net/what-we-do/medical-ethics/declaration-of-helsinki/>
4. Read the Belmont Report [Internet]. HHS.gov. 2018 [cited 2020 Jun 27]. Available from: <https://www.hhs.gov/>



[ohrp/regulations-and-policy/belmont-report/read-the-belmont-report/index.html](https://www.fda.gov/ohrp/regulations-and-policy/belmont-report/read-the-belmont-report/index.html)

5. ISO 15189:2012(en), Medical laboratories — Requirements for quality and competence [Internet]. [cited 2020 Jun 27]. Available from: <https://www.iso.org/obp/ui/#iso:std:iso:15189:ed-3:v2:en>

6. Task Force on Ethics (TF-E) - IFCC [Internet]. [cited 2020 Oct 7]. Available from: <https://www.ifcc.org/taskforce-ethics/>

7. TF-E Toolkit - IFCC [Internet]. [cited 2020 Oct 7]. Available from: <https://www.ifcc.org/taskforceethics/toolkit/>

8. Gallicchio VS, Gallicchio LM. Ethics and Quality Assurance in Biomedical Laboratory Science. 2013;(2):4.

9. ICMR Ethical Guidelines [Internet]. [cited 2020 Jun 29]. Available from: [https://ethics.ncdirindia.org/ICMR\\_Ethical\\_Guidelines.aspx](https://ethics.ncdirindia.org/ICMR_Ethical_Guidelines.aspx)

10. Gronowski AM, Budelier MM, Campbell SM. Ethics for Laboratory Medicine. Clin Chem. 2019 Aug 21.

# Research ethics committees in laboratory medicine

Lamis Beshir<sup>1,2</sup>

<sup>1</sup> Department of Clinical Immunology, Sudan Medical Specialization Board, Khartoum, Sudan

<sup>2</sup> On behalf of the IFCC Task Force on Ethics (TF-E)

---

## ARTICLE INFO

### **Corresponding author:**

Lamis Beshir  
Department of Clinical Immunology  
Sudan Medical Specialization Board  
Qasr Street, Khartoum  
Sudan  
E-mail: [Lamis.beshir@gmail.com](mailto:Lamis.beshir@gmail.com)

### **Key words:**

research ethics, research ethics  
committees, institutional review boards,  
laboratory medicine, biological samples,  
autonomy, informed consent, justice,  
beneficence, non-maleficence

---

## ABSTRACT

Biomedical research that involves human subjects requires compliance with ethical principles and guidelines. The ethical and scientific standards of research have been thoroughly discussed by international ethical guidelines and declarations. Compliance with these ensures the autonomy, dignity and well-being of research subjects; as well as the integrity and credibility of research results. Research ethics committees (RECs) are mandated to ensure that research proposals are scientifically sound and ethical. In this review, we define RECs in laboratory medicine and describe their role based on the examination of the requirements of ethical research; discuss particular ethical issues that arise in laboratory medicine research using biological samples, what challenges they face and how they can ensure the quality of their review. RECs need to be put into a broader framework that ensures institutional governance with continuous evaluation and auditing that ensure the quality of ethical review.

## INTRODUCTION

There has been a global increase in research productivity during the last decades. A potential concern with that is the adherence of these researches with ethical principles and the safeguarding of research participants. A balance between human subject protection and the progress of science should always be maintained.

Laboratory medicine like any other medical disciplines is bound to adhere to ethical standards in practice and research. Yet, there is still great variability in research ethics education in laboratory medicine programs (1). With advancements in the field and complexities arising in research, biomedical researchers and research ethics committees should be well trained to identify unique ethical issues that arise during the process of ethical review. Some of these ethical issues represent some new dimensions to old themes. Some of these include the use of biological samples that remain following routine investigation, sometimes using additional research tests in surplus of clinical requirements, storage of samples, research commercialization, methodology validation or methods comparison as well as incidental findings in genetic research, etc. A large proportion of the research in the field is retrospective where the conventional human subject is not directly involved. This poses an important question whether this type of research requires ethical approval and informed consent (2).

In this review, the role of research ethics committees (RECs) in ethical review, their operational function, particular issues arising for RECs in laboratory medicine along with their challenges and opportunities will be comprehensively discussed.

RECs or their equivalent, the institutional review boards (IRBs), are committees that provide protection to research subjects through their mandate of providing independent ethical and

scientific review of research proposals (3). They play a pivotal role in enhancing the quality of research conducted within educational and clinical institutions. They are also considered as a bridge between researchers, institutions, and ethical guidelines.

The primary mandate of RECs is to review research proposals before any data collection ensues. This process includes a rigorous scientific review and a detailed examination of ethical issues that may arise. This ensures research subjects are respected, autonomous and not exposed to excessive risks without direct benefits. Additionally, RECs have a secondary mandate of protecting the integrity of their research institution from any misconduct that may tarnish their reputation and result in public mistrust (4).

## THE ETHICAL FRAMEWORK FOR RESEARCH EVALUATION BY RECs

In 2000, Emanuel *et al* published a systematic framework consisting of seven general requirements that make human subject research ethical (5). This practical framework is a valuable tool to guide the review process conducted by RECs. We use this framework in relevance to laboratory medicine to discuss particular ethical issues. These requirements should be satisfied by research proposals before a REC grants final approval.

### 1. Social value

The submitted research and expected findings should lead to advancement in laboratory medicine knowledge.

### 2. Scientific validity

The research should be methodologically sound with clear scientific aims and objectives. It should not be biased, minimize confounders, and use the right analytical tests. Ethical research should be conducted in a rigorously sound methodological

approach. As stated in the CIOMS guidelines: “Scientifically unsound research on human subjects is *ipso facto* unethical in that it may expose subjects to risks or inconvenience to no purpose.”

The research methodology and data analysis must be valid, sound, and feasible. The methods used and sampling must be appropriate to achieve the research objectives. A research that is scientifically invalid will not enable the achievement of the overall research goal and therefore will expose subjects to unnecessary risks (6). Methodological scrutiny in laboratory medicine is of great importance. Different methodologies may exist to perform a single test; all of which differ in their sensitivity, specificity, positive and negative predictive values. Some tests may be quantitative, semi-quantitative while others are qualitative. Issues that arise in the pre-analytical, analytical and post-analytical phases should also be taken into consideration. It is therefore important to have the right expertise to scrutinize laboratory methods among REC membership.

### 3. Fair subject selection

The selection of enrolled subjects should be fair to ensure the principle of distributive justice is achieved (7)(8). It should ensure that no vulnerable populations are chosen without a justification. It should also ensure that inclusion and exclusion criteria are clear. Fair subject selection implies that, as much as possible, individuals who will bear the burdens and harms of the research should be able to enjoy its benefits and those who will benefit from research should share some of the anticipated risks. Retrospective research on biological samples is often anonymous or anonymized and therefore no issues arise from subject selection since the identity of samples cannot be readily ascertained. In a prospective study, the selection of a vulnerable population (e.g children) is not justified if the research can equally be conducted in adults.

### 4. Favorable risk-benefit ratio

Favorable risk-benefit ratio should ensure that there is an acceptable risk-benefit ratio and embodies the moral principles of beneficence and non-maleficence (7)(8). Beneficence implies that the benefits of research should be maximized as much as possible. In laboratory medicine, few increments above minimal risk are identified particularly in genetic research(9). Although researchers aim to ensure confidentiality of research subjects at all times, it is difficult in the era of genomic datasets and electronic health records to be certainly sure of that. Non-maleficence implies that harm should not be disproportionate to the benefits.

### 5. Informed consent

Informed consent is the application of the moral principle of respect for persons and autonomy (7)(8). It allows individuals to control their decisions and ensures that they make independent, informed decisions whether they want to be part of a research or not. To give informed consent, individuals must be informed about: the purpose of the research, its description, the anticipated risks, the potential benefits, the confidentiality of participants, any compensation for injury if applicable, and a reference person from the research team who should address any questions. It should be emphasized that participation is voluntary and withdrawal is possible without any negative consequences. The decision-making should be free of coercion, undue influence, or pressure. Participants should be competent and have adequate understanding of the information. Surrogate or proxy consent should be obtained in the case of incompetent individuals. Research in laboratory medicine does not always need informed consent; this is the case in anonymous and anonymized biological samples. In general, coded and prospective samples require informed consent since the risks of privacy and confidentiality are present.

## 6. Independent review

In laboratory medicine, research stakeholders have different interests which may differ from those of the participants and thus conflict of interests (COI) may arise. There are often collaborations between industry and academia. Some REC members may also have COI such as being consultants or own shares in biomedical companies. Maintaining the independence of RECs review is vital to research governance and public accountability. This is usually achieved by having clear COI policies. CIOMS require that RECs members should not review research in which they have competing direct interests as investigators or funders (6). Including a lay person or a public representative as a REC member contributes to the independence of review.

## 7. Respect for human subjects

Research subjects should be respected throughout the research process. Their privacy and confidentiality should always be maintained. Any new information that arises should be made available and disclosed to the participants. Permitting subjects to withdraw and change their mind during the research is key to achieving autonomy and ensuring the welfare of subjects is always respected.

## RECs: STRUCTURE, ETHICAL ISSUES AND CHALLENGES

RECs were originally established to protect the health, safety and wellbeing of researchers and of research participants. Over time, their role has been expanded and diversified beyond the ethical review to rather become a role of research governance. Previous comments have described them as “gatekeepers or “adjudicators” (4). Whittaker suggests that: “ethical review boards have become established as one of the most authoritative, if not authoritarian, gatekeepers in research history” (10). For these

reasons, it is important to ensure RECs have the right membership.

Different institutions have different membership for their ethical committees. Albeit, there is a consensus that the membership is generally multidisciplinary with broad representation from across specialties.

Additionally, a member of the community that represents its values and norms in the committee, contribute to the independent review and RECs’ efforts to maintain transparency and accountability to the public (11). He/she may also reflect the public’s nonscientific point of view and opinions; and ensures that the informed consent is comprehensible to the nonscientist research person.

There is no specific guidance on the membership of RECs in laboratory medicine but it is agreed that whenever expert advice is needed for complex proposals then the appropriate consultants (eg. Clinical chemist, immunologist, geneticist, hematologist, etc) may be called upon by the REC (Table 1).

## ETHICAL ISSUES IN RESEARCH USING BIOLOGICAL SAMPLES

### *Blood samples and the human subject*

RECs evaluating proposals in laboratory medicine often encounter complex ethical issues that differ from their clinical counterpart mainly due to the nature of research that uses blood samples and human tissues (figure 1). These samples and data are not the living, identifiable humans that research regulations were designed to protect. Many ethical debates discussed whether research on blood samples is human subject research. Federal regulations define a human subject as a “living individual about whom an individual conducting research obtains data through intervention or interaction with that individual or identifiable private information” (14). Albeit,

**Table 1** Regulatory requirement of RECs

Regulation	Requirement
Membership	<ul style="list-style-type: none"> <li>• At least 5 members of varying backgrounds with equal gender opportunities</li> <li>• At least 1 scientific member, 1 nonscientific member, and 1 unaffiliated member</li> <li>• The chair should not be a member of the institution</li> <li>• Members should have adequate experience and expertise to safeguard subjects' rights and welfare. Expertise should include research methodology, ethics and laws, regulations, institutional commitments, and professional standards; as well as content expertise in the different fields of biomedical research. Avoid selection bias</li> <li>• At least 1 member knowledgeable about research in vulnerable groups</li> <li>• Members should declare conflicts of interest</li> <li>• Ad hoc experts or independent consultants invited as needed</li> </ul>
Function	<ul style="list-style-type: none"> <li>• Quorum requirement (more than half members); with distribution of expertise requirements over the quorum</li> <li>• a confidentiality agreement regarding meetings, applications, information on research participants, and related issues should be signed by members;</li> </ul>
Review	<ul style="list-style-type: none"> <li>• Meetings should be planned in advance according to the workload allowing time to study submitted documents beforehand</li> <li>• Approve, ask for corrections and disapprove research</li> <li>• Follow a systematic scientific and ethical framework for review</li> <li>• Approve informed consent and ensure suitability</li> <li>• Waive the requirement for informed consent whereas applicable</li> </ul>
Documentation & archiving	<ul style="list-style-type: none"> <li>• All meeting minutes of RECs</li> <li>• Submitted protocols, corrections, approval and disapproval decisions</li> <li>• Correspondence between REC members; and applicants</li> <li>• Follow up</li> <li>• Final reports of studies</li> </ul>

*The table summarizes the operational guidelines of RECs including the membership terms, function, review, documentation and archiving in accordance to the World Health Organization guidelines and the Common Federal Rule (12) (13) (14).*

a lot of biomedical research is carried out on deceased people or on biological materials donated before death. Although these are not living subjects but the protection of the interests of the deceased is increasingly recognized as respect for persons ethical principle (7)(15).

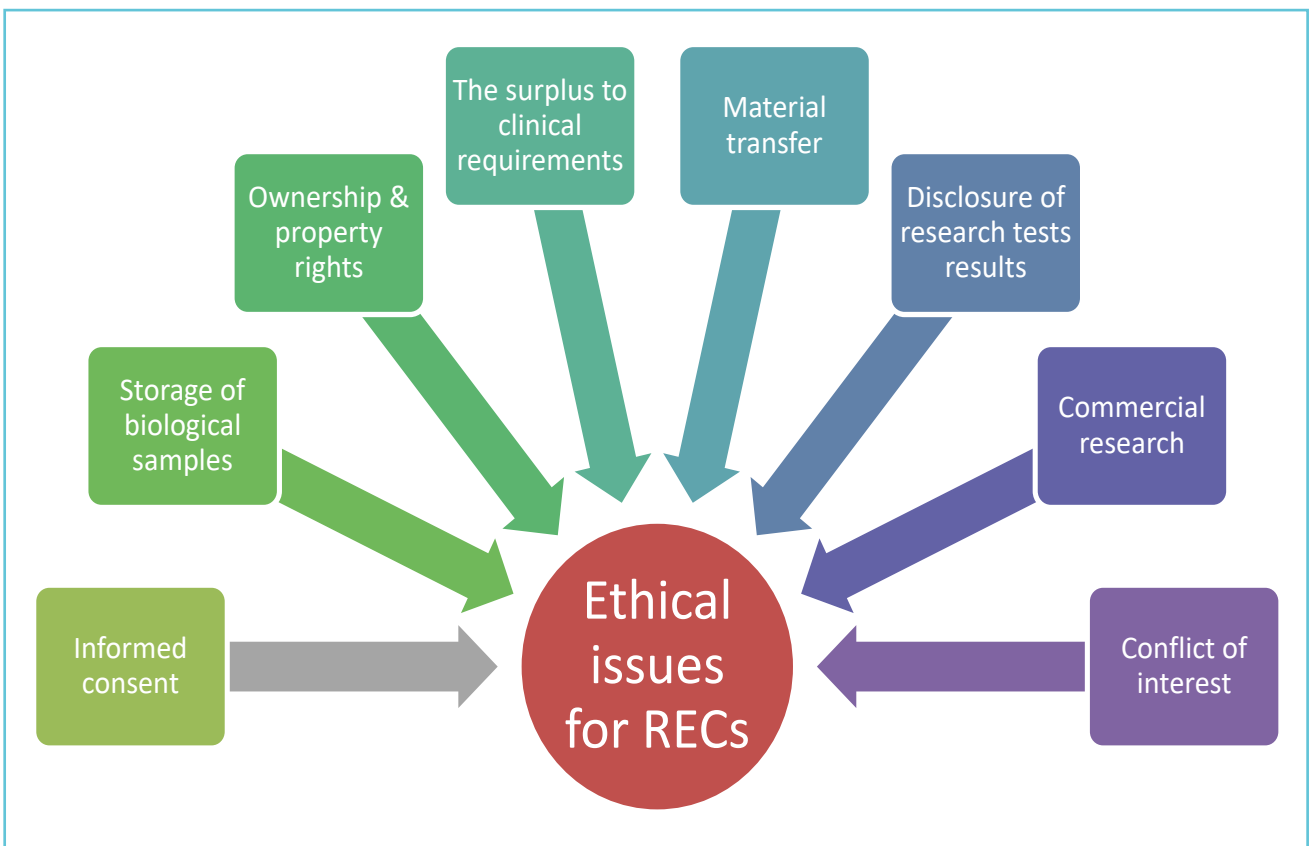
### Informed consent

Research in laboratory medicine commonly involves existing samples in which the participants did not give consent to, but such research may be quite valuable. What counts as informed consent when a sample may be stored for years and used for unforeseen research? The traditional concept of informed consent where a participant is informed about the important aspects of a study (purpose, risks and benefits) may not

be a good fit for research with bodily materials and data stored for future purposes. In many instances, a broad consent has been suggested to overcome such hurdles. Yet, it is still argued that a broad consent is not bona fide consent and thus, the concept of autonomy is not really fulfilled. Obtaining consent under institutional status quo may not necessarily be a consent that ensures autonomy of research subjects but is rather safe for regulatory purposes.

RECs should thoroughly discuss what would be the best model of informed consent to be used in such cases. Issues to be discussed within the informed consent include the storage of samples for future research and whether samples may be part of a repository or a biobank (Figure 1).

**Figure 1** The ethical issues encountered by research ethics committees in laboratory medicine



*Material transportation, ownership and commercialization.*

Research in laboratory medicine may sometimes require the transfer of biological sample to different institutions or even countries in international collaborations. Material transfer agreement is a process to facilitate exchange of samples and technology between researchers and institutions and to protect the interest of both (16). It is usually an agreement on materials that are owned by the originator but of which has no propriety rights or patents (17) (18). Although this can be a simple process, more complexities are gained with the increment in collaborations between industries and academia. There has also been an increasing movement in universities to commercialize their research (18). Biotechnology research has also been changing and moving towards genomics and generation of *in vitro* research models, nucleic acid tools, molecular probes for drug discovery and other tools that are meant to be disseminated (19).

Many disputes have arisen over the ownership of leftover “abandoned” blood samples. There are no specific regulations to govern this issue. However, the interest of research participants should always be safeguarded. Many bioethicists

consider subjects to no longer have any property or ownership rights over the material (20). This is because leftover materials are no longer functional. Some participants have expressed even if the donor has no continuing property right, the laboratory must act in accordance with ethical regulations if this material is to be used for research (21). It is for RECs to ensure that the confidentiality of the data is kept by researchers all time.

Commercialization of research and commercial spin-off companies may automatically imply a COI for RECs which may not always be the case in research. For instance, a researcher who is developing a diagnostic method may not acquire enough funding to develop such tool. However, biotechnology companies may be interested to sponsor such type of research. This collaborative partnership could result in a synergistic relationship that may lead to the development of new knowledge. It is therefore important for RECs to understand such complex entities and ensure that benefits are not skewed towards companies nor researchers are driven by competing interests (Table 2).

**Table 2** General guidelines for researchers in laboratory medicine before conducting research on human tissue specimens

1.	Understand the regulations in reference to biological samples and laboratory medicine.
2.	Understand the components of informed consent form in biological samples research (specially in genetics).
3.	Inform research participants as much as possible about risks, how their specimens will be used now and in the future, plans to return incidental findings.
4.	Have research protocols and informed consent forms reviewed and approved by a REC.
5.	Always maintain confidentiality.
6.	Understand when a waiver of informed consent can be obtained by REC.



## CHALLENGES

RECs have been criticized generally for a number of issues. The administrative work of RECs has been previously described as a slow bureaucratic process (22)(20) (23). It is also costly and serves little to enhance the quality of the research process (24) (25). There have been suggestions to reassess RECs to ensure their purpose is fulfilled to encourage research within acceptable ethical frameworks. However, there are still no metric tools that could assess and measure the effectiveness of RECs (26) (13).

Sometimes, inflexible requirements for adherence to narrow literal interpretations of regulations and other policies have led to a system that is more concerned with “legal” protection of the institution than the protection of human research participants (18) (27). Some challenges that face RECs are the inconsistencies across different committees even though they may be using the same guidelines (28). However, Edwards *et al* argue that not all inconsistencies should be perceived negatively and may sometimes be considered a desirable part of research (29). This possibly has its origin from a moral pluralism philosophy. However, it is the inconsistencies that are due to the lack of expertise in identifying ethical issues that is undesirable.

In addition to scientific and ethical review, it is crucial for RECs to ensure researchers have sufficient research experience and qualifications or alternatively collaborating with an experienced colleague in the relevant field of research. In the case of laboratory medicine, the researcher needs to provide evidence to the committee that they commit to good laboratory practice, they are trained in laboratory health and safety rules, and they are experienced in using laboratory equipment and techniques. Researchers should ensure they are using the right laboratory method in line with their research objectives. Working with biohazardous materials, with toxic

chemicals or with radioisotopes is risky and must be governed by the bioethical principle of non-maleficence (7) (8), the REC have a duty to minimize the risk of individuals being exposed to harm. Qualifications that ensure the investigators have achieved these competencies or received formal training should be confirmed by RECs in laboratory medicine.

## OPPORTUNITIES AND THE WAY FORWARD

The identified challenges faced by RECs point towards an opportunity for quality assurance and continuous improvement (13). Independent auditing is key to a quality assessment that can be followed by accreditation (30) (31). For these purposes, few tools have been previously developed and many local guidelines may include guidelines for accreditation of RECs (18).

RECs maybe unexpectedly be faced with the increasing workload. It is imperative for those committees particularly in laboratory medicine to have well trained members that can efficiently review protocols in due time (32). It is increasingly recognized that adequate training of committee members improves the efficiency of RECs (33). This has been recognized by Levine who emphasizes the need to add an educational system for REC staff and members followed by an accreditation system for RECs and certification system for the staff (31). As the face of biomedical research is changing and gaining complexities, RECs members will need to undergo more formal continuous professional training. Enhancing a model of self-assessment, certification of members and accreditation are all strategies that may be used to ensure the professional research review (22).

## REFERENCES

1. Bruns DE, Burtis CA, Gronowski AM, McQueen MJ, Newman A, Jonsson JJ, et al. Variability of ethics education in laboratory medicine training programs: results of an international survey. *Clin Chim Acta* [Internet]. 2015;442:115–8.

- Available from: <http://europepmc.org/abstract/MED/25437910>
2. Borovecki A, Mlinaric A, Horvat M, Supak Smolicic V. Informed consent and ethics committee approval in laboratory medicine. *Biochem medica* [Internet]. 2018 Oct 15;28(3):30201. Available from: <https://pubmed.ncbi.nlm.nih.gov/30429665>
  3. McCarthy C, Emanuel E, Grady C, Crouch R, Lie R, Miller F, et al. *The Oxford Textbook of Clinical Research Ethics*. 2008;
  4. Ekberg M. Reassessing the Role of the Biomedical Research Ethics Committee. *J Acad Ethics* [Internet]. 2012;10(4):335–52. Available from: <https://doi.org/10.1007/s10805-012-9171-6>
  5. Emanuel EJ, Wendler D, Grady C. What Makes Clinical Research Ethical? *JAMA* [Internet]. 2000 May 24;283(20):2701–11. Available from: <https://doi.org/10.1001/jama.283.20.2701>
  6. Council for International Organizations of Medical Sciences. & WHO (CIOMS). *International ethical guidelines for biomedical research involving human subjects*. Geneva; 2016.
  7. US Department of Health and Human Services. *The Belmont report* [Internet]. 1979. Available from: [http://www.hhs.gov/ohrp/human\\_subjects/guidance/belmont.html](http://www.hhs.gov/ohrp/human_subjects/guidance/belmont.html)
  8. Beauchamp TL, Childress JF. *Principles of biomedical ethics*. Oxford University Press, USA; 2001.
  9. McGuire AL, Fisher R, Cusenza P, Hudson K, Rothstein MA, McGraw D, et al. Confidentiality, privacy, and security of genetic and genomic test information in electronic health records: points to consider. *Genet Med* [Internet]. 2008;10(7):495–9. Available from: <https://doi.org/10.1097/GIM.0b013e31817a8aaa>
  10. Whittaker E. Adjudicating entitlements: the emerging discourses of research ethics boards. *Health*. 2005;9(4):513–35.
  11. UNESCO. *Bioethics Committees at work: Policies and Procedures*. Available from: [www.unesco.org](http://www.unesco.org)
  12. Team WHOPR and D. *Operational guidelines for ethics committees that review biomedical research* [Internet]. Geneva PP - Geneva: World Health Organization; Available from: <https://apps.who.int/iris/handle/10665/66429>
  13. Grady C. Institutional Review Boards: Purpose and Challenges. *Chest* [Internet]. 2015 Nov;148(5):1148–55. Available from: <https://pubmed.ncbi.nlm.nih.gov/26042632>
  14. PublicWelfarePofHS.45CFR§46[Internet].2005.Available from: <https://www.hhs.gov/ohrp/regulations-and-policy/regulations/45-cfr-46/index.html>
  15. Beauchamp TL. Informed consent: its history, meaning, and present challenges. *Cambridge Q Healthc Ethics*. 2011;20(4):515–23.
  16. Bubela T, Guebert J, Mishra A. Use and misuse of material transfer agreements: lessons in proportionality from research, repositories, and litigation. *PLoS Biol*. 2015;13(2).
  17. Rodriguez V. Material transfer agreements: open science vs. proprietary claims. *Nat Biotechnol*. 2005;23(4):489–91.
  18. Mirowski P. Livin'with the MTA. *Minerva*. 2008;46(3):317–42.
  19. Kahl L, Molloy J, Patron N, Matthewman C, Haseloff J, Grewal D, et al. Opening options for material transfer. *Nat Biotechnol* [Internet]. 2018;36(10):923–7. Available from: <https://doi.org/10.1038/nbt.4263>
  20. Allen G. Getting beyond form filling: the role of institutional governance in human research ethics. *J Acad Ethics*. 2008;6(2):105.
  21. Petrini C. Ethical and legal considerations regarding the ownership and commercial use of human biological materials and their derivatives. *J Blood Med* [Internet]. 2012/08/07. 2012;3:87–96. Available from: <https://pubmed.ncbi.nlm.nih.gov/22977316>
  22. Shaul RZ. Reviewing the reviewers: the vague accountability of research ethics committees. *Crit Care*. 2002;6(2):121.
  23. Loff B, Black J. Research ethics committees: what is their contribution? *Med J Aust*. 2004;181(8):440.
  24. Sugarman J, Getz K, Speckman JL, Byrne MM, Gerson J, Emanuel EJ. The cost of institutional review boards in academic medical centers. *N Engl J Med*. 2005;352(17):1825–7.
  25. Wagner TH, Murray C, Goldberg J, Adler JM, Abrams J. Costs and benefits of the national cancer institute central institutional review board. *J Clin Oncol*. 2010;28(4):662.
  26. Grady C. Do IRBs protect human research participants? *Jama*. 2010;304(10):1122–3.
  27. Kotsis S V, Chung KC. Institutional review boards: what's old? What's new? What needs to change? *Plast Reconstr Surg* [Internet]. 2014 Feb;133(2):439–45. Available from: <https://pubmed.ncbi.nlm.nih.gov/24469174>
  28. Abbott L, Grady C. A systematic review of the empirical literature evaluating IRBs: What we know and what we still need to learn. *J Empir Res Hum Res Ethics*. 2011;6(1):3–19.

29. Edwards SJL, Ashcroft R, Kirchin S. Research ethics committees: differences and moral judgement. *Bioethics*. 2004;18(5):408–27.

30. Cummins D. The Professional Status of Bioethics Consultation. *Theor Med Bioeth* [Internet]. 2002;23(1):19–43. Available from: <https://doi.org/10.1023/A:1019567812747>

31. Levine RJ. Institutional review boards: a crisis in confidence. *Ann Intern Med*. 2001;134(2):161–3.

32. Gronowski AM, Budelier MM, Campbell SM. Ethics for Laboratory Medicine. *Clin Chem* [Internet]. 2019 Dec 1;65(12):1497–507. Available from: <https://doi.org/10.1373/clinchem.2019.306670>

33. Organization WH. Research ethics committees: basic concepts for capacity-building [Internet]. Geneva PP-Geneva: World Health Organization; Available from: <https://apps.who.int/iris/handle/10665/44108>

# Conflicts of interest and an approach to managing them

Nilda E. Fink<sup>1,2</sup>

<sup>1</sup> PROES Program, Biochemical Foundation of Argentina

<sup>2</sup> On behalf of the IFCC Task Force on Ethics (TF-E)

---

## ARTICLE INFO

### **Corresponding author:**

Dr. Nilda E. Fink  
Chair, IFCC Task Force on Ethics (TF-E)  
Director, Programa PROES  
Fundación Bioquímica Argentina  
Viamonte 1167  
Buenos Aires C 1053  
Argentina  
E-mail: [NFink@fbpba.org.ar](mailto:NFink@fbpba.org.ar)

### **Key words:**

conflict of interest, disclosure,  
management, ethics

---

## ABSTRACT

Conflicts of interest (COI) exist in every step of life, including in medicine and science. Professionals who work in different areas of Health systems, such as physicians in care patient, in pharmaceutical and biomedical devices industries, in management positions, in teaching or in research, all must apply rigid ethical principles.

It is possible with these actions that COI were detected in several circumstances such as in the prescribing therapy, in production or employment of technology in services of Health system, in article publications, and in decision-making for those who have decided to occupy positions of responsibility in scientific and healthcare institutions, in industry or professional associations, among others. These actions must be consistent with the essential principles of Bioethics.

At present, COI disclosure has been irreversibly installed in Medicine. A detailed description of the classification of conflicts of interest and its ethical and legal implications in the practice of health sciences such as those that appear in the practice of clinical and laboratory medicine, pharmaceutical industry and also,

research and publications are included. Final considerations on the management of COI are also included.



Conflicts of interest (COI) exist in every step of life and in every corner of society, including medicine and science (1, 2). Professionals who work in different areas of Health systems, such as physicians in patient care, in pharmaceutical and biomedical devices industries, in management positions, in teaching or in research, must apply rigid principles even if their main objective is not to produce new knowledge as researchers are interested in. It is possible with these actions that COI were detected in several circumstances such as in the act of prescribing a therapy (3), in production or employment of technology in services of Health system, in article publications, and in decision-making for those who have decided to occupy positions of responsibility in scientific and healthcare institutions, in industry or professional associations (4- 6) among others. These actions must be consistent with the essential principles of Bioethics in the field of Health that are implicit in documents and fundamental principles such as those arising from the Helsinki declaration, the Belmont report and others (7-10). This is extended to modern and serious societies where justice and social ethics require absolute transparency in decision-making involving third parties (11).

When the term COI is analyzed in a search database such as Pubmed (12), it appears registered – as part of the title – between 1962 and 2018 there are 1,288 registered articles, with a sustained increase. It was noted though that it obtained more relevance in the field of medicine from the 80s. This shows that COI consideration has been irreversibly installed. In this article several aspects pertaining to conflict of

interest, in terms of its definition, classification, applications, management and other challenges are described.

## DEFINITION OF THE CONCEPT

Sometimes the term is used unclearly or used as a prejudice that leads to anticipated moral condemnation. It is necessary to have a more flexible look since the presence of a conflict of interest is a situation that appears frequently and sometimes it is not possible to avoid that other people were involved. Therefore, biased conclusions can be drawn to qualify a behavior as reprehensible when in facts it is not. So it is essential to review and adjust its meaning. According to the Thompson definition (13), COI “is a set of conditions in which professional judgment concerning a primary interest tends to be unduly influenced by a secondary interest.” It can also be defined as a situation where a judgment or action that should be determined by a primary value established for professional or ethical reasons (protection of research subjects, production of safe knowledge, adequate assistance to the patient), may be influenced or appear skewed to obtain a secondary benefit.

For example, a person in a high professional position is in a COI when the decision he/she has to take may affect his/her personal interests, of an economic or professional nature, as his/her decision could attract benefit or harm to those interests. For a better understanding of the nature of personal interests, Table 1 enumerates an ordered description of different types of factors involved besides own interests (relatives, affective and professional relations).

The term conflict of interest is a moral concept that means a challenge to the behavior of those who have an obligation or a duty that collides with a personal interest. Such interest can therefore distort a judgment in an irrational or unacceptable way, thus creating a mantle of

suspicion if justice has been damaged (11,13). In other words, it is the person's actions in the context of a particular situation that may be a cause for concern (14).

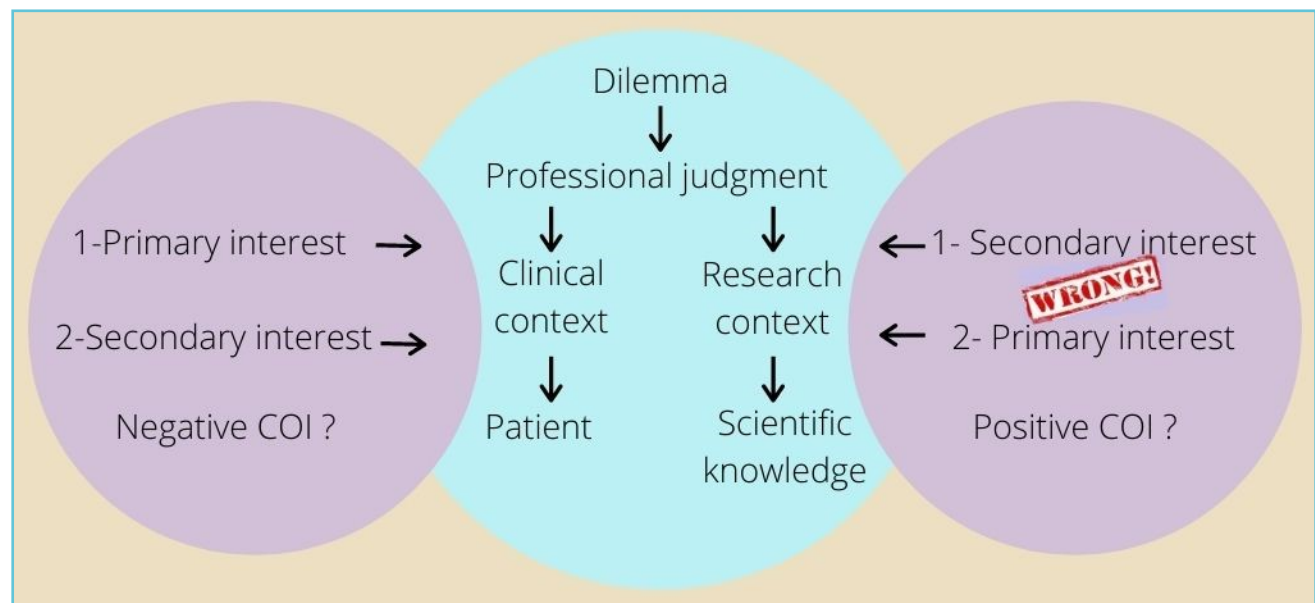
Accordingly, the ethical dilemma that the presentation of a COI entails has more to do on

how to proceed and resolve the conflict since the appearance of a COI does not imply by itself reprehensible conduct or taking a reprehensible position by the professional involved. The challenge is the management and resolution of the dilemma (Figure 1).

**Table 1** Classification of personal interests

a)	Own interests.
b)	Family interests, including close relatives or persons with whom the professional lives in a relationship of affectivity and relatives within the fourth degree of consanguinity or second degree of affinity.
c)	Interests of persons with whom the professional has a dispute pending.
d)	Interests of persons with whom they have close friendship or manifest enmity.
e)	Interests of legal persons or private entities with whom the professional has been related by an employment or professional relationship of any kind in previous years.
f)	Interests of legal persons or private entities to which professional relatives mentioned in point (b) are related by an employment or professional relationship of any kind, provided that it implies management, advisory or administration functions.

**Figure 1** Scheme for decision making to solve dilemmas



**DIFFERENTIAL CHARACTERISTICS OF THE CONCEPT**

At this point it is convenient to start by asking ourselves what we are talking about in professional terms or in a colloquial environment when we say COI since it is possible to give it a meaning that is not appropriate. A particular situation may arise when there is a dispute over “conflicting interests” between two or more persons or entities, for example in a fight between two or more manufacturers when a contract is in dispute. In this case, the genesis of the conflict may appear if the institutional evaluator who must give his opinion on proposals quality has held a position in one of the companies that aspires to obtain the contract.

There would be a real conflict of interest due to the hypothetical possibility that this evaluator could make a biased opinion that unjustifiably benefits one of the companies. Different situations can occur such as receiving important gifts as an exchange for those benefits or receiving a hidden payment, for example to favor one of the companies, thus criminal figures of bribery is produced. Another possibility of conflict for a professional who works as chief staff in an industry or in a public administration department who could have a “conflict of duties”. As an

example this could appear when an employee requests to be absent in his/her labor by a non-contemplated statutory cause, although important from personal point of view, which may be understandable within the framework of a labor relationship. That person, exercising his/her work with professionalism, must decide between the obligation to respond to their superiors who trust on his/her actions to allow, only for valid reasons, the withdrawal of a person. On the other hand, to consider that the reason for the request is not appropriate although understandable from other points of view (14).

Another condition that is in fact a conflict of interest appears when a health professional gives preferential treatment to a family member on a waiting list. A different situation that can generate conceptual confusion occurs when a physician is under the effect of an emotional involvement with a patient and does not handle the situation with sufficient professional distance, which can lead to failures associated with a loss of objectivity. These arguments have led to the existence of codes of ethics that specifically prohibit medical professionals, especially in the field of psychiatry, to engage in personal relationships with patients. Table 2 summarizes these concepts and in Table 3 detailed types of secondary interests are described.

**Table 2** Decision-making conflicts in Medicine

Type of conflict	Classification	Comments
Conflicts of interest	Due to different types of interests <ul style="list-style-type: none"> <li>• Economic</li> <li>• Non-economic</li> <li>• Related to third parties interests</li> </ul>	To protect moral integrity at decision-making challenges
Other conflicts		
Conflicting interests	-	Commercial arguments

Bribery	-	Criminal behavior. There is ethical and legal conflict
Conflict of duties	-	Social responsibility to guarantee the administration regulations compliance

**Table 3** Categories of secondary interests

Class	Comments
Direct sponsored	Payment for conferences, articles writing, patient registrations for clinical studies , for research of a product
Indirected sponsored	Gifts , travels, meals
Laboral and academic development	For prestige, academic acknowledgment or promotion
Others	Physician in charge of patients care and also as researcher, and/or as administrator of Health resources and/or as consultant for marketing strategies.

**ETHICAL AND LEGAL IMPORTANCE OF CONFLICTS OF INTEREST IN MEDICINE AND HEALTH SCIENCES**

Since 1978 when the Belmont report (10) was produced, basic fundamental principles of justice, autonomy and beneficence/non-maleficence support, among other aspects, the decision-making on patients’ own body. This position completely relegated the paternalistic model of physician-patients relationships. Nevertheless, some very unprotected and needy sectors request assistance, fully trusting on suitability and integrity of physicians without perceiving that they may be in a disadvantageous situation in which COIs could occurs. Harmful decisions can be made, biased towards defenseless people, so these COIs must be treated more rigorously than in other settings in medical fields.

In relation to COI in medical research, it is well known that there are research studies where results have been reported in a biased way as

a consequence of COI, these effects may prove harmful and may be reversed only after a considerably long time. In relation to teaching and in recommendations elaboration, deviations can occur due to COI that installs concepts that later are transferred for years to clinical practice and that persist over time. Likewise, there are doubts in society about the integrity of health professionals who lose credibility in front of public opinion. This loss of credibility is due to the mantle of doubt that is installed before the eyes of patients who may consider that gifts, invitations or other types of benefits can affect their health and finally, their lives. It is difficult to prevent all negative effects of COI as Chren et al mentioned (15) “preserving justice, the trusteeship relationship with our patients, and our own altruism are regulative ideals — that is, standards not always achievable by all of us, but useful templates ‘against which all efforts can be measured.”



From a legal point of view, the responsibility for the administration done by anybody who acts for or on behalf of another person has limits on his/her autonomy and freedom as there is a more powerful party entrusted to protect interests of a less powerful party. This could be similar to physician/patient relationship so any possibility of COI should be avoided to preserve the interests of the protected part. In order to regulate health care professionals, specific codes for various professionals have been approved. These codes for example prohibit members of a health care professional for practicing the profession if they are in a COI or in a professional misconduct (16).

Many professions have their own COI legal documents. These documents were composed quite some years ago, but for medicine it was not until about 40 years ago that legal bodies were incorporated. Many medical organizations and journals were concerned about interactions between the industry and the medical profession and have introduced requirements for authors to disclose any financial interest they have in a study (16). Moreover, different types of articles written by authors with a financial interest were rejected. For example a very complete and excellent disclosure form was approved by American Association of Clinical Chemistry (AACC) (17).

In the 1990s, many Colleges and Associations of Physicians from several countries outlined in position papers how to deal with relations between physicians and the industry, and it was acknowledged that not only real bias but also perceived bias should be avoided (18). Moreover, gifts or subsidies from industry “ought not to be accepted if acceptance might influence or appear to others to influence the objectivity of clinical judgment”. Also detailed recommendations on gifts and subsidies can be found in an opinion of the Council on Ethical and Judicial Affairs, which the American Medical Association

(AMA) incorporated into its Code of Medical Ethics (18,19). Sections on continuing education, research, clinical evaluation and surveillance studies were included, nonetheless, it was remarked that the main focus on patient care must be included. The guidelines do not forbidden research and education sponsoring by industry but only under regulated nature of manufacturers’ and physicians’ alliances. In relation to this, conference and courses organizers must have under control the content of events and no reference for endorsement of a sponsor’s product should be done. As regards to industry gifts, some associations guidelines are stricter. Furthermore, health professionals are discouraged from investing in companies where commercial success could be affected by practices of the professional. Criticism has frequently emerged and proposal for improvement have been sought (16).

In 2012, the International Federation of Clinical Chemistry (IFCC) approved a statement for all members integrating its structure at different levels that they are required to read this policy and sign the disclosure statement (20). As part of the statement definition, conditions for participant’s exclusion, examples of potential COI and disclosure statement were included. At the same time also a statement for Ethics publication was also approved. Also Council of International Medical Societies (CIOMS) in its Ethical Guidelines for clinical research in COI guideline 25 dedicated to researchers, research institutions and research ethics committee mention, as an important topic for disclosure, requirements for education and highlight recommendations on disclosure to research ethics committees and to participants (21).

The World Medical Association produced a statement on conflict of interest (22) with recommendations for research, needs in education and also items for health organization and Institution conflicts were included. Two other aspects of interest for laboratory medicine are included.

One is related to self-referrals and fee-splitting as a point to keep in consideration and expresses that “all referrals and prescriptions (whether for specific goods or services) should be based on an objective assessment of the quality of the service or of the physician to whom the patient has been referred. Referral by physicians to health care facilities (such as laboratories) where they do not engage in professional activities but in which they have a financial interest is called self-referral. This practice has the potential to significantly influence clinical decision-making and is not generally considered acceptable unless there is a need in that particular community for the facility and other ownership is not a possibility (for example, in small rural communities)”. Also mentions that “kickbacks (or fee-splitting) occur when a physician receives financial benefit for referring a patient to a specific practitioner or for a specific service for which a fee is charged. This practice is not acceptable”. Second important point considers for “patient convenience, occurs if many physician offices are located in close geographic proximity to other medical services such as laboratories. The physician should not receive any financial compensation or other consideration either for referring a patient to these services, or for being located in close geographical proximity to them” (22).

Some authors have researched the extent to which physicians interact with industry. Considering this goal there are many different types of articles published from 70s up to now referred to COI, industry and medical specialties and organizations in which frequency, relevance and implications of gifts from companies to researchers and others members in academic life, such as scientists, and found that these studies indicate that interactions are common and in various forms. Unfortunately studies have shown these interactions influence physicians and medical researchers for example on prescribing patterns (23), and the outcome of research studies

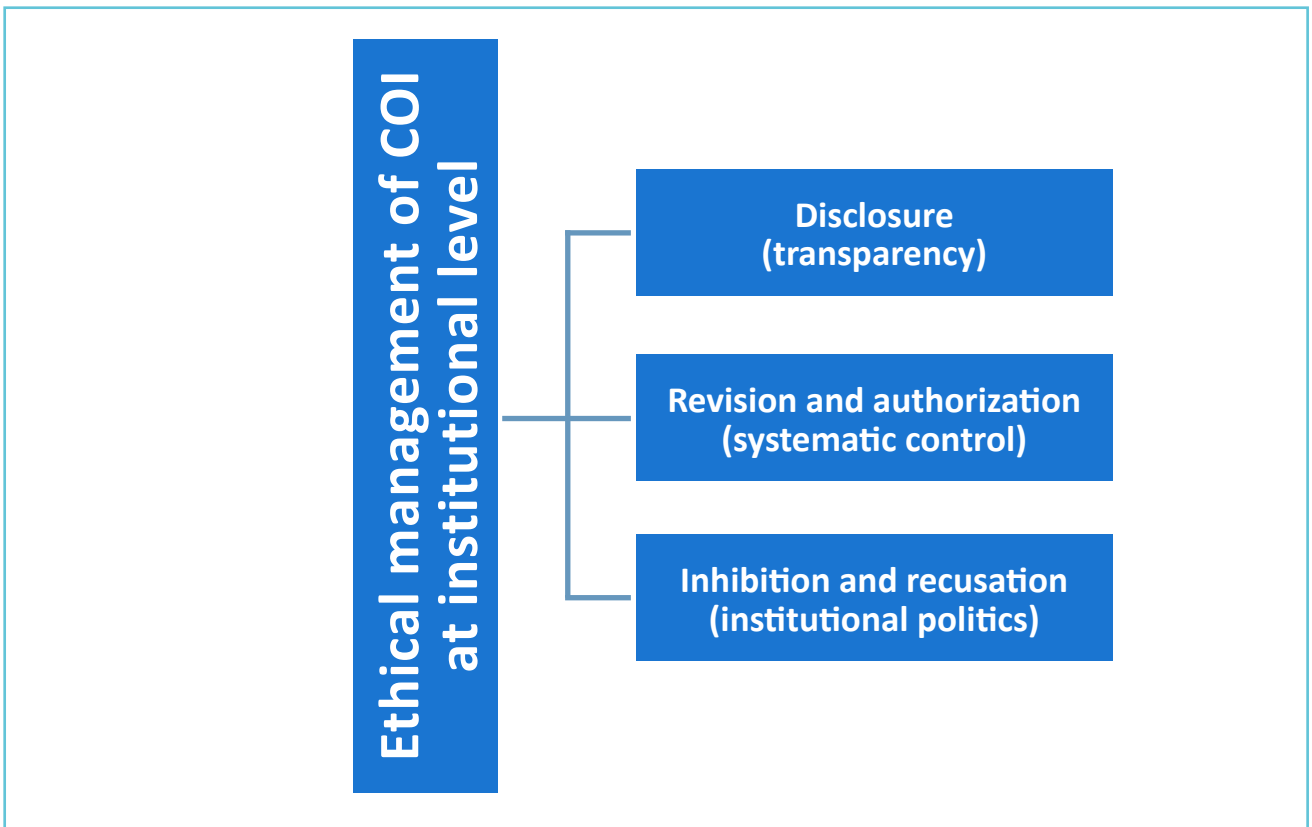
to support a product. These findings did not show something unexpected and are in parallel with the budget spent for products promotion. Even taking into account these finding, many physicians consider that they are not likely to be influenced by their interactions with industry.

In relation to publication ethics, focus on duplicate publication, inappropriate authorship, fabrication of data, plagiarism, and conflict of interest (COI) is paramount. The issue of COI is an important problem for medical journals. Relevant documents related to COI definition and disclosure include the COI Disclosure Form are those done by the International Committee of Medical Journal Editors (ICMJE) (24) and flow-charts by the Committee on Publication Ethics (COPE) dealing with the omission of essential COI notes in research publications (25), which provide advice for publishers and editors.

#### **AN APPROACH TO MANAGEMENT OF CONFLICTS OF INTEREST**

As was mentioned earlier there are conflicts of interest in every aspect of human activity, including medicine and science. The moral problem arising when the influence of a secondary interest that can threaten the ethics of a professional decision is accepted as natural. A conflict of interest could be an inducement or temptation that must be distinguished from its acceptance. However, there are situations that are consciously ignored. Awareness, acknowledgement of COI and evaluation of the influence of secondary interests are important in these situations. As such, focus must be directed to recognize and manage COI appropriately. The ethical management of COIs by institutions must be carried out through explicit regulations with corporative supervision through their governing bodies or by special committees. The types of conflict and the potential for real or perceived damage can be approached by different

Figure 2 Strategy for an approach to management of conflicts of interest



strategies and they are: disclosure, hierarchical steps of review and authorization, and prohibition (Figure 2) (16).

### **Disclosure**

The main action to solve a conflict of interest is its disclosure. Transparency by public declaration is considered as the golden rule. It is morally very healthy to highlight the secondary interests that could affect the rigor of professional judgment because it implies to show potential bias intentionally desired to be prevented by offering their exposure openly. To judge whether one is in a conflict of interest, it can be revealing to ask a recommended question in relation to how comfortable patients and others people are about his/her interest in the matter under discussion and in function of answers.

### **Review and authorization**

By means of laws and regulations, formal review systems were introduced to control conflict of interest additional to disclosure as is the case of medical research. Research ethics boards have a duty to determine, among other items, whether conflicts of interests are affecting clinical trials and the health care of patients participating in trials. Members of review boards should themselves not be in a conflict of interest. Universities have systems of authorization and reception of financial interests from researchers to the university administration and that could verify if the required conditions are achieved (16).

### **Prohibition**

But disclosure and review and authorization are not always enough and adequate. Some con-

flicts of interest may so deeply affect trust that they have to be forbidden (16). The policies of some entities disapprove of researchers being remunerated over and above reasonable compensation for extra work and in a condition of decrease of other income (16). The remuneration for merely including research subjects in a clinical trial, should not be accepted. In that case, the temptation to include subjects without proper informed consent and without respecting selection criteria is huge. The policy further discourages physicians from accepting a fee from industry in exchange for meetings with representatives or for attending promotional activities. The organizers of educational program events are also requested not to be in a potential COI by virtue of any relationship with companies that fund such events. Additionally, to ensure quality of clinical practice guidelines, organizations need to formulate policies related to COI (26).

In conclusion, it is important to arise awareness for COI acknowledgment to evaluate the influence of secondary interests. In this article we include a detailed description of conflicts of interest classification and its ethical and legal implications in the practice of health sciences such as those that appear in the practice of clinical and laboratory medicine, pharmaceutical industry and also, research and publications. Final considerations on the management of COI are also included.

## REFERENCES

1. Lo B, Field MJ, Eds. Committee on Conflict of Interest in Medical Research, Education, and Practice. Board on Health Sciences Policy (consensus report). Washington: The National Academies Press; 2009.
2. Muth CC. Conflict of interest in Medicine. JAMA. 2017; 317:1812.
3. Grande D. Limiting the influence of pharmaceutical industry gifts on physicians: Self-regulation or government intervention? J Gen Intern Med. 2010;25:79-83.
4. Fabbri A, Gregoraci G, Tedesco D, Ferreti F, Giraldi F, Lemmi D, et al. Conflict of interest between professional medical societies and industry: A cross-sectional study of Italian medical societies' websites. BMJ Open. 2016;6: e011124.
5. Ngo-Metzger Q, Moyer V, Grossman D, Ebell M, Woo M, Miller T, et al. Conflicts of interest in clinical guidelines: Update of U.S. Preventive Services Task Force Policies and Procedures. Am J Prev Med. 2018;54(1S1):S70-80.
6. Nissen SE. Conflicts of interest and professional medical associations: Progress and remaining challenges. JAMA. 2017;317:1737-8.
7. World Medical Association (WMA). Declaration of Geneva. Available at <https://www.wma.net/policies-post/wma-declaration-of-geneva/>. Accessed 9 July 2019.
8. World Medical Association (WMA). Declaration of Helsinki: ethical principles for medical research involving human subjects. 2008. Available at <https://www.wma.net/wp-content/uploads/2018/07/DoH-Oct2008.pdf>. Accessed 9 January 2016.
9. The Nuremberg Code (1947). Text as evolved. Available at <https://history.nih.gov/research/downloads/nuremberg.pdf>. Accessed July 2019.
10. National Commission for the Protection of Human Subjects of Biomedical and Behavioral Research [USA]. The Belmont Report. Ethical Principles and Guidelines for the Protection of Human Subjects of Research. 1979. Available at <https://www.hhs.gov/ohrp/regulations-and-policy/belmont-report/read-the-belmont-report/index.html>. Accessed 15 March 2016
11. Morreim EH. Conflict of Interest. En: Reich WT, editor. Encyclopedia of Bioethics. New York: Simon & Schuster Macmillan; 1995. p. 459-65.
12. PubMed. <https://pubmed.ncbi.nlm.nih.gov>. Accessed 31 August 2020
13. Thompson DF. Understanding financial conflicts of interest. N Engl J Med. 1993;329:573-6.
14. Brody H. Clarifying Conflict of Interest. The American Journal of Bioethics. 2011;11:23-8.
15. Chren MM, Landefeld CS, Murray TH. Doctors, drug companies, and gifts. JAMA 1989;262(24):3448-51.
16. Lemmens T, Singer PA. Conflict of interest in research, education and patient care. CMAJ. 1998;159:960-5.
17. American Association for Clinical Chemistry (AACC). Policy #2-11: Conflict-of-Interest Disclosure Statement and Copyright Assignment. Available at [https://www.aacc.org/~media/files/about-aacc/governance/aacc-policies/2\\_11\\_conflict\\_of\\_interest\\_disclosure\\_statement\\_and\\_copyright\\_assignment.pdf?la=en](https://www.aacc.org/~media/files/about-aacc/governance/aacc-policies/2_11_conflict_of_interest_disclosure_statement_and_copyright_assignment.pdf?la=en)
18. American College of Physicians. Physicians and the pharmaceutical industry. Ann Intern Med 1990;112:624-6.

19. McMurray RJ, Clarke OW, Barrasso JA, MD; Clohan DB; Epps CH Jr, John Glasson J, et al. Gifts to physicians from the industry. *JAMA* 1991;265:501.

20. International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) Conflict of Interest Policy and Disclosure Statement 2012. Available at [https://www.ifcc.org/media/158532/Conflict of Interest Statement Mar 2012.pdf](https://www.ifcc.org/media/158532/Conflict%20of%20Interest%20Statement%20Mar%202012.pdf)

21. Council for International Organizations of Medical Sciences (CIOMS) International Ethical Guidelines for Health-related Research Involving Humans. Fourth Edition. Geneva. Council for International Organizations of Medical Sciences (CIOMS); 2016

22. World Medical Association. WMA statement on conflict of interest. 2015. Available at <https://www.wma.net/policies-post/wma-statement-on-conflict-of-interest/#:~:text=A%20conflict%20of%20interest%20is,influenced%20by%20a%20secondary%20interest.&text=Other%20interests%20must%20not%20be,the%20potential%20to%20do%20so>

23. Fickweiler F, Fickweiler W, Urbach E. Interactions between physicians and the pharmaceutical industry generally and sales representatives specifically and their association with physicians' attitudes and prescribing habits: a systematic review *BMJ Open*. 2017; 7(9): e016408.

24. International Committee of Medical Journal Editors (ICMJE). Recommendations for the Conduct, Reporting, Editing, and Publication of Scholarly Work in Medical Journals. 2019. Available at <http://www.icmje.org/icmje-recommendations.pdf>. Accessed December 2018

25. Committee on Publication Ethics (COPE). What to do if a reviewer suspects undisclosed conflict of interest (Col) in a submitted manuscript. Available at <https://publicationethics.org/files/COI%20submitted.pdf>

26. Norris SL, Holmer HK, Burda BU, Ogden LA, Fu R (2012) Conflict of Interest Policies for Organizations Producing a Large Number of Clinical Practice Guidelines. *PLoS ONE* 7(5): e37413.

# A framework to ethically approach incidental findings in genetic research

Lamis Beshir<sup>1,2</sup>

<sup>1</sup> Department of Clinical Immunology, Sudan Medical Specialization Board, Khartoum, Sudan

<sup>2</sup> On behalf of the IFCC Task Force on Ethics (TF-E)

---

## ARTICLE INFO

### **Corresponding author:**

Lamis Beshir  
Department of Clinical Immunology  
Sudan Medical Specialization Board  
Qasr Street, Khartoum  
Sudan  
E-mail: [Lamis.beshir@gmail.com](mailto:Lamis.beshir@gmail.com)

### **Key words:**

genetic research, genomics, research ethics,  
incidental findings, unsolicited findings

---

## ABSTRACT

With the advancement of science in the area of genetics and genomics, special ethical considerations should be taken in addition to the general ethical framework followed in research.

Genetic research can reveal information about the susceptibility of an individual to disease and hence about his/her future health. Such information may be of interest and benefit to research participants, especially if preventive strategies exist. It may also expose them to other risks or anxieties when incidental findings that were not the primary scope of the study are found. Ethical guidelines acknowledge the duty of researchers to disclose incidental findings (IFs) to participants. In this review, we recommend four steps approach that researchers can use to disclose incidental findings: plan for IFs, discuss IFs in informed consent, identify and disclose IFs. Verification and identification of IFs should follow a categorical stratification based on the importance of the findings and the presence of a beneficial intervention to the participants.

## INTRODUCTION

Genetic testing is “a (laboratory) procedure to detect the presence or absence of, or change in, a particular gene or chromosome, including an indirect test for a gene product or another specific metabolite that is primarily indicative of a specific genetic change”(1). It is the analysis of human DNA, RNA, genes and/or chromosomes, or the analysis of human proteins or certain metabolites, with the primary purpose of detecting a heritable genotype, gene mutation, phenotype or karyotype. Next generation sequencing (NGS) has revolutionized clinical genomics by enabling the detection of large genetic variations in patients. This new type of advanced DNA analysis may fundamentally alter medicine and be used as genetic health screening tool. The knowledge of genomic information may allow healthy individuals to explore their susceptibility to certain gene disorders. Consequently, this knowledge provides an interventional opportunity for screening programs, prevention and personalized medicine.

Genetic research should not be conducted with the primary aim to provide research subjects with specific medical information about their genetic status or overall wellbeing. However, if there is a possibility that the research may yield incidental findings of significance to their health, prior to the research, the participant should be informed of this possibility and offered the choice of whether he or she would like to receive such information.

An incidental finding (IF) can be defined as a finding concerning an individual research participant that has potential health or reproductive importance and is discovered in the course of conducting research but is beyond the aims of the study (2).

The enhanced capacity, rapid pace of NGS technology advances along with the falling costs of the test allow new ethical and psychosocial

questions to be asked. Since the information contained in genome sequencing is vast, diagnostic NGS may not only provide information about the genetic basis of disease, but potentially further IFs of certain or uncertain significance. Unsolicited information can be generated from sequencing and there is still an active debate about which information should be disclosed to the patient.

In the first part of this paper, we examine the characteristics of genetic testing that pose additional risks and ethical issues compared to other types of research. The vulnerabilities caused by these risks imply that researchers have ancillary obligations towards their participants as they disclose IFs. We also discuss genotype/phenotype correlation to point towards the importance of interpreting IFs and their significance suggesting guidance from experts and clinical geneticists should be sought whenever required. Lastly, we examine the current guidelines and ethical debates and suggest a framework to deal with IFs.

## CHARACTERISTICS OF GENETIC/GENOMIC TESTING

**Table 1** The characteristics of genetic information

- Personal
- Permanent
- Predictive, pre-symptomatic
- Prejudicial
- Pedigree-sensitive

The unique characteristics of genetic/genomic testing gives rise to special ethical issues that have been increasingly identified and discussed by international guidelines (3). They should be taken holistically in risk-benefits analysis and in guiding informed consent.

### 1. Personal

Genetic data is unique to each individual. Genetic variability between individuals is identified as .11% of 3.2 billion bases of nucleotides (4). Genetic databases can therefore be used to match individuals based on a small set of single nucleotide polymorphisms (5).

Most regulations have emphasized some additional safeguards to protect such type of information but the extent to how successful they are remains unclear (6).

### 2. Permanent

Individual genomes are immutable that is, they do not change throughout the person's life. Albeit, some somatic mutations that affect the DNA maybe acquired resulting in some alteration of some parts of the DNA. Therefore, researchers should consider that the genetic results are long lasting and have lifelong considerations when disclosed to the participants.

### 3. Predictive, pre-symptomatic

Some genetic testing may have predictive values in disease development. The accuracy in interpreting such predictability is a complex and critical subject. Therefore, careful interpretation of the results should be made by experts.

### 4. Prejudicial

Genetic testing may reveal private information that raises concerns of stigmatization and discrimination. Breach of confidentiality could result in financial risks such as loss of employment or insurance (7). Researchers, in particular epidemiologists, may wish to study genes in populations to determine their contribution to disease incidence and prevalence in the community. This information may result in social stigmatization of certain populations or ethnic groups such as the case of the Havasupai tribe (8).

### 5. Pedigree-sensitive

Genetic testing has the potential to reveal information about family members as in germline mutations. A lot of information may be revealed about individuals who have not consented to have their genetic material tested in the first instance.

It is widely known that individuals who carry a certain disease-causing mutation or a genotype may not exhibit all the pathological features or phenotype. This is a phenomenon known as reduced penetrance. This might be explained by allelic expression, modifier genes, digenic inheritance, imprinting, the influence of age, sex and environmental variants and epigenetics on gene expression and post-translational modification (9) (10). The type of mutation (e.g. missense, nonsense, frameshift, or deletion) would have an impact on the exhibited phenotype. In monogenic disorders, the presenting phenotype depends on the presence of a mutation in another gene. Somatic mosaicism also results in phenotypic variations. There is also a difference between loss of function and gain of function mutations. For instance, distinct mutations of STAT3 at the same position may cause either loss of function or gain of function. Loss of function may results in hyperimmunoglobulin E syndrome also known as Job's syndrome while gain of function may results in early onset lymphoproliferation, autoimmunity and myelodysplastic syndromes (11)(12)(13). In some instances, complete penetrance may require the presence of mutation variants at other loci.

Autosomal recessive conditions are known to have reduced penetrance with varying clinical implications since depending on the function of the second allele. Autosomal dominant conditions were previously thought to be penetrant. However, it is now increasingly described that the alleles that are not completely



penetrant may act in a recessive fashion such as PKD1 alleles in renal manifestations (14) (15). Penetrance should be distinguished from the expressivity. Expressivity refers to the phenotypical variations among the same genotype (10). Although used in an inter-related manner, making a distinction between the two phenomena is important when informing research subjects regarding what the results imply.

Although a discussion of genotype/phenotype correlation is not the purpose of this article, it is important to highlight the importance of understanding individual results in genetic research in the presence of such uncertainties. A careful interpretation of such genetic variants should be made and discussed with geneticists prior to reaching a conclusion. Sometimes, giving a black or white answer to whether a specific phenotype will be manifested as a result of a certain mutation is not always feasible. It is also of importance to highlight these inevitable risks and uncertainties within the process of informed consent.

## CURRENT GUIDELINE RECOMMENDATIONS

Incidental findings are becoming the most pressing issue in genetic research today and are increasingly recognized by guidelines and research ethics committees. The issue raises a fundamental question regarding whether researchers have an ethical obligation to disclose incidental or unsolicited findings to participants, guidelines recognize the need to do so. The National Bioethics Advisory Commission (NBAC) recommended that when the risks identified in the study are both valid and associated with a proven intervention for risk reduction, disclosure may be appropriate (17). The Council for International Organizations of Medical Sciences (CIOMS) also recommends that a prior informed plan on how to manage unsolicited findings

should be disclosed to research participants “a procedure for determining whether unsolicited findings should be disclosed, and if so, how they should be managed; how the quality of the material is controlled; how confidentiality of the link between biological specimens and personal identifiers is maintained” (3). The National Heart, Lung, and Blood Institute conditions the return of genetic research results on a significant risk of disease (specific relative risk >2.0). The disease should have fatal or debilitating morbidity or reproductive implications, and the availability of therapeutic or preventive interventions (16).

These established ethical justifications to disclose IFs have not identified a specific implementation approach. Given the characteristics of genetic information and the implications of results in relevance to genotype/ phenotype correlation, it is important to be careful in determining the significance and validity of tests prior to interpretation.

## A FRAMEWORK TO DISCLOSE IFs

### 1. Planning for IFs

The possibility of obtaining IFs should be included in the study plan. The plan should include a procedure to verify IFs, interpret and evaluate their implications taking into consideration genotype, phenotype correlation, and reproductive importance. The plan should explain the possibility of IFs whether foreseeable or not in their informed consent and include the intent of researchers to disclose these findings should they arise.

There should be a process to ensure data validity and quality to avoid any false positives that may result from data mistakes. Research ethics committees (RECs) should review this plan and ensure it is satisfactory to minimize risks and safeguard participants and their autonomy.

## 2. Discussing IFs in the process of informed consent

Research ethics guidelines consider informed consent a cornerstone document in maintaining the autonomy of research participants. The informed consent should explain the genetic test that will be conducted in research and the characteristic of the data produced from it. This lays the foundation to discuss and explain the potential IFs whether anticipated or unanticipated. IFs may result in psychological, social, and financial risks. The prejudicial nature described earlier recognizes risks of stigmatization, loss of employment or insurance as a result of disclosure. Researchers should allow subjects to ask questions, verify any queries and understand the scope of the research and resultant risks in order to make an informed decision.

## 3. Verifying and identifying IFs as they arise

An old generic framework by Reilly suggests that investigators should differentiate three categories of findings in research: those “of such potential importance ... that they *must* be disclosed immediately”; those that “are of importance to subjects ... but about which ... [the investigator] should exercise judgment” on disclosure; and those “that do *not* require special disclosure.” (17). This article similarly recommends a three categories stratification framework in which the investigator initially carries an assessment and interpretation of the findings after consulting with genetic experts – should this be of need – to determine the implication of a genetic variant on an individual’s health. Currently, many analysts place only findings of health importance in the “should return” category, even though individuals may assign high importance to findings with major reproductive implications. Although, most recommendations to date have conditioned “should return” on the “actionability” of findings, this remains ambiguous. It is under

debate whether the utility of findings should be viewed from the standpoint of clinicians, the standpoint of individual participants, or some combination (17) (18). For instance, in the case of identifying a colorectal cancer gene, actionability from the standpoint of a physician is undertaking yearly colonoscopy, while from a participant’s standpoint it could mean making different choices in life.

## 4. Disclosure of IFs

A compassionate subject-centered approach should be used to disclose IFs. The plan to disclose should take into consideration the autonomy of individuals and their preferences as well as the ancillary care obligations of researchers. Current models perhaps stratify IFs to inform the appropriate decision of action but it is still debatable what kind of action should be taken exactly. What is it that is owed to the participants by researchers? To what extent are researchers obliged to minimize the risks resulting from the vulnerabilities created from IFs, even though they have not been created by them.

Offering genetic counseling with disclosure contributes to risk minimization. The OHRP IRB Guidebook states that “[a]ppropriate counseling should be provided to educate subjects about the meaning of the genetic information they have received, and to assist them in coping with any psychosocial effects of participation.” (18).

There has been a debate whether researchers have the duty to provide further clinical work-up and care as a result of these IFs. Researchers may also be requested to share information with the participant’s treating physician, which should not be denied (20).

This positive moral duty in the disclosure of IFs has been emphasized by ancillary care frameworks discussed by Beskow et al (19). The ancillary approach supports the notion the researchers and

RECs should take into consideration the degree of entrustment in genetic research that individuals put into researchers that creates participants' vulnerabilities. This vulnerability becomes more complex when it is combined with medical, financial and social vulnerabilities. Therefore, they are ought to contribute to addressing the consequences of IFs. Such ancillary obligation needs to be considered when planning and budgeting research.

Research differs from clinical care that it is conducted with the motivation to advance generalizable knowledge rather than create individualistic benefits. Despite the pluralistic nature of the research enterprise, it is advocated that researchers are motivated by some fiduciary obligations to maintain public entrustment especially in the context of genomic research and biobanking.

## OTHER ISSUES

### *Incorporating participants' preferences to guide the decision of disclosure*

An outstanding question is whether patients' preferences should be considered in the decision-making process regarding the disclosure of IFs. Although guidelines have emphasized on the principle of respect for persons and the exercise of autonomy (20)(21), it remains unclear how to incorporate that. Just like physicians who tend to overestimate potential benefits of testing over the unanticipated harms, researchers are not an exception. Similarly, research subjects may have an underestimation of the risks arising from disclosure. This may possibly be due to the lack of information regarding the nature of genetic testing and the disease implication of discovered mutations or genetic variants. If a participant states that they do not wish to have IFs returned, and their results come back with potentially important information such as the presence of BRCA1/BRCA2 gene,

does the researcher have a "duty to rescue"? Will ignoring the person's autonomy driven by beneficence be considered undue paternalism? It is best to use a planning approach that considers both participants' preferences and current recommendations from ethics guidelines. Emphasis should be put on the process of informed consent where researchers take the time to provide information, resources and examples of the foreseeable and unforeseeable risks associated with IFs.

It should be reiterated that research ethics have evolved into a model of collaborative partnership between researchers and research subjects that are now increasingly referred to as participants to imply this collaborative nature. Guidelines encourage community representative groups to develop guidelines on evidence-based best practices for managing incidental in genetic research. The characterization of preferences about the disclosure and management of these IFs is yet to be understood (20). Many commentators are recognizing the importance of the public's view to understand community norms. As these become available to researchers and RECs, they can take it into considerations to inform plans to deal with IFs.

### *Access to results*

Another question is whether participants have the right to access their results. The National Human Genome Research Institute recommends the allowance of this: Upon their request, "research participants should have access to experimental research data except when the research results are of unproven clinical validity, and the IRB has judged that there is no benefit to the research subjects" (22). Even though the access to results of benefit to participants is advocated, it is subject to logistical arrangements by researchers. Researchers must specify whether they intend to send results upon request while maintaining the privacy and confidentiality of

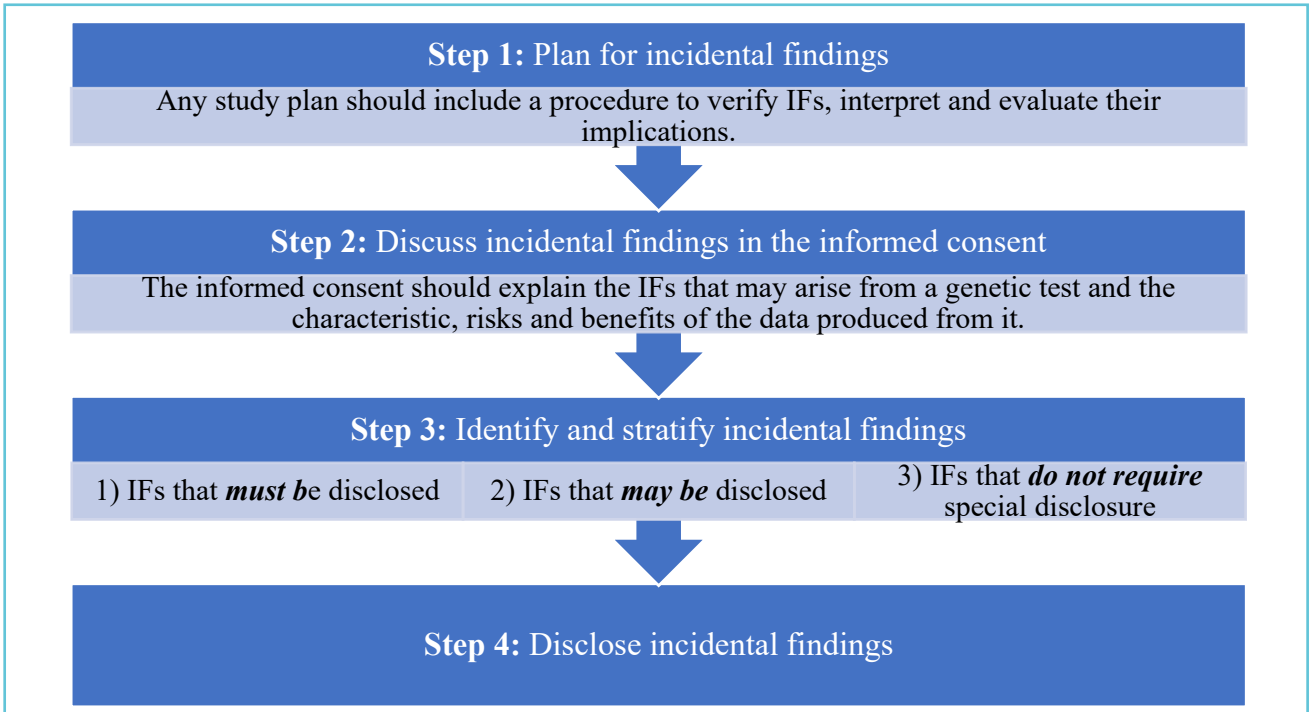
participants, time frame to do so and other details they think are worth highlighting. Discussing this in the process of informed consent ensures participants' expectations are met.

Biobanks are a research resource rather than a research project. Therefore, it is difficult to determine what an IF is when no clear objectives of the research is available. Researchers should have a policy for returning IFs that are preventable or treatable conditions of early onset to participants. Details of this should be included in the consent forms (Figure 1).

### KEY POINTS

- Guidelines have acknowledged the duty of researchers to disclose IFs.
- Researchers should have a plan to address IFs in their protocol and informed consent.
- Informed consent should explain potential risks from genetic research in general and IFs specifically including the researchers' approach to disclose them.
- Researchers should verify and interpret these results carefully. Experts and geneticists should be consulted to determine their clinical and reproductive implications. Emphasis on the quality and validity of data should be made.
- Stratification of the IFs using a **three categories approach**:
  1. Important findings with strong net benefit of disclosure
  2. Potentially important with possible net benefit if disclosed
  3. Unknown variants with no benefit

**Figure 1** A framework to ethically address incidental findings in genetic research



*This framework suggests a 4-step approach where IFs are initially planned for, discussed, stratified following identification and finally disclosed to research participants. The IFs may be stratified into three categories based on the significance of those results and their health implications. These include IFs that must be disclosed, those that may be disclosed based on the researchers' judgement and those that do not require disclosure because of their insignificance. Such stratification may need the expertise of a geneticist.*

- A compassionate participant-centered approach should be opted for.
- The autonomy and preferences of participants should always be respected.

## REFERENCES

1. Abbing HD. International Declaration on Human Genetic Data. *Eur J Health Law*. 2004;11(1):93–107.
2. Wolf SM, Lawrenz FP, Nelson CA, Kahn JP, Cho MK, Clayton EW, et al. Managing incidental findings in human subjects research: analysis and recommendations. *J law, Med ethics*. 2008;36(2):219–48.
3. Council for International Organizations of Medical Sciences (CIOMS). International ethical guidelines for biomedical research involving human subjects. Geneva; 2016.
4. Li W-H, Sadler LA. Low nucleotide diversity in man. *Genetics*. 1991;129(2):513–23.
5. Lin Z, Owen AB, Altman RB. Genomic research and human subject privacy. American Association for the Advancement of Science; 2004.
6. Lowrance WW, Collins FS. Identifiability in genomic research. *Science* (80- ). 2007;317(5838):600–2.
7. Morrison PJ. Insurance, unfair discrimination, and genetic testing. *Lancet*. 2005;366(9489):877–80.
8. Garrison NA. Genomic Justice for Native Americans: Impact of the Havasupai Case on Genetic Research. *Sci Technol Human Values* [Internet]. 2013;38(2):201–23. Available at: <http://europepmc.org/abstract/MED/28216801>
9. Cooper DN, Krawczak M, Polychronakos C, Tyler-Smith C, Kehrer-Sawatzki H. Where genotype is not predictive of phenotype: towards an understanding of the molecular basis of reduced penetrance in human inherited disease. *Hum Genet* [Internet]. 2013;132(10):1077–130. Available at: <https://doi.org/10.1007/s00439-013-1331-2>
10. Zlotogora J. Penetrance and expressivity in the molecular age. *Genet Med*. 2003;5(5):347–52.
11. Mogensen TH. STAT3 and the Hyper-IgE syndrome: Clinical presentation, genetic origin, pathogenesis, novel findings and remaining uncertainties. *JAK-STAT* [Internet]. 2013 Apr 1;2(2):e23435–e23435. Available at: <https://pubmed.ncbi.nlm.nih.gov/24058807>
12. Chandrasekaran P, Zimmerman O, Paulson M, Sampaio EP, Freeman AF, Sowerwine KJ, et al. Distinct mutations at the same positions of STAT3 cause either loss or gain of function. *J Allergy Clin Immunol*. 2016;138(4):1222–4.
13. Milner JD, Vogel TP, Forbes L, Ma CA, Stray-Pedersen A, Niemela JE, et al. Early-onset lymphoproliferation and autoimmunity caused by germline STAT3 gain-of-function mutations. *Blood* [Internet]. 2015 Jan 22;125(4):591–9. Available at: <https://doi.org/10.1182/blood-2014-09-602763>
14. Vujic M, Heyer CM, Ars E, Hopp K, Markoff A, Örndal C, et al. Incompletely penetrant PKD1 alleles mimic the renal manifestations of ARPKD. *J Am Soc Nephrol*. 2010;21(7):1097–102.
15. Rossetti S, Kubly VJ, Consugar MB, Hopp K, Roy S, Horsley SW, et al. Incompletely penetrant PKD1 alleles suggest a role for gene dosage in cyst initiation in polycystic kidney disease. *Kidney Int*. 2009;75(8):848–55.
16. Bookman EB, Langehorne AA, Eckfeldt JH, Glass KC, Jarvik GP, Klag M, et al. Reporting genetic results in research studies: summary and recommendations of an NHLBI working group. *Am J Med Genet A* [Internet]. 2006 May 15;140(10):1033–40. Available at: <https://pubmed.ncbi.nlm.nih.gov/16575896>
17. Reilly P. When should an investigator share raw data with the subjects? *IRB Ethics Hum Res*. 1980;2(9):4–12.
18. Office for Human Research Protections (OHRP). IRB Guidebook [Internet]. 1993. Available at: [http://www.hhs.gov/ohrp/archive/irb/irb\\_guidebook.htm](http://www.hhs.gov/ohrp/archive/irb/irb_guidebook.htm)
19. Beskow LM, Burke W. Offering individual genetic research results: context matters. *Sci Transl Med*. 2010;2(38):38cm20–38cm20.
20. US Department of Health and Human Services. The Belmont report [Internet]. 1979. Available at: <http://www.hhs.gov/ohrp/human-subjects/guidance/belmont.html>
21. Beauchamp TL, Childress JF. Principles of biomedical ethics. Oxford University Press, USA; 2001.
22. National Human Genome Research Institute. Federal Policy Recommendations Including HIPAA. 2013. Available at: <https://www.genome.gov/11510216/federal-policy-recommendations-including-hipaa>

# Ethics and the electronic health record: description of an integrating system of electronic health records in Argentina and a proposal to shift towards a patient-centered conception

Julián Verona<sup>1,2,3</sup>

<sup>1</sup> Central Laboratory, Hospital Dr. Felipe A. Fossati, Balcarce, Argentina

<sup>2</sup> Confederación Unificada Bioquímica de la República Argentina, CUBRA

<sup>3</sup> On behalf of the IFCC Task Force on Ethics (TF-E)

---

## ARTICLE INFO

### **Corresponding author:**

Julián Verona  
Av. Favaloro 706, Balcarce (7620)  
Buenos Aires  
Argentina  
Phone: +542266662757  
E-mail: [julianverona@hotmail.com](mailto:julianverona@hotmail.com)

### **Key words:**

patient-centered electronic health record, patient's rights, bioethical principles, reciprocity

---

## ABSTRACT

The Electronic Health Record (EHR) constitutes a complete information system useful for patient care, epidemiological studies and public health policies development.

We describe the Integrating System of EHRs of the Autonomous City of Buenos Aires (CABA), established by Law 5669, of 2016.

Although we consider the Integrating System of EHRs implemented by CABA very appropriate, we propose, first, that health services no longer store comprehensive EHRs. Instead, complete information would reside in one or several servers sheltered by civil society. Second, information would become integrated only when patients require it and grant access.

The patient would now be in a position of strength (complete autonomy). Instead of asking for his data he would be asked for them. In this sense, the patient will have to exercise the emerging responsibility of

reciprocity to the benefit of his own care and the care of others.



## INTRODUCTION

The neoteric SARS-CoV-2 pandemic exposed like never before the need to disclosure personal information to allow health system to better identify potential COVID-19 infections and track the spread over time. However, current digital solutions for monitoring have implications for privacy and data protection (1). The development of a versatile and innovative system to handle health information in a useful way for the requirement of the society and respecting the rights of people becomes increasingly necessary.

The health record is the obligatory, foliated, systematic and comprehensive documentation of a single patient medical history across time within one particular health provider's jurisdiction. It contains the day it started, patient's identification data and composition of his family nucleus including a list of the health status of immediate family members as well as their causes of death, family common diseases distributed by gender. This information may give clues about genetic, physiological and pathological backgrounds. There may also contain a family genealogical chart. For children and teenagers health record will include growth charts and developmental history to compare with age-matched mates. Children's behavior such as timing of talking, walking, etc. is also significant. Social history is helpful to know what sort of community support the patient may expect during a major illness. It would also disclose possible causes of illnesses. Habits are important too, especially those related tobacco use, alcohol intake, exercise, diet, and sexual behavior or orientation. There are also incorporated records of the acts carried out by physicians and assistants such as:

prescription and supply of medications, studies and practices conducted, treatments established, dietary prescriptions, interventions carried out by specialists and surgical protocols. Surgical history includes surgeries performed to the patient with a narrative detailing what the surgeon did. Obstetric history lists prior pregnancies and their outcomes including any complication. Internments include medical admissions and discharges, diagnoses, prognoses and evolution. In addition, allergies, vaccination certificates, the patient's willingness to donate their organs and the condition of voluntary blood donor, as well as informed consents that have been issued will be enclosed. In general, it is necessary and obligatory to incorporate in the health record any data that reflects the real information on patient's health status (*veracity*) (2). Based on subjective/objective assessment plan (SOAP) notes, problem-oriented health record (POHR) is a method of recording data about the health status of a patient in a problem-solving system and in an easily accessible way that encourage ongoing assessment and revision of the health care plan, by every actor in the health care team. The POHR consists of four components: the database containing all the information about the patient, a complete problem list (complaints), initial plans for each problem and daily progress notes, organized by problem and written in the SOAP format (3).

According to the International Standards Organization (ISO), the Electronic Health Record (EHR) is the repository of information regarding the health status of an individual in computer processable format, stored and transmitted securely and accessibly by multiple authorized users, having a standardized format that is independent of the EHR system used and aimed at the support of continuing, efficient and quality integrated health care (4).

In parallel, the Pan American Health Organization defines EHRs as records of the health information

of each patient in electronic format which can help health professionals in decision-making and treatment. In any case, the EHR is a document contained in a database, administered through computer programs, in which records of each health care event are endorsed with the provider's electronic or digital signature. It is compiled and maintained by health care agents from institutions or services. The concept of EHR goes beyond the mere digitalization. It constitutes a complete information system useful for patient care, epidemiological studies and public health policies development. The EHR offers a wide list of benefits: it guarantees accurate identification of patients, their safety and confidentiality, accelerates medical consultation appointments and scheduling, helps professionals to save time in administrative tasks, enables to share clinical information, improves legal certainty of professionals, increases the quality of health care, avoids the waste of unnecessary drugs and diagnostic studies, improves decision-making processes, reduces mistakes derived from illegibility of drug prescriptions, reduces paper expenses and helps to preserve the environment, and facilitates epidemiological surveillance, among others. There are also risks associated with its implementation: to be short-sighted and not understand the significance of implementing it for the benefit of the health system as a whole, confidentiality issues (also identified as a benefit), violation of privacy rights, problems arising when trying to computerize inefficient processes, interferences during medical consultation caused by the need to dedicate increase attention to the electronic system, among others (5).

A systematic review conducted by Kruse *et al.* (6) identified 39 barriers to EHR adoption within the literature selected ( $n = 27$ ). The most frequently mentioned were initial cost (67%), technical support (33%), resistance to changing work habits (30%), maintenance/ongoing costs (30%), training (26%), privacy concerns (19%),

insufficient time (19%) and workflow challenges (19%). In addition, Katurura *et al.* (7) performed a systematic literature review concerning e-health technology in South African organizations and found that social, technical and environmental factors influenced the success or failure of the implementation.

It is necessary to emphasize that data cannot be used for purposes other than those that motivated its collection and that public or private health facilities and professionals can collect and process personal data related to physical and mental health of patients, provided they respect professional secrecy, according to Argentine Law 25036 (8).

The health record is of patient's property. At their simple request, a copy of it, authenticated by competent authority of the health care institution, must be provided within 48 hours. Faced with the refusal, there is a legal remedy called action to protect personal data or *habeas data* in order to ensure access and collection. The Ombudsman may intervene in this process in a cooperative manner.

Health care facilities and professionals, owners of private offices, are in charge of health record's care, custody and administration, and must implement the necessary means and resources to prevent access to the information contained therein by unauthorized persons.

In Argentina, the EHR must be conceived in a way that allows effective compliance with National Law 26529 (9), "*Patient's rights, medical history and informed consent*", approved in 2009. These rights include assistance by health care professionals, without impairment or distinction due to their ideas, religious beliefs, socio-economic status, race, sex, sexual orientation, etc.; receive dignified and respectful treatment by health care agents, preserve privacy and confidentiality, and accept or reject certain medical or biological therapies or procedures, with or without



expression of cause, as well as later to revoke the manifestation of will. Argentine National Law 26061 (10), “*Law for the integral protection of the rights of girls, boys and adolescents*”, of the year 2005; establishes their right to intervene in decision-making regarding their health. Patients who present irreversible, incurable diseases or who are in a terminal stage have the right to express their will regarding the rejection of surgical procedures, artificial resuscitation or the withdrawal of life support measures, when they were disproportionate in relation to the perspectives of improvement. In these cases, measures and actions that lead to adequate control and relief of the patient’s suffering must be maintained. The patient has the right to receive health information related to their health and to carry out medical consultations to obtain second opinions on the diagnosis, prognosis and/or treatments of their diseases.

The Law also refers to the informed consent understood as the declaration of the patient’s will regarding the actions carried out on his body for his health. The consent will be issued by the patient after receiving clear, pertinent and opportune information about his health status, the proposed procedure, the expected benefits, the warning about the risks and adverse effects, the mention of the existence of alternative procedures and the foreseeable consequences for not performing the medical practice. Informed consent must be documented in the EHR.

The implementation of the EHR should lead to better compliance with bioethical principles of *autonomy, justice, beneficence* and *non maleficence*. The former implies the right to decide with which professionals personal health information will be shared. It also implies prior consent of the patient before undergoing medical practices. The second refers to the State should guarantee equitable health care and adequate accessibility to all its citizens. And the latter two, indicate professional acts always have as

their end the benefit on patient’s health, avoiding or minimizing harm.

## CASE HISTORY

The Integrating System of EHRs of the Autonomous City of Buenos Aires (CABA), established by Law 5669 – “*Electronic Medical Record Law*” (11) of 2016, is a good example to be considered, since it has many characteristics to guarantee the rights of patients and bioethical principles.

In the area of CABA, 35 hospitals, 74 primary care centers, 1 ambulatory reference medical facility, 2 children’s dental centers and 2 mental health centers are in function. It is structured into 12 geographical areas to organize health care delivery. The health system has a total of 41,000 employees (12).

The implementation of EHR requires registration in the EHR Registry and compliance with the requirements for its certification. The Integrating System of EHR is under the orbit of the Ministry of Health of the CABA, which offers the necessary software and hardware to allow the interconnection of the various databases of public, private and social security health care facilities, constituting a repository of health information contained on them. In order to harmonize the information generated in different health services, technical standards are set for the data and the information contained in the EHRs.

The Integrating System of EHRs stores minimum people identification data and basic sanitary information. The patients are incorporated through a registration process that allows their identity to be verified and accredited in order to assign an EHR. The EHR identifier assigned at each health facility must be associated with patient’s unique identifier in the Integrating System of EHRs.

The Integrating System of EHRs accounts for the information contained in each EHR, the accesses

to the database and the modifications. It ensures that the information was available to the patient and authorized users. It offers the possibility of managing appointments and preparing recipes through remote access. In addition, it permits the continuity of health care by allowing attention in different facilities, through health information exchange (*interoperability*). As an added value, it provides demographic information that can be used for public health policies design.

The CABA Ministry of Health is the certifying authority for the digital signature that will identify each of the users registered in the EHR Registry. This registry provides advice, training and technical support to health care institutions to implement EHRs and achieve its *certification*.

Professionals and patients can access the system through an authentication process using username and password. There are three levels of *access*: consultation, update and modification. Since the information contained in the EHR is confidential, access requires the authorization of the patient. In cases in which he/she was disabled to authorize it, the intervening health professional may do so compulsively, leaving record of it and respecting the principle of *confidentiality*.

Beyond the updates and modifications made by authorized persons, the EHR has the quality of remain unchanged (*integrity*) over time (*durability*). Every time one of these actions is carried out, it should be kept a record of it, even if it was intended to correct an error. Once validated, no data can be removed and, if a correction is necessary, the new data is added with the date, time and validation of the person responsible for the correction, without deleting the corrected. The system allows associating the actions carried out on the information unequivocally to an individual or entity, leaving trace of the respective access (*traceability*).

The testimony of the health care event must be simultaneous or immediately after the provision

of the service (*opportunity*) and, since the patient has access, he can verify the effective performance of this act in a timely manner. That is, he can track access to his / her clinical information. The patient has the right to know the data entered in their EHR (*privacy*) and to request a copy (*portability*). In case of incapacity or inability to understand the information due to his physical or mental state, it must be provided to his legal representative or beneficiary.

The system guarantees the *security* of the information avoiding the illegal or illegitimate use that could harm the interests or the rights of the EHR owner.

## DISCUSSION AND PROPOSAL

Currently, the EHR constitutes a significant part of patient-centered care, defined as care provision consistent with the values, needs and desires of patients and achieved when health care professionals involve patients in health care discussion and decisions (13). Patient-centered care includes effective communication, partnership and health promotion. In other words, it means exploration of the patient's diseases and illnesses to develop an understanding of patient's health care experiences, finding common ground upon which a health care plan can be developed mutually and tailoring health care plans based on reflections on the patient's past health history and current health context (14).

The Integrating System of EHRs described above represents a fairly good example of what we can do with new technology. The Federal District (CABA) took the initial step and, surely, replication by the other districts (the provinces) will follow. Over time, the system may become universal and would cover the whole country. However, as stated by Hägglund and Scandurra, introducing a national patient accessible EHRs service is a complex socio-technical challenge (15).

The Integrating System of EHRs has essential characteristics directly implicated with patient’s rights and bioethical principles: *veracity, interoperability, certification, access, confidentiality, integrity, durability, traceability, opportunity, privacy, portability and security.*

We endorse the extensive use of the EHR and consider the Integrating System of EHRs implemented by CABA very appropriate. However, we pretend to go far beyond and propose a shift in three main aspects: data ownership, storage and administration.

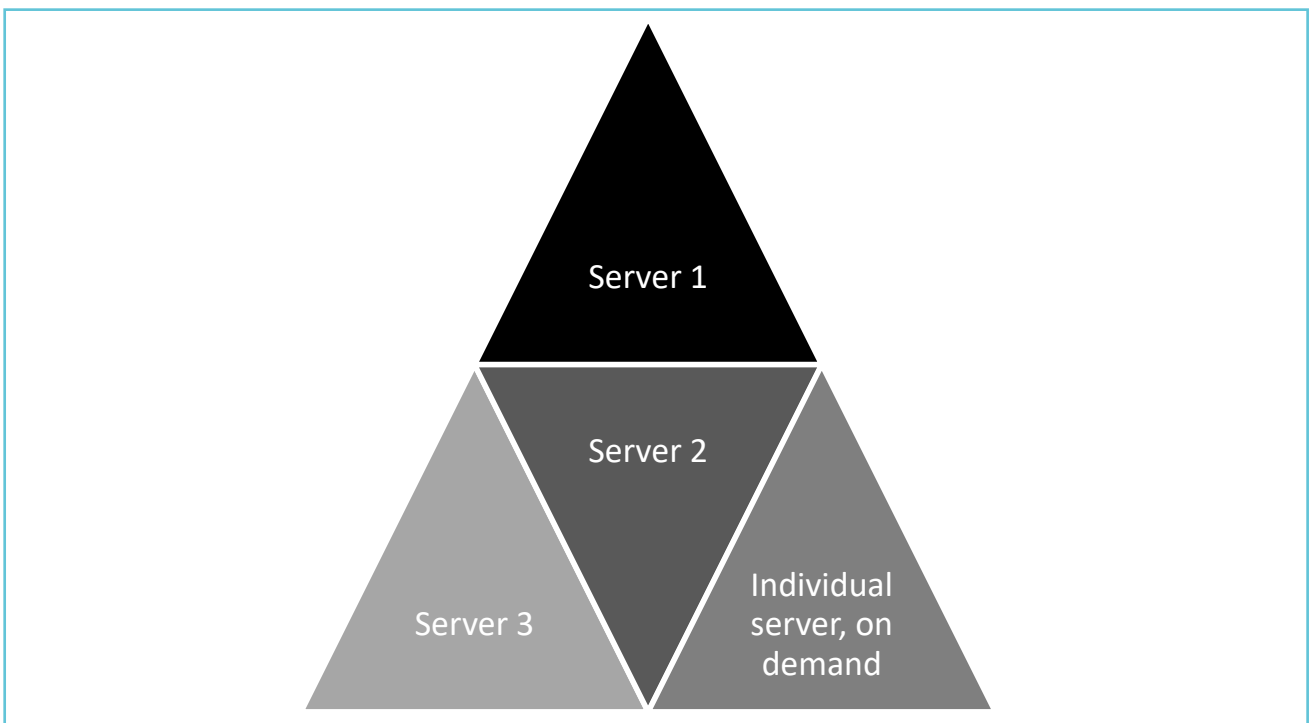
Since the patient has a recognized right over the information contained in his/her health record, he/she is actually in a position of weakness given the information resides in and is administered by an institution. Then, he/she is in the need to ask for something that actually belongs to him/her.

From our point of view, institutions should no longer safeguard EHRs. Instead, they should be

sheltered by the civil society, constituting a true social value. They should be free from political interference or economic interests. Physically, the information could be contained in one or several servers or it could even reside at the individual level, on demand (Figure 1). This does not mean health services will no longer keep records of the actions they carry out or the information they generate. Instead, we propose they should have disaggregated information while the integrated should be in patient’s hands in the form of patient-centered EHR.

Health institutions would get the integrated personal information only under express permission granted by the patient (Figure 2). In other words, the EHR would be administered by patients. A system of this nature would guarantee patient’s rights in a real way, avoiding any type of unauthorized review by the health care system and restricting the use of the information only for the purposes authorized by patients.

**Figure 1** Health information storage

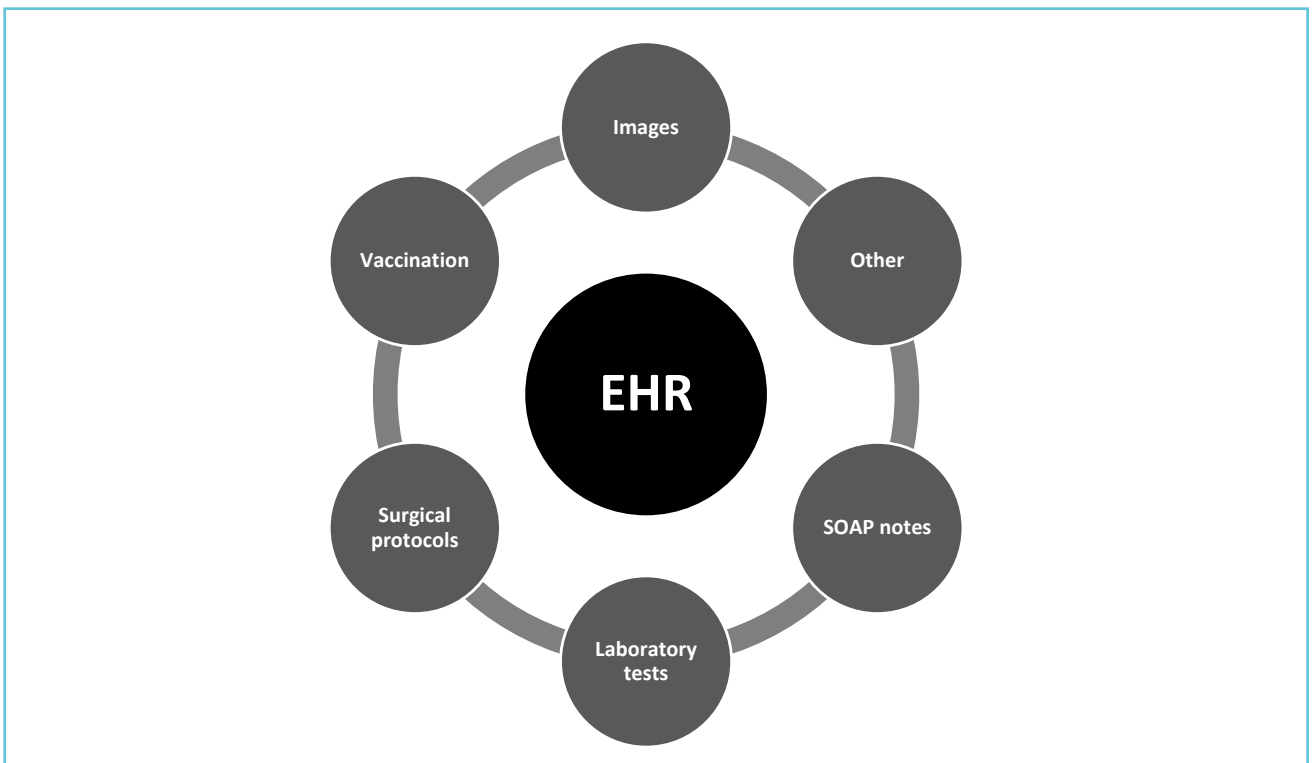


*Health information contained in several servers, including individual on demand.*

To pose an example, suppose a patient attends to a laboratory service and, then, undergoes an imaging study. Laboratory will generate, store and upload data to the patient-centered EHR. Image service will do so with images. Laboratory and image services individually could look for the studies they have done, but each piece of information will remain disaggregated until the patient attends to consultation and grants access. At this moment, the physician will be able to see both studies in an integrated EHR automatically generated in real time (Figure 2). Now suppose the patient went to two different health care services and, in the second visit, it is necessary to seek into his/her entire health record. In the same way, information coming from the two health centers and stored in external servers would become integrated as a whole under patient permission.

This kind of administration would give patients complete autonomy with respect to who views their integral health information. Actually, this restriction could affect the principle of *justice* since ignorance of information recorded in the health record could generate the need to carry out studies and tests already done. Clearly, this situation would generate a misuse of resources. It could also be affected the right of health care professionals to have access to the information contained in the health record; the principles of *beneficence* and *non maleficence* could then be compromised (*primum non nocere*, not to harm) (16). In fact, professionals would have the right to ask patients for access to their respective EHRs, to perform with professionalism. At this point, a new responsibility for the patients emerges: that of *reciprocity*. It implies that patients would allow their EHR data to be use to the

**Figure 2** Real time generation of patient-centered electronic health record



Health care institutions having disaggregated information while the electronic health record is generated at the patient's request.

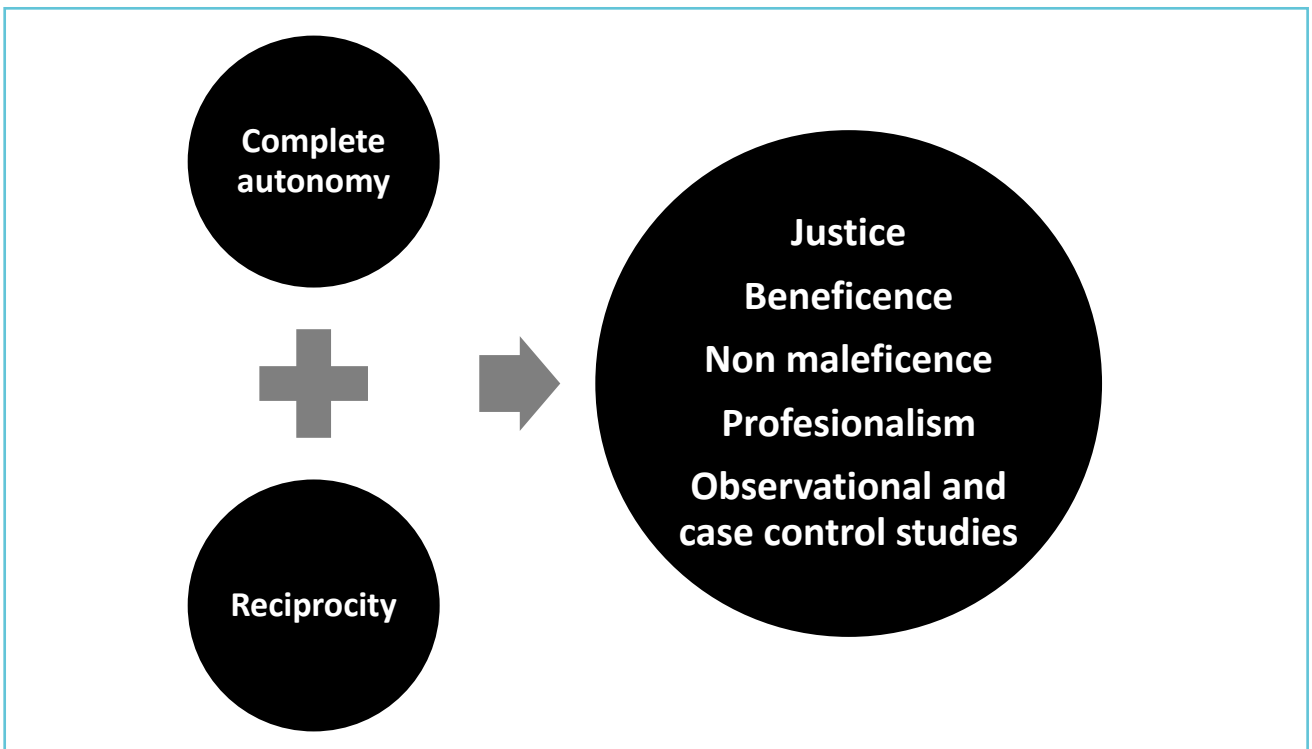
benefit of their own care and the care of others (Figure 3) and this should include the release of the information for population studies and epidemiological research (observational and case-control studies). For these purposes, data quality is a subject of mayor concern and patient’s reviews are highly important. Data must be accurate and reliable to avoid potential hazards that could range from individual harm to erroneous conclusions caused by bias at population level (17).

Although, the patient is the owner of his health record, there are issues that should be open access to health care providers: blood type, allergy to certain drugs, pathologies of epidemiological relevance or any meaningful information for health care in an emergency or serious context. The above could be expressed in a general way, with pre-established formulas, without going

into details that could affect *dignity, integrity, vulnerability, professional secrecy*, among other issues.

From an operational point of view, an application designed in this way should be easy to use. Verbal expression or a simple exchange of text messages should be sufficient to have the necessary access code and be able to view the health record for a certain period of time. This would give full participation to the patient and allow professionals to consult and upload health events. Institutions or individual professionals would initiate a session to access the application and thus the responsibility of each professional act would be established. Personal preferences and predetermined informed consent would be incorporated into the application in digital format, which would constitute valid consent for

**Figure 3** Relational equation of bioethical principles including reciprocity and professionalism



*Patient exertion of complete autonomy plus the emerging responsibility of reciprocity warrant bioethical principles of justice, beneficence and non maleficence, ensure that health care providers perform with professionalism and permit to conduct epidemiological research (observational and case control studies).*

situations where the patient's circumstances made it impossible to obtain. The application would support different types of data formats: text, portable document format (pdf), audio, image, video, etc. The system would also include vaccination programs and the recommended frequency of carrying out medical practices in order to ensure adequate prevention and would issue the corresponding alerts. In special circumstances (accidents, loss of consciousness, etc.), compulsory access by institutions or health care professionals would be possible, but the system would register the entry. This could be done through fingerprints or facial recognition. The application would also have the ability to manage geographically referenced epidemiological information through the use of global positioning capabilities (GPS) of smart phones and would allow public health centers to extract real time global data. For these centers, interfaces specifically designed for data acquisition and analysis would be developed in accordance with international regulations. Then, the reflection made in the first paragraph of the introduction refers to a well conceived EHR.

Finally, a SWOT (strengths, weaknesses, opportunities, threats) analysis must be carried out on the system adopted by the CABA, trying to find out its ability to coordinate with equivalent systems from other jurisdictions that may constitute their own integrated EHRs systems in the future (18). A strategic plan proposal should be considered to formulate an alternative for the growth, development and articulation of integrated EHRs systems, based on introducing proactive changes: that of the repository and that the administration of individual EHRs were in patient's hands, as the unique owner of his health data.

## CONCLUSIONS

After reviewing the Argentine legislation on the matter and the auspicious initiative conducted

by the CABA, here we propose a radical change in the conception of EHRs. It would happen to belong by right and now also in fact to the patient. It would reside in one or several servers protected by the civil society or even in individual repositories, on demand. Institutions would only have disaggregated information. The EHR would be administered by patients. In this way, greater guarantees regarding their rights would be given to the patient, and bioethical principles would be observed. The patient would now be in a position of strength (complete *autonomy*). Instead of asking for his data he would be asked for them. In this sense, the patient will have to exercise the emerging responsibility of *reciprocity* both for his/her own wellness and for the wellness of the others. However, the implementation of such a system will take a long time and will require the design of an appropriate information network and the change of mind of many actors including patients. Clearly, patient-centered EHR as described above constitutes a tangible expression of bioethical principles and a paradigm shift in health information ownership, storage and administration.

## REFERENCES

1. Organisation for Economic Co-operation and Development (OECD). 23rd report "Tracking and tracing COVID: protecting privacy and data while using apps and biometrics". April 2020.
2. Guillum RF. From papyrus to the electronic tablet: a brief history of the clinical medical record with lessons to the digital age. *Am J Med* 2013;126(10):853-857.
3. Wright A, Sittig DF, McGowan J, Ash JS, Weed LL. Bringing science to medicine: an interview with Larry Weed, inventor of the problem-oriented medical record. *J Am Med Inform Assoc.* 2014;21(6):964-8.
4. International Organization for Standardization (ISO). Health informatics — Electronic health record — Definition, scope and context. ISO/TR 20514:2005(en). <https://www.iso.org/obp/ui/#iso:std:iso:tr:20514:ed-1:v1:en>. Accessed, May 2020.
5. Pan American Health Organization. Electronic Medical Records in Latin America and the Caribbean: An Analysis

of the current situation and recommendations for the Region. Washington, DC: PAHO, 2016.

26riers to Electronic Health Record Adoption: a Systematic Literature Review. *J Med Syst.* 2016;40(12):252.

7. Katurura MC, Cilliers L. Electronic health record system in the public health care sector of South Africa: A systematic literature review. *Afr J Prim Health Care Fam Med.* 2018;10(1):e1-e8

8. Ley Nacional 25036, “Ley de Protección de Datos Personales”, Argentina, 2000.

9. Ley Nacional 26529, “Derechos del paciente, historia clínica y consentimiento informado” Argentina, 2009.

10. Ley Nacional 26061. “Ley de protección integral de los derechos de las niñas, niños y adolescentes”, Argentina, 2005.

11. Ley 5669, “Ley de Historia Clínica Electrónica”, Ciudad Autónoma de Buenos Aires, 2016.

12. Faretta F, Levi D, Marques L, Ferrante D, GiussiBordoni MV, Baum A, de Quirós FGB. Developing an Electronic Record Tool Representative of Primary Health Care in the

Public Health Care System of Buenos Aires City. *Stud Health Technol Inform.* 2019;264:516-520.

13. Mead N, Bower P. Patient-centredness: a conceptual framework and review of the empirical literature. *Soc Sci Med.* 2000;14:1087–1110.

14. Little P, Everitt H, Williamson I, Warner G, Moore M, Gould C, Ferrier K, Payne S. Preferences of patients for patient centered approach to consultation in primary care: observational study. *BMJ.* 2001;322(7284):468-72.

15. Hägglund M, Scandurra I. A Socio-Technical Analysis of Patient Accessible Electronic Health Records. *Stud Health Technol Inform.* 2017;244:3-7.

16. Meslin EM, Schwartz PH. How bioethics principles can aid design of electronic health records to accommodate patient granular control. *J Gen Intern Med.* 2015;30 Suppl 1:S3-6.

17. Lee LM. Ethics and subsequent use of electronic health record data. *J Biomed Inform.* 2017;71:143-146.

18. Shahmoradi L, Darrudi A, Arji G, FarzanehNejad A. Electronic Health Record Implementation: A SWOT Analysis. *Acta Med Iran.* 2017;55(10):642-649.

# How does the MedTech Europe Code of Ethical Business Practice affect the activities of professional societies in laboratory medicine?

Valdas Banys<sup>1,2,3</sup>

<sup>1</sup> Vilnius University, Faculty of Medicine, Institute of Biomedical Sciences, Department of Physiology, Biochemistry, Microbiology and Laboratory Medicine, Vilnius, Lithuania

<sup>2</sup> Vilnius University Hospital Santaros Klinikos, Center of Laboratory Medicine, Vilnius, Lithuania

<sup>3</sup> Lithuanian Society of Laboratory Medicine

---

## ARTICLE INFO

### **Corresponding author:**

Valdas Banys, MD, PhD  
Santariskiu str. 2  
08661 Vilnius  
Lithuania  
Phone: +370-5-2501361  
E-mail: [valdas.banys@santa.lt](mailto:valdas.banys@santa.lt)

### **Key words:**

MedTech Europe, the Code of Ethical Business Practice, continuous professional development, laboratory medicine

---

## ABSTRACT

The MedTech Europe Code of Ethical Business Practice came into effect on 1 January 2018. It was created by the medical technology industry. It addresses the importance of fair management of educational grants: public disclosure of provided educational grants, compliance of conferences with the Conference Vetting System; allocation of grants to healthcare organizations (HCOs) but not to the healthcare professionals (HCPs); the need for written contracts with HCOs, etc. As a National Society and member of IFCC and EFLM, the Lithuanian Society of Laboratory Medicine (LLMD) has created a fund dedicated to the continuous professional development of LLMD member HCPs. The fund, as an instrument for the ethical use of money, corresponds to the principles of the MedTech Code of Ethical Business Practice and is an example on how HCOs can implement it to ensure ethical communication between the IVD (In Vitro Diagnostics) industry,



HCOs and their member HCPs. Scarce data exists on the level of MedTech acceptance and implementation among HCOs and HCPs, thus more effort has to be made to better communicate and consequently improve fair use of the funds received from the industry, and to improve the ethical behavior of HCPs.



## CHANGE IS HERE

Diagnostic companies all around the Europe are no longer allowed to pay specialists in laboratory medicine directly to attend third-party educational conferences. Since 2015, when medical technology companies joined together to form the alliance we know today as MedTech Europe, the world of continuous professional development has changed dramatically. The changing situation affected all healthcare professionals (HCPs) and healthcare organizations (HCOs), including those involved in laboratory medicine. The pharmaceutical industry did not stop direct sponsorship, but rather continued it while agreeing to declare all relevant relations. But MedTech Europe went a huge step further, declaring that “...there are certain conflicts of interest that maybe we should prohibit. Transparency is not enough,” (1) and started to foster the highest ethical standards in the medical technology industry, including for all activities related to training, medical education and professional relationships with HCPs (2).

It is now undoubtedly clear that not only the International Federation of Clinical Chemistry and Laboratory Medicine (IFCC), the European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) and their National Societies (NSs), but also every individual laboratory medicine specialist should be aware of the importance of the ethical issues described in the MedTech Code (2, 3). Briefly, the MedTech Code sets stronger rules for educational grants:

1) medical technology companies should publicly disclose educational grants provided to HCOs, ensuring increased transparency of the funds allocated to medical education; 2) conferences supported by these companies must comply with the Conference Vetting System; 3) companies are only able to provide grants, charitable donations, scholarships or fellowships to HCOs but never to individuals; 4) companies are able to define the category of HCPs eligible for financial support under the grant but not to choose individual HCPs; 5) companies are required to sign a written contract with HCOs setting out the terms and conditions for grants, charitable donations, scholarships or fellowships; and 6) companies must establish an internal and independent process based on objective criteria to review grant requests (4).

## TWO LEVELS OF INTERACTION

MedTech Europe focuses on several levels of interactions between Member companies, HCPs and HCOs. The individual level of interaction with HCPs covers two areas of interest. First, HCPs are interested in participating in scientific meetings, conferences or other third-party-organized events. Secondly, HCPs might be involved as consultants and advisors to provide bona fide consulting and other services, including but not limited to research, inclusion on advisory boards, presentations at Company Events and product development (2). The latter form of involvement might be remunerated by the IVD industry and is subject to clear rules to be followed by the industry and by HCPs themselves. The other level of interactions is organizational and requires that HCOs set their rules and criteria for ethical organization of scientific events (5) and ethically support their member HCPs who participate in those events. The focus of this article is on individual HCPs who are in need of financial support for the purposes of training and continuous professional development, and also

HCOs who are expected to apply high ethical standards for the aforementioned interactions.

It is almost impossible to judge how HCOs behave and which practices they apply without conducting a properly organized survey. The first attempt to investigate the landscape of MedTech acceptance and implementation among European professional societies of laboratory medicine was carried out by the Committee on Education and Training (C-ET) and the Working Group for Congresses and Postgraduate Education (WG-CPE) at EFLM (3). The results of the surveys conducted are heterogeneous but provide interesting ethical aspects, which should be discussed by each HCO and transferred to their HCPs. The authors found that there still are some National Societies who have not yet adopted the MedTech Europe Code, thereby raising the risk of direct financing of HCPs by the IVD industry. The survey results have shown that even up to one fifth of respondents always use direct IVD financing. Detailed survey results and outcomes can be found in the EFLM journal *Clinical Chemistry and Laboratory Medicine* (3). Meanwhile the Legal and Compliance team at MedTech Europe have documented in their Compliance Report 2019 a significant increase in the percentage of National Associations that have banned direct sponsorship (from 13 % in 2017 to 95 % in 2019) (6).

### **AN EXAMPLE OF ONE SOCIETY**

As a National Society and member of the IFCC and EFLM, the Lithuanian Society of Laboratory Medicine (LLMD) felt obliged to act fairly. Thus, for the purposes of compliance with the MedTech Europe Code of Ethical Business Practice, the LLMD implemented a special measure in 2017 – the fund for continuous professional development of the LLMD member HCPs. It has determined the rules for the ethical use of the fund. By way of these rules the LLMD defines the procedures for the organization of

scientific conferences and support of the continuous professional development of its member HCPs (specialists in laboratory medicine). The purpose of the fund is the financing of scientific conferences organized by the LLMD (LLMD events) and the financing of the expenses of LLMD member HCPs dedicated to their continuous professional development in external conferences not organized by the LLMD (external events). As a beneficiary, the LLMD accumulates resources in the Fund as educational financial support/sponsorship from support givers according to the national law of charity and sponsorship. The Meeting of the Board members of the LLMD plans the usage of the Fund yearly and makes plans for LLMD events and external events to be funded.

The funds are most frequently used to provide LLMD member HCPs with the opportunity to participate in external (third-party-organized) events, which are normally costly and beyond the financial means of LLMD members. However, it is not possible to grant funding to all applicants, and so the LLMD has listed the priorities according to which its member HCPs are funded. First, funds are allocated if an LLMD member HCP gives an oral presentation, has provided an abstract and/or poster, is a member of the scientific/organizing committee or the chair of a scientific event. Secondly, an LLMD member HCP is granted funding if he or she participates as a corresponding member, national representative or a member of the committee/working group of the IFCC, EFLM or another subspecialty professional society. Following the aforementioned individuals, priority is then given to LLMD member HCPs who are members of working groups created by the Ministry of Health of the Republic of Lithuania and young scientists under the age of 35 years, and who in the past 12 months have published at least one article in the national journal of the LLMD (*Laboratorine medicina*)

or any other peer-reviewed journal. Finally, any other LLMD member HCPs who have submitted a written application for financial support are considered.

Only certain expenses can be covered by the Fund. When an applicant is selected for an external scientific event, only 1) the registration fee (preferably an early bird registration fee), 2) accommodation expenses (where the external event takes place abroad and the cost of the accommodation per night does not exceed the amount defined by the relevant Order of the Minister for Finance) and 3) travel expenses (only standard/economy flight tickets, or bus/train tickets if flights to the destination city are unavailable) are eligible for funding. Also, pursuant to the MedTech Code, the LLMD does not pay for 1) participation in social events (i.e. gala dinners), 2) taxis, public transport or other transportation services at the destination city where the external event is held, 3) personal expenses of LLMD member HCPs, 4) expenses of persons accompanying LLMD member HCPs, 5) travel insurance and 6) daily allowances. These must be covered by the applicant himself/herself.

Funding of the LLMD's HCPs was not always consistent before the implementation of the funding rules. In 2012, it was discovered that a substantial number of applications for educational grants from the LLMD were coming indirectly from IVD companies or their national distributors. The IVD industry was selecting HCPs to be funded and granted funds to the LLMD. After implementation of the rules for ethical funding, the situation changed. In our opinion for the better, because even if the IVD industry is interested in certain HCPs, the LLMD would decide itself whether or not the HCP in question complies with the rules, and then finally allocate funds irrespective of the wishes of the IVD industry.

From 2013 to 2019, the LLMD was able to identify 19 attempts at targeted funding of LLMD member HCPs (Table 1) and was able to successfully change their funding policy to block these attempts. The increasing number of detected targeted funding attempts reflects the Society's efforts to ensure fair funding and successful implementation of the MedTech Europe Code of Ethical Business Practice. Interestingly, all targeted HCP funding attempts were performed by the distributors of IVD companies, acting as independent legal entities, which work as distributors of several IVD companies (brands) at once. This creates an impression that IVD companies (original brands) do not pay enough attention to the compliance of their distributors. Thus HCOs (in this particular case the LLMD itself) have to act fairly and start communicating with irresponsible entities (national distributors) within the IVD industry chain.

In terms of internal LLMD events, the LLMD has experience in organizing international congresses in the field of laboratory medicine, namely BALM (Baltic Congress of Laboratory medicine). One of the last congresses took place in 2018 in Vilnius (Lithuania), which was already compliant with the MedTech Europe Code's General Criteria for Events and successfully approved in the Conference Vetting System. All 6 criteria were met successfully: scientific programme, geographic location, venue, hospitality, registration package benefits and communication (5), according to the IFCC's guidance and recommendations (7).

## **FUTURE PERSPECTIVES**

The diversity of the EFLM survey results (3) and the lack of communication between the IVD industry and leading HCOs (8) leads to several questions for the future of conferences and their attendance. Will there be any provisions and changes in the MedTech Code? Will HCPs

**Table 1** Overview of educational grants at the Lithuanian Society of Laboratory Medicine (LLMD) in 2013-2019

Financial year	Number of funded HCPs	Number of external events	Average expenses per HCP, EUR	Number of agreements with IVD companies or their distributors	Number of blocked targeted HCP funding attempts
2013	33	16	1171	14	0
2014	37	15	1253	30	0
	107	16 (including BALM)	628		
2015	75	19	1372	31	1
2016	33	17	855	22	3
	110	18 (including BALM)	449		
2017	64	19	967	9	5
2018	36	14	1114	23	7
	138	22 (including BALM and 7 other small national events)	447		
2019	58	17	1165	11	3

**Abbreviations in the table:** BALM – Baltic Congress of Laboratory Medicine (usually involving smaller registration fees and less travel expenses); IVD – in vitro diagnostics; HCP – healthcare professional.

and HCOs achieve full compliance? Will there be less attendees at congresses and conferences? It seems that the Code is not going to change, and our profession has to live with it. Still, we have to put more effort into achieving better communication between the IVD industry and HCPs in order to guarantee improved and fair use of the funds received from the industry, and we must also redouble efforts

dedicated to organizing and attending scientific events (3). There is a need to follow up on C-ET and WG-CPE surveys in terms of how countries have managed to implement the principles of the Code or to improve the ethical behavior of their HCPs, as well as in terms of whether individual HCPs become more conscientious (9) in maintaining fair interactions with the IVD industry.

The year 2020 has thus far proven challenging to everyone involved in postgraduate training and organization of scientific events due to the global COVID-19 pandemic. Many highly valuable congresses and conferences have had to be cancelled or postponed for a long time, while others have been held virtually in order to give HCPs the option to attend events online, sometimes even without any registration fee. This format is an option for the future of scientific events and has to be considered by HCOs and event organizers.

#### REFERENCES:

1. Arie S. The device industry and payments to doctors. *Br Med J* 2015; 351:h6182.
2. MedTech Europe Code of Ethical Business Practice. [https://www.medtecheurope.org/wp-content/uploads/2017/06/2020\\_MTE\\_MedTech-Europe-Code-of-Ethical-Business-Practice-QA-DG.pdf](https://www.medtecheurope.org/wp-content/uploads/2017/06/2020_MTE_MedTech-Europe-Code-of-Ethical-Business-Practice-QA-DG.pdf). Accessed: 27 Jul 2020.
3. Pasalic D, Homsak E, Buno A, Bergmann K, Carru C. Practice in financial support of third party organized conferences and courses at a national level for health care professionals in Europe. *Clin Chem Lab Med* 2019; 57(9): 1319-28.
4. MedTech Europe Code of Ethical Business Practice. Leaflets for Healthcare Organisations (HCO). <https://www.medtecheurope.org/resource-library/support-to-medical-education-and-interactions-with-healthcare-organisations/>. Accessed: 28 Sep 2020.
5. MedTech Europe. <https://www.ethicalmedtech.eu/conference-vetting-system/objective/>. Accessed: 27 Jul 2020.
6. MedTech Europe Activity Report 2019. <https://www.medtecheurope.org/resource-library/medtech-europe-activity-report-2019/>. Accessed: 28 Sep 2020.
7. Wesenberg JC. IFCC Guidelines for compliance with applicable codes of ethical business practice. *IFCC eNews* 2018; 6:3-4.
8. Howard AJ. New Medtech Code of Ethics and Cardiology. *Eur Heart J* 2018; 39:3270-1.
9. Wijeratne N, Benatar SR. Ethical issues in laboratory medicine. *J Clin Pathol* 2010; 63(2): 97-8.

# The clinical laboratory: a key player in diagnosis and management of COVID-19

Sojit Tomo, Sreenivasulu Karli, Karthick Dharmalingam, Dharmveer Yadav, Praveen Sharma

Department of Biochemistry, All India Institute of Medical Sciences, Jodhpur, Rajasthan, India

---

## ARTICLE INFO

### **Corresponding author:**

Dr. Dharmveer Yadav  
Associate Professor  
Department of Biochemistry  
All India Institute of Medical Sciences  
Jodhpur, Rajasthan, India  
E-mail: [dharam143s@gmail.com](mailto:dharam143s@gmail.com)

### **Key words:**

COVID-19, SARS-CoV-2, laboratory,  
clinical parameter, diagnosis, prognosis

---

## ABSTRACT

The Coronavirus disease 2019 (COVID-19) outbreak, caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), had emerged as a pandemic affecting almost all countries in the world in a short span after it was first reported in December. Clinical laboratories have a crucial role in mitigating this new pandemic. Timely and accurate diagnosis of COVID-19 is of paramount importance for detecting cases early and to prevent transmission. Clinical laboratories have adopted different test modalities and processes to tackle this unprecedented situation with directives from regulatory bodies such as the WHO. The varying presentations, as well as complications attributed to comorbidities in COVID-19, have created hurdles in the management of these patients. Various clinical laboratory parameters have been investigated for their potential for diagnosis and prognosis of the disease, prediction of complications and monitoring of treatment response. Different routine and uncommon parameters have been shown to have the diagnostic and prognostic capacity. This update discusses the role of the laboratory in diagnosis, prognosis and

monitoring of treatment response. Different methodologies for diagnostic testing as well as various clinical laboratory parameters having diagnostic and predictive powers have been discussed. This compilation organises relevant available information on various clinical laboratory parameters and their role in COVID-19 mitigating pandemic.



## **1. INTRODUCTION:**

Novel Coronavirus induced pneumonia, which was given the name of coronavirus disease 2019 (COVID-19) by the WHO on the 11th of February 2020, has rapidly amplified to the full scale of a pandemic since it was first reported in Wuhan, China, back in December 2019 (1,2). COVID-19 is the clinical syndrome associated with SARS-CoV-2 infection. The disease signifies a respiratory syndrome starting from mild upper respiratory illness to severe pneumonia and acute respiratory distress syndrome (ARDS). SARS-CoV-2 belongs to the beta coronavirus genus of the coronaviruses. Although Severe Acute Respiratory Syndrome (SARS) and Middle East Respiratory Syndrome (MERS) also belongs to the same genus, SARS-CoV-2 leads to milder infections.

However, SARS-CoV-2 have a broader community transmission when compared with SARS and MERS. Hence, laboratory testing is of paramount importance to distinguish between COVID-19 and other respiratory diseases. Moreover, extensive testing will help in COVID diagnosis and a better understanding of disease prevalence in asymptomatic infections. As of November 10, 2020, there have been over 50 million confirmed cases of COVID-19 and over 1.2 million deaths across the world. Contribution of Laboratory medicine in diagnosis, prognosis, risk prediction and management is indispensable in most

of the human pathologies, and COVID-19 is not an exception. The current COVID-19 pandemic has reconfirmed that laboratory diagnostics will remain the core of every clinical decision made. This review covers recent laboratory modalities available for diagnosis, prognosis and monitoring of treatment response in COVID-19.

## **2. LABORATORY TESTING IN COVID-19**

Clinical Laboratories are of paramount importance in mitigating the COVID-19 pandemic. From early diagnosis, Clinical Laboratories play a crucial role in monitoring comorbidities, diagnosing complications, assessment of treatment responses and in assessing the prevalence of diseases in the community. Timely and accurate diagnosis of the disease is essential for early initiation of treatment as well as to prevent the transmission to contacts.

Different countries had followed and implemented different testing strategies targeting different genes based on the availability of diagnostic methods and consumables (Table 1). Further, the WHO has meanwhile taken strict steps and created the diagnostics available with the mission to “detect, protect and treat” to break the chain of transmission of SARS-CoV-2(3). Early diagnosis and immediate treatment will significantly decrease future COVID-19 cases. Therefore, early laboratory diagnosis of SARS-CoV-2 plays a vital role in controlling the COVID-19 pandemic.

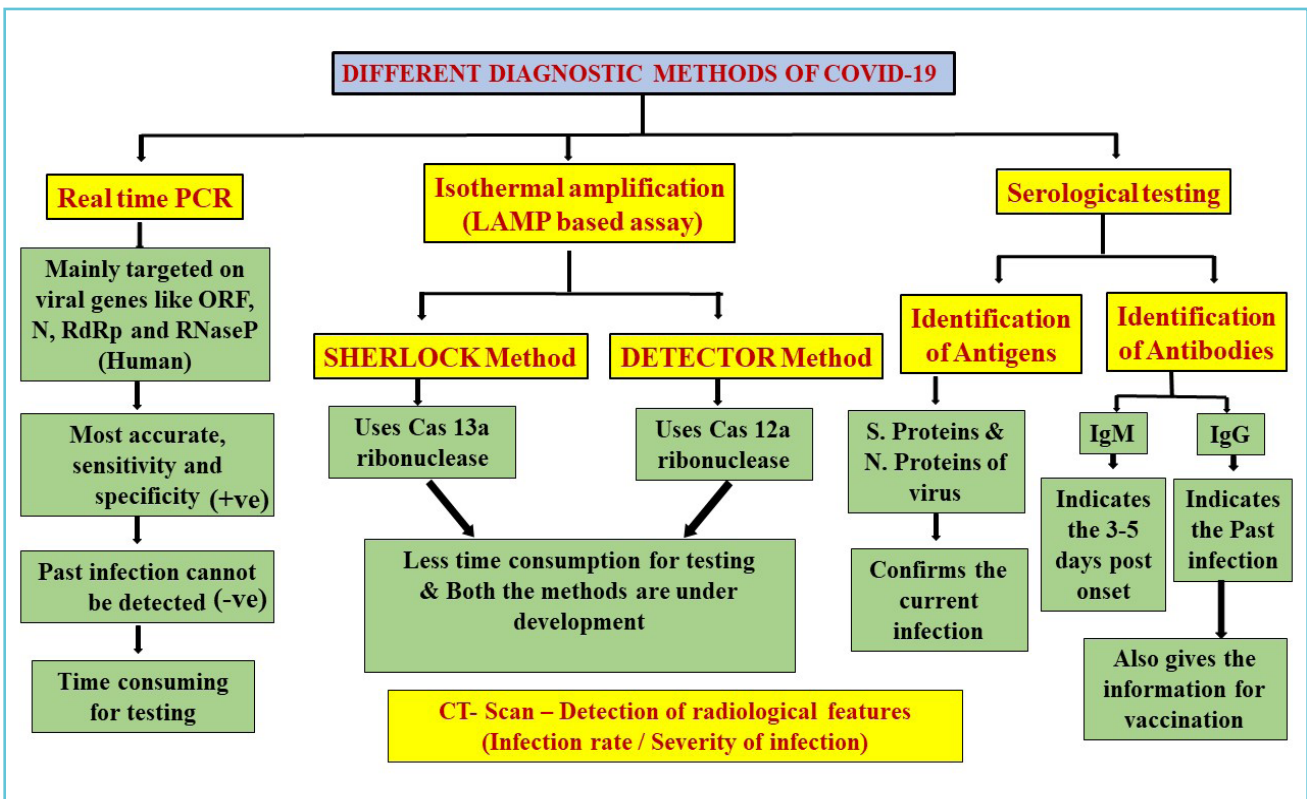
### **3.1 DIAGNOSTIC METHODS FOR COVID-19**

Compared to symptomatic testing and CT scan method, the molecular techniques are more appropriate in accurate diagnosis since they target the identification of pathogens (Figure 1). Despite this, the Real-time reverse transcriptase-polymerase chain reaction (rRT-PCR) serves as a gold standard method for nucleic acid screening

**Table 1** Currently targeting different genes by the different country protocol as per WHO

Country	Institute	Targeting gene	References
China	China CDC	ORF 1ab and N genes	(4)
Hong Kong SAR	Hong Kong University	ORF 1b-nsp14, N genes	(5)
Germany	Charitè	RdRp, E, N genes	(6)
Japan	National Institute of Infectious Diseases	N gene	(7)
Thailand	National Institute of Health	N gene	(8)
USA	US CDC	Three targets in N gene (N1, N2, and N3) RP-RNase	(9)

**Figure 1** Different diagnostic methods of COVID-19





of SARS-CoV-2. Since this a time consuming and sophisticated method, rRT-PCR serves better as a diagnostic tool than a screening tool(10). Considering the current stage of the pandemic, a large number of patient screening is needed using novel screening methods which require lesser equipment and materials.

The advancement of molecular techniques is mainly reliant on understanding the genomic and proteomic composition of the pathogen. Similarly, the changes in the host gene or protein expressions induced by the pathogen after infection (11). World Health Organization (WHO) and China jointly described genome sequence of SARS-CoV-2 and its genetic characterisation (12,13). This genome sequencing has given a road map to researchers for designing primers and probe sequences for rRT-PCR and some other nucleic acid amplification tests.

### **3.2 SPECIMEN COLLECTION, SAMPLE STORAGE AND SAMPLE STABILITY**

The World Health Organization (WHO), Centre for Disease Control and Prevention (CDC) and Indian Council of Medical Research (ICMR) have recommended a set of guidelines to collect the samples from patients affected or suspected by COVID-19 (14–16). For the safety of clinicians and researchers, it is highly necessary and suggested that the specimens should collected in a BSL-2 laboratory. It is compulsory by law for the individuals concerned or suspected to cooperate appropriately with health departments for collection, storage and shipment of the specimens. In the case of unavailability of immediate testing, store the specimens at 2-8°C for up to 72 hours after collection. If a delay in testing or shipping is expected, store specimens at -70°C or below. Rodino et al demonstrated the stability and reliable detection of SARS-CoV-2 RNA in stored swabs in viral transport medium, saline, PBS and minimal essential medium after

seven days at 2-8°C and frozen at -20°C using an in-house Emergency Use Authorization (EUA) assay as well as the Roche Cobas EUA assay (17). In another study Perchetti GA et al. shown that SARS-CoV-2 stability can be retained at 4°C for up to a month if the storage of -80°C is not available (18).

The sample should be isolated from two main sites, the lower respiratory or upper respiratory tract, depending on WHO's suggested guidelines. The nasopharyngeal or oropharyngeal swab specimens should be collected from the upper respiratory tract, while tracheal aspirate, bronchoalveolar lavage, and sputum should be collected from the lower respiratory tract. Bronchoalveolar lavage fluid specimens remain the ideal sample for detection of COVID-19. Sputum, nasal swabs, fibre bronchoscope brush biopsy, pharyngeal swabs and faeces demonstrated different rates of positivity for COVID-19 virus. Urine was not found to be a suitable sample for detection of COVID-19 (19).

### **3.3 NUCLEIC ACID AMPLIFICATION TEST (NAAT)**

Since the outbreak of COVID-19, nucleic acid amplification testing is the primary method of diagnosis. Multiple real-time reverse transcription-polymerase chain reaction (rRT-PCR) kits have been invented to detect SARS-CoV-2. Corman et al. aligned and scrutinised SARS-CoV-2 related viral genome sequences to construct a set of oligo primers and probe sequences(6). Among these mainly three conserved sequences have been revealed. 1) In open reading frame ORF1ab region the RdRP gene (RNA-dependent RNA polymerase gene), 2) Envelope protein gene (E gene), 3) Nucleocapsid protein gene (N gene) (6). Different countries submitted their primary probe designs to the WHO. As an example, the rRT-PCR can be designed as two genes target system or three genes target systems, where

one primer set detects family of coronaviruses, the second set detects specifically SARS-CoV-2 and third is human RNase P as the internal control (Table 1). Similarly, ICMR also released some recommendations for COVID-19 diagnosis. The ICMR has recommended the use of US-based RT-PCR probes distributed to national laboratories (16).

The United States Centers for Disease Control and Prevention (CDC) set up a panel of genes through RT-PCR for the specific finding of SARS-CoV-2 and overall detection of SARS-like beta coronaviruses (9). Primarily designed by targeting three different sets of primers to the N gene among these two primers sets are specific to SARS-CoV-2 and one primer set is specific to all beta coronaviruses. If all three genes are positive, then it specifies the COVID-19 confirmation. Similarly, in Germany Charite (6) developed two sets of nucleic acid tests for detection of SARS-CoV-2, SARS-CoV and bat-like beta-CoVs by targeting the RdRp and E genes, if both tests were positive then COVID-19 confirmation through SARS-CoV-2 specific RdRp gene. The results of the Chu et al. study suggested targeting the N gene as primary screening and ORF1ab as a confirmative target. Studies targeted at two or more genes, thus had a stronger outcome performance compared to single genes alone (20). Now, molecular testing was developed as the gold standard for the diagnosis of SARS-CoV-2, hence the E and RdRb genes suggesting better analytical sensitivity compared to the N and ORF1ab genes combination.

While different institutions have developed various SARS-CoV-2 research protocols, it remains uncertain if the findings of nucleic acid tests based on multiple targets are comparable. In a recent study compared the analytical sensitivities of the United States, Germany, Hong Kong and China qRT-PCR assays by using RNA transcripts isolated from a COVID-19 patient (21).

They found that all primer-probe sets used in the qRT-PCR tests could detect SARS-CoV-2, but the significant difference was observed in the limit of detection (LOD) and the ability to distinguish the positives and negatives while the viral load is at lower levels. The highest sensitivity of primer-probe sets was found E-gene (Germany), N1 gene (US CDC), ORF1 (Hongkong) but RdRp gene (Germany) showed the lowest sensitivity. In another study from Germany Konrad et al. found that by using a single-step qRT-PCR method, the E gene target was more sensitive than the RdRp target (22).

### **3.4 DIRECT RT-PCR**

The positive controls (2019-nCoV pseudovirus) provide a nucleic acid extraction and a reverse transcription control to validate the entire procedure and reagent integrity. Similarly, the RNase P internal control provides an RNA extraction of practical control and secondary negative control. However, RNA extraction from clinical samples creates a major bottleneck in the diagnostic process, as it either runs manually and thus is laborious or automated and expensive. To overcome this, recently, some research groups developed direct RT-PCR by omitting RNA extraction procedure (23-25). In this method, after the collection of patient material and deposition of potential SARS-CoV-2 viral particles in transport medium followed by the inactivation of the virus through detergent/chaotropic reagents or heating process step. Then, transfer the lysate to single-step RT-PCR format in which cDNA synthesis by RT and detection by qPCR may take place. Wee Sk et al. showed that direct RT-PCR has a high sensitivity of 6 RNA copies per reaction and is quantitative over a dynamic range of 7 orders of magnitude (25). Direct amplification of SARS-CoV-2 viral RNA from samples without RNA purification allows the reducing hands-on-time, time-to-results, and costs.

As per WHO guidelines, one of the following conditions should be met for considering a case as a NAAT-confirmed laboratory in areas with no circulation of SARS-CoV-2(18).

- 1) A positive NAAT result for at least two different targets on the SARS-CoV-2 virus genome, of which at least one target is preferably specific for SARS-CoV-2 virus using a validated assay;
- 2) One positive NAAT result for the presence of beta coronavirus, and SARS-CoV-2 virus further identified by sequencing partial or whole genome of the virus as long as the sequence target is larger or different from the amplicon probed in the NAAT assay used.

At the moment, it's important to identify that a negative result may not eliminate the possibility of COVID-19, it might be due to the poor-quality specimens, early or late collection, inadequate sample, and incorrect test procedures. When a patient with a high level of suspicion obtains a negative result for SARS-CoV-2 virus infection, especially when only upper respiratory tract specimens have been collected, additional specimens should be collected and tested, including, where possible, from the lower respiratory tract (26).

### **3.5 LOOP-MEDIATED ISOTHERMAL AMPLIFICATION**

Isothermal amplification depended nucleic acid tests are currently under progression for SARS-CoV-2. Recently a few studies reported the development of reverse transcription LAMP (RT-LAMP) tests(27–29) and some are clinically tested for SARS-CoV-2(30,31).

Primarily RT-LAMP is based on the DNA polymerase and 4-6 primers to bind at distinct regions on the target genome. RT-LAMP is a highly specific method since it uses a greater number of primers, like two inner primers and two outer primers on different regions on the genome. In

LAMP diagnostic tests, SARS-CoV-2 family genes such as ORF1ab, spike (S), envelope (E) or/and N gene can be targeted, and the procedure will be done in a single step at 63 °C isothermal conditions, and within 15-40 minutes the results will be obtained (27,28,30,31). For the POCT of SARS-CoV-2, many institutes are keen to implement isothermal nucleic acid amplification technology, eliminating the need for a highly costly thermal cycler. The most promising alternative to PCR may be loop-mediated isothermal amplification (LAMP) because it provides many advantages in terms of precision, sensitivity, reaction efficiency and product yield. Recently, a reverse transcription (RT)-LAMP assay targeting non-structural protein 3 (Nsp3) for SARS-CoV-2 detection was developed by Park et al., whose LOD was 100 copies per reaction (32). Similarly, RT-LAMP assay within 60 min targeting an ORF1ab and the S gene, whereby the LOD was 20 copies/reaction and 200 copies/reaction, was prepared by Yan et al. (33).

### **3.6 CRISPR BASED METHODS**

Along with isothermal amplification, another category of nucleic acid tests that could be used for SARS-CoV-2 detection based on dyes employing inherent by-products of comprehensive DNA synthesis, such as calcein, malachite orange, and hydroxynaphthol blue can be utilised for performing visual detection methods. Clustered regularly interspaced short palindromic repeats (CRISPR) based diagnostic tests have been developed for point-of-care nucleic acid detection (34), such as Specific High-sensitivity Enzymatic Reporter unlocking (**SHERLOCK**) or DNA Endonuclease-Targeted CRISPR Trans Reporter (**DETECTR**).

CRISPR based method nucleic acid tests mainly in a combination of Recombinase Polymerase Amplification with CRISPR–Cas enzymology for specific recognition of targeted RNA or DNA

sequences (34). In SHERLOCK testing strategy is based on Cas13a ribonuclease for RNA sensing (35). Recently, studies have reported the development and evaluation of a CRISPR based Diagnostic For 2019-Novel Coronavirus (36). Similar to the SHERLOCK method, CRISPR–Cas12-based assay was developed termed as DETECTR (DNA Endonuclease-Targeted CRISPR Trans Reporter). Broughton et al. reported the development and initial validation of a CRISPR–Cas12-based assay for detection of SARS-CoV-2 from extracted patient sample RNA (37). In addition to that, Broughton et al. compared the detection strategies of DETECTR, and the RT-qPCR which is recommended by CDC/WHO for SARS-CoV-2 detection, however, they found that the limit of detection these methods is ten copies/ $\mu\text{L}$ , 1 or 3.2 copies/ $\mu\text{L}$  input sample, respectively. Also, the assays turnaround time is 45 min and four hours, respectively (37). Since less time consumption and equipment requirement, these methods can be set up in emergency departments and local community hospitals. Recently, Hou et al. exploited polymerase mediated amplification by the combination of recombinase polymerase amplification (RPA) and CRISPR-Cas13-mediated enzymatic signal amplification for detection of SARS-CoV-2 with high sensitivity and 7.5 copies/reaction within 40 min. The CRISPR-Cas13-based assay has a higher detection potential than the RT-PCR assay, according to a comparative clinical study. (38). In another study, Ding et al. developed the protocol by integrating RT-RPA and CRISPR-based detection in a one-pot reaction and incubating at a single temperature (39). This “All-In-One Dual CRISPR-Cas12a” (AIOD-CRISPR) assay detected as little as 4.6 SARS-CoV-2 RNA copies per  $\mu\text{L}$  input at 40 minutes per  $\mu\text{L}$  input.

### **3.7 SEROLOGY TESTING**

It is emphasised that nucleic acid-based testing methods need to extract nucleic acid in

advance, the requirement of trained technicians, complex operation, expensive equipment; it is complicated to do in epidemiological and surveillance purposes. With the aid of viral protein antigen and antibodies which are produced in response to a SARS-CoV-2 infection can be used for diagnosis. Since variations in the viral load throughout infection, it may difficult to detect the viral proteins. In contrast to this, the detection of antibodies which are generated to viral proteins may enable the indirect ways to detect SARS-CoV-2.

Serology testing involves the screening test by qualitative assays and measurement of different classes of immunoglobulins (IgA, IgM, IgG) against SARS-CoV-2 by using quantitative assays for establishing whether a person has been infected by SARS-CoV-2. Zhang et al. detected immunoglobulin G and M (IgG and IgM) from the human serum of COVID-19 patients using an enzyme-linked immunosorbent assay (40). Although recent reports suggesting that detection of antibody-based methods targeted to IgM and IgG by using recombinant N and S proteins of SARS-CoV-2 are consistent with the results obtained by real-time RT-PCR (41–43). In addition to this, the receptor-binding domain (RBD) of the viral S protein presented a better antigenicity than viral N protein in the diagnosis of SARS-CoV-2 infection (44). Also, IgA levels in patient serum have positively correlated with the severity of SARS-CoV-2 infection, signifying that serum IgA can be used as a biological marker (44). In clinical diagnosis, the IgA and IgM antibodies against viral proteins can be detected seven days after SARS-CoV-2 infection or within 3-4 days after symptoms appear, as well as for IgG antibodies appears in 7-10 days later SARS-CoV-2 infection.

Serology testing has some advantages over other techniques, apart from being inexpensive. The primary application of serology testing is to identify individuals who previously

had SARS-CoV-2 infections. This knowledge can be used to guide studies of epidemiology and seroprevalence, and to facilitate contact tracing. Serology tests can also be used to determine possible convalescent donors of plasma and to assess the immune response to candidate vaccines. Finally, serology tests can also aid in diagnosing Covid-19 in patients with clinical suspicion but having repeated RT-PCR-negative results (45,46). Serology testing has its limitations too. The serology test cannot be used to diagnose acute or recent COVID-19 cases. Antibody tests for COVID-19 may also interact with other pathogens, including other human coronaviruses and leads to false-positive results. Based on current data, the WHO does not recommend the use of antibody-detecting rapid diagnostic tests for patient care but encourages the continuation of work to establish their usefulness in disease surveillance and epidemiologic research (14).

Serology testing helps in the assessment of seroprevalence of COVID-19 disease in the community. Nationwide serology testing would help in tailoring the public health measures to control and avoid renewed COVID-19 epidemic wave (47). Serologic surveillance also can help in anticipate and modify treatment modalities as in perinatal clinical practices pregnant women (48). Seroprevalence surveys can also help in understand the geographical profile of the COVID-19 disease and help in creating a regional level approach in controlling the pandemic (49). However, serology testing cannot be used to determine the infectivity status or the susceptibility to reinfection for the patient. The presence of antibodies does not render the patient non-infectious, as the antibodies can be of non-neutralising in nature (50,51). Virus neutralisation tests have to be performed to assess the neutralising capability of antibodies generated by the body against the SARS-CoV-2 virus. Hence clinical laboratories

are recommended not to promote so-called “immunity passports” due to a lack of evidence for the neutralising capability of antibodies (52).

### **3.8 VIRAL SEQUENCING**

Sequencing does not play a part in the initial SARS-CoV-2 laboratory diagnosis but can be beneficial in the following circumstances; 1) Provides evidence of virus existence; 2) Monitoring for viral genome mutations that could affect medical countermeasure performance, including diagnostic testing; 3) Virus sequencing of entire genomes can also inform studies on molecular epidemiology. Virus isolation, currently, is not recommended as part of the routine diagnostic methodology.

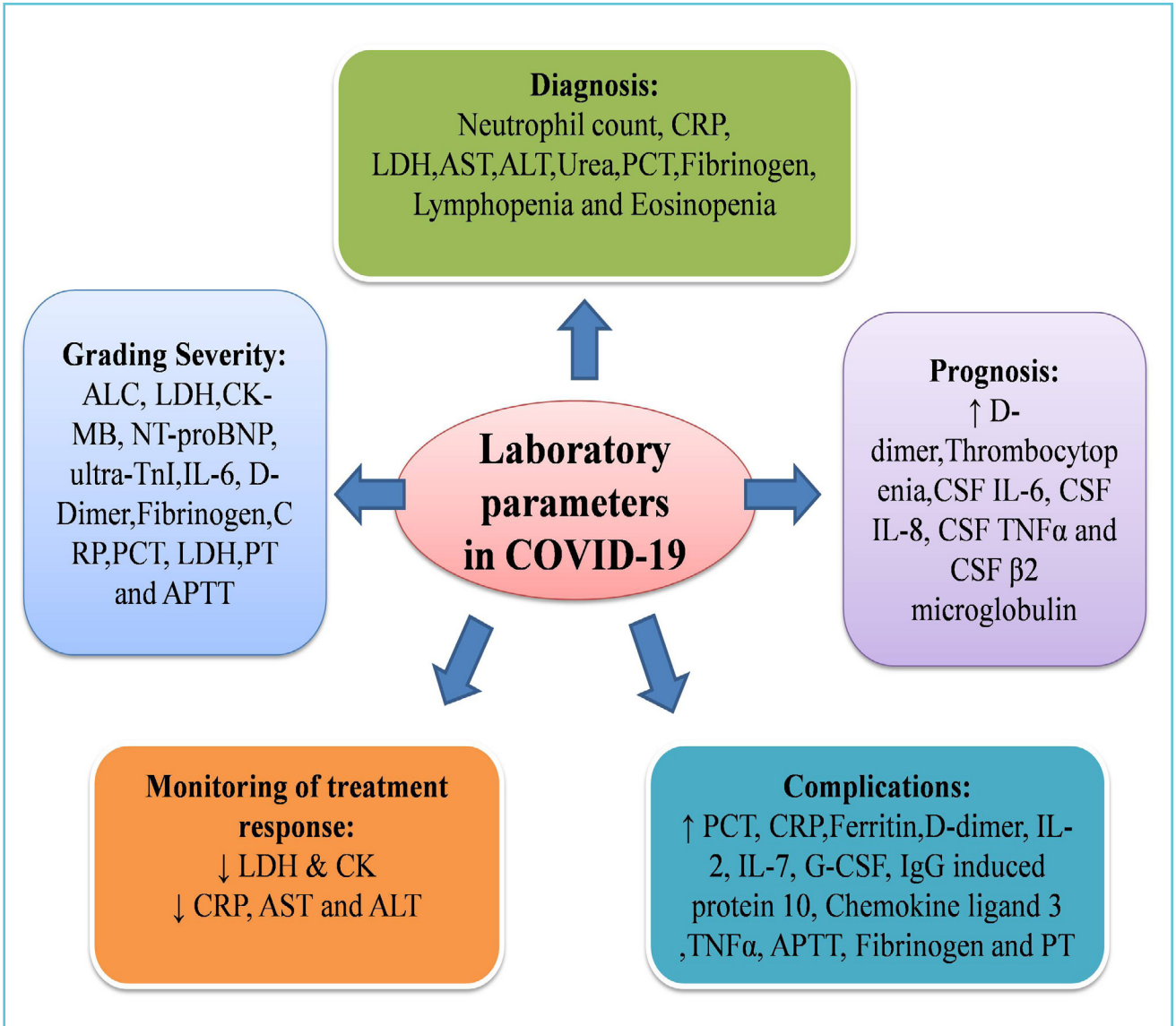
### **4.1 CLINICAL LABORATORY PARAMETERS IN COVID-19**

Biochemical and haematological parameters have been investigated to assess their role in diagnosis and prognosis. Further, the role of laboratory testing in assessing severity and selecting treatment modalities and monitoring the effectiveness of treatment has been elucidated through multiple studies. Figure 2 depicts the important parameters that can be used for determining diagnosis, prognosis and treatment response.

### **4.2 LABORATORY PARAMETERS AIDING IN DIAGNOSIS OF COVID-19**

Several Laboratory Parameters are significantly increased in COVID-19 positive patients when compared with others. RT-PCR diagnosed COVID-19 patients had significantly higher neutrophil (NEU) count, and C-reactive protein (CRP), aspartate aminotransferase, alanine aminotransferase (ALT), lactate dehydrogenase and Urea levels in serum(53). Serum albumin levels and White blood cell (WBC) count are decreased

**Figure 2** Important parameters used for determining diagnosis, prognosis and treatment response



in COVID-19 positive patients when compared to others (53). More number of control patients had a higher procalcitonin (PCT) level of more than 0.5 ng/ml than that of COVID-19 patients (54). At admission, the COVID-19 patients showed elevated levels of fibrinogen than the control group. Further, a greater percentage of COVID-19 patients had fibrinogen levels >400 mg/dL compared to the control group (55). Normal or decreased number of leukocytes,

lymphopenia, eosinopenia, and elevated hs-CRP were presented in COVID-19 patients when compared with controls. It has been found that the use of eosinopenia alone or the combination of eosinopenia and elevated hs-CRP improves the predictive capacity for the detection of COVID-19 patients (56). Table 2 depicts the Laboratory parameters assessed to distinguish COVID-19 Positive patients from Negative patients.

**Table 2** Laboratory parameters in COVID-19 positive and negative patients

Study	Sample size	Parameter	Remarks
Mardani et al. (53)	Two hundred cases RT-PCR for COVID-19 was positive in 70	Neutrophil (NEU) count, C-reactive protein (CRP), Aspartate aminotransferase, alanine aminotransferase, Lactate dehydrogenase, Urea, Lower white blood cell (WBC) count, Lower serum albumin level	ALT (AUC = 0.879), NEU (AUC = 0.858), CRP (AUC = 0.870), LDH (AUC = 0.835), Urea (AUC = 0.835)
Chen et al (54)	78 COVID-19 patients 26 control patients	PCT	-
Di Micco et al. (55)	67 COVID-19 patients and 67 patients with non-COVID-19 acute respiratory illness	Increased levels of fibrinogen	-
Li et al. (56)	458	Normal or decreased number of leukocytes, lymphopenia, eosinopenia and elevated hs-CRP	Eosinopenia the sensitivity of 74.7% and specificity of 68.7%  Combination of eosinopenia and elevated hs-CRP showed a sensitivity of 67.9% and specificity of 78.2% (AUC=0.730).
Ferrari et al. (57)	207	WBC, AST, ALT, CRP, and LDH	For LDH cut off 210 U/L: positive or negative with PPV: 83.3% and NPV: 90.6%.
Liu et al. (58)	119	Presence of urine occult blood and proteinuria.  Lower urine specific gravity	-

### 4.3 LABORATORY PARAMETERS HELPING IN ASSESSING THE SEVERITY OF COVID-19

Multiple parameters are useful in assessing the severity of the disease. The parameters that were found to have a significant difference between mild and severe disease include interleukin-6 (IL-6), d-dimer (d-D), glucose, fibrinogen, thrombin time, and C-reactive protein (59). Fibrinogen was found to be higher in COVID-19 patients with SARS compared to those without SARS (55).

The role of laboratory parameters indicating inflammation have been discussed elsewhere (60). IL-6, an inflammatory cytokine, was found to have a potential value for monitoring the process of severe cases (61). The increased concentration of ultra-TnI, MYO, and NT-proBNP was also found to be associated with the severity of COVID-19 (62). The dysregulated activity of CD3<sup>+</sup> CD8<sup>+</sup> T lymphocytes, CD16<sup>+</sup> CD56<sup>+</sup> NK cells and altered C1q and IL-6 have been found to accentuate the severity of disease and death (63). Further, on correlation analysis between multiple cytokines and coagulation indicators

in critically ill COVID-19 patients, a high correlation was observed between IL-6 and the International normalised ratio (INR) (64).

The severity of lung abnormalities is quantified by chest imaging. Different laboratory parameters are associated with stages of lung diseases in COVID-19 patients as quantified on chest CT. Early-stage as per CT scoring was found to be correlated with the neutrophil count. The progressive stage was correlated with the neutrophil count, white blood cell count, C-reactive protein, procalcitonin, and lactate dehydrogenase. Contrastingly, peak and absorption stages were not correlated with any parameter (65). The paradoxical increase in D-dimer levels despite decreased fibrinolytic capacity had prompted the researchers to hypothesise that the major source of D-dimer could be the lungs (66).

Apart from altered coagulation profile, low activities of natural anticoagulants, increased factor VIII level and antiphospholipid antibodies presence have also been found to accentuate the severity of the disease in COVID-19 patients (67). In severe and critically ill patients, the

**Table 3** Laboratory parameters associated with severity of the disease in COVID-19 patients

Study	Sample size	Parameter	Category
Fan et al. (53)	Between the ICU (n=9) and non-ICU (n=58) patients	ALC and LDH, ALC and Absolute Monocyte Count (AMC) nadir	ICU vs Non-ICU
Han et al. (62)	mild (198 cases), severe (60 cases) and critical (15 cases)	CK-MB, MYO, ultra-TnI and NT-proBNP	Severity and case fatality



Gao et al. (59)	43 COVID-19 patients mild group (28 patients) and severe group (15 patients).	Interleukin-6 (IL-6), d-dimer (d-D), glucose, thrombin time, fibrinogen, and C-reactive protein IL 6 (AUC=0.795) D-Dimer (AUC=0.75) Glu, TT, CRP and FIB (AUC<0.75) IL 6 + D-Dimer (AUC=0.84)	Mild vs severe
Zhang et al. (63)	84 COVID-19 patients	Early stage: neutrophil count . Progressive stage: neutrophil count, white blood cell count, C-reactive protein, procalcitonin, lactate dehydrogenase.	early and progressive stages of lung abnormalities (CT Finding)
Tan et al. (64)	27 COVID-19 and 75 Flu patients	Progression and the peak stages: lymphocytes decreased Initial and progression stages: C-reactive protein (CRP) higher. Correlation analysis showed that CRP, erythrocyte sedimentation rate and granulocyte/lymphocyte ratio were positively associated with the CT severity scores. CRP (AUC=0.87) at 20.42 mg/L cut-off, with sensitivity and specificity 83% and 91%, respectively.	Mild vs Severe
Di Micco et al. (55)	SARS: 24 Without SARS: 43	Fibrinogen	Mild vs Severe
Fu et al. (65)	75	WBC, NLR, D-dimer, and fibrinogen levels Increased. Lymphocyte level Decreased. AUC is NLR (0.88), AUC of D-dimer and fibrinogen was 0.74, and AUC of lymphocyte and PCT were 0.72 and 0.67 respectively.	mild/moderate COVID-19 group
Zhu et al. (61)	127 16 severe cases	High level of interleukin-6 (IL-6), C-reaction protein (CRP). The area under the ROC curve was 0.835 for IL-6, sensitivity was 87.50%, specificity was 74.77%.	severity of COVID-19

Liu et al. (58)	119	The positive rates of urine glucose (GLU-U) and PRO in the severe and critical groups were higher	mild/moderate COVID-19 group
Tang et al. (71)	183	Higher D-dimer and FDP levels, longer PT and APTT	Between survivors and non-survivors

specific immunoglobulin G antibodies to the SARS-CoV-2 were found to be significantly low when compared with patients with mild disease (68).

Different parameters have been assessed for their dynamic trend in different stages as well as the severity of the disease. Lymphocytes in the severe COVID-19 were found to be progressively decreasing at the progression and the peak stages. C-reactive protein (CRP) was higher in the severe group at the initial and progression stages than those in the mild group (64). Table 3 depicts the laboratory parameters associated with severity of the disease in COVID -19 patients.

#### 4.4 LABORATORY PARAMETERS INDICATING PROGNOSIS OF THE DISEASE IN COVID-19 PATIENTS

Laboratory parameters at admission have been investigated for their prognostic power for the severity of the disease. Logistic regression analysis showed that IL-6 and D-Dimer could be important predictors in the severity of COVID-19. Further, it had also been found that combined detection using IL-6 and D-Dimer was more efficient than independent detection (59). Various parameters have also been used to predict admission to ICU. ALC and LDH stood out as parameters that can, with the levels at admission, reliably predict the admission of the patient to ICU (53). The change of neutrophil to lymphocyte

ratio (NLR) and D-dimer level has been found to help in discriminating severe COVID-19 cases from mild/moderate ones on consequent days after admission (65). The early increase in Fibrinogen in COVID-19 patients makes it a good risk stratification marker for the early detection of a subgroup of COVID-19 patient at increased risk to develop SARS (55).

Non-survivors mainly presented with laboratory abnormalities of serious inflammation response and multiple organ failure, manifesting as high levels of cytokines and deranged coagulation parameters. Neutrophil count, hypersensitivity C-reactive protein, creatine kinase, and blood urea nitrogen were identified to help in early detection of COVID-19 severe patients with poor outcomes on admission (72). Further, the non-survivors of COVID-19 disease revealed significantly higher D-dimer and FDP levels compared to survivors on admission (71). Hence, the use of Sepsis-induced coagulopathy scoring system for early assessment and management have been advised in patients with the critical disease (73). mRNA clearance rates indicate the resolution of the disease. It has been found that the decline of serum creatine kinase (CK) and lactate dehydrogenase (LDH) levels significantly correlated with mRNA clearance rates (74). CSF analyses revealed relatively slightly increased levels of interleukin 6 (IL-6), interleukin 8, tumour necrosis factor-alpha and  $\beta$ 2-microglobulin. Ten days after the admission, CSF IL-8 and TNF- $\alpha$  decreased, whereas IL-6

**Table 4** Laboratory parameters determining prognosis in COVID-19 patients

Study	Sample size	Parameter
Li et al. (75)	279	The higher D-dimer levels on admission progressively improved only in the mild disease group.
Liu et al. (77)	383	Thrombocytopenia. An increment of per $50 \times 10^9/L$ in platelets was associated with a 40% decrease in mortality (hazard ratio: 0.60, 95% CI: 0.43, 0.84).

and  $\beta$ 2-microglobulin values were stable (76). Table 4 depicts the important laboratory parameters that can be used to determine the prognosis in COVID-19 patients.

#### 4.5 LABORATORY PARAMETERS IN COMORBIDITIES AND TREATMENT IN COVID-19 PATIENTS

Various laboratory parameters have been assessed for its role in complications in COVID-19 patients. Patients with abnormal liver function had higher levels of procalcitonin and C-reactive protein(78). Various inflammatory markers are elevated in patients with COVID-19

related cardiac injury. They include C- reactive protein (CRP), procalcitonin, ferritin, D- dimer, Interleukin - 2 (IL-2) interleukin – 7 (IL-7), granulocyte – colony-stimulating factor, IgG- induced protein 10, chemokine ligand three and tumour necrosis alpha(79). Lymphocyte counts, activated partial thromboplastin time (APTT) and D-dimer was found to be different in patients with venous thromboembolism when compared with the non-VTE group. The significant increase of D-dimer observed in severe patients makes it a good index for identifying high-risk groups of VTE(80). On comparison of COVID-19 patients with and without HBV co-infection, although the level of liver function parameters

**Table 5** Laboratory parameters associated with complications and response to treatment in COVID-19 patients

Study	Sample size	Parameter	Remarks	Category
Yuan et al. (74)	94 COVID-19 patients	Decline in Serum LDH or CK	-	Response to treatment
Fan et al. (78)	148	Increased levels of procalcitonin and C-reactive protein	-	Abnormal liver function
Han et al. (79)	273	Increased levels of CK-MB, MYO, ultra-Tnl, and NT-proBNP	-	COVID-19 related cardiac injury

Duan et al. (83)	10	Lymphocytopenia tended to be improved after CP transfusion. Decreased in C-reactive protein (CRP), alanine amino-transferase and aspartate aminotransferase.	-	Convalescent plasma therapy
Cui et al. (80)	81	Lymphocyte counts, activated partial thromboplastin time (APTT), D-dimer	D-dimer cut-off 1.5 µg/mL for predicting VTE had a sensitivity of 85.0%, the specificity of 88.5%, and the negative predictive value (NPV) of 94.7%.	VTE /NonVTE group
Wright et al (84)	44	Elevated D-dimer, fibrinogen, PT, and PTT	A D-Dimer cutoff of 2600 ng/ml predicted need for dialysis with an AUROC of .779	Predict thromboembolic outcomes and new-onset renal failure
Lin et al. (85)	137	CD8+ T cells	HR=2.376	Duration of SARS-CoV-2 viral positivity

showed no differences, prealbumin levels were found to be lower in HBsAg+ patients(81). In solid organ transplant recipients with COVID-19, a biphasic pattern was observed with initial increases in inflammatory markers, followed by an increase in WBC, CRP, ferritin and D-dimer (82).

To assess the efficacy of treatment, the primary tool for analysis have been the trend shown by Laboratory parameters. Lymphocytopenia improved after Convalescent Plasma transfusion. C-reactive protein (CRP), alanine aminotransferase, and aspartate aminotransferase decreased after treatment (83). Table 5 depicts the laboratory parameters which are associated with

complications and monitoring of response to treatment in COVID-19 patients.

#### 4.6 LABORATORY PARAMETERS AND BODY FLUIDS IN COVID-19

Different fluids have also been assessed in COVID-19 patients for different parameters. The presence of urine occult blood and proteinuria were found to be higher in COVID-19 patients than in healthy controls, whereas urine specific gravity was found to be lower in patients than in healthy controls. The presence of urine glucose and proteinuria were higher in the severe and critical groups when compared with that of

the moderate group (58). CSF analyses have revealed relatively slightly increased levels of interleukin 6 (IL-6), interleukin 8, tumour necrosis factor-alpha, and  $\beta$ 2-microglobulin in a single patient (76).

## **5. POOLED AND META-ANALYSIS OF LABORATORY PARAMETERS IN COVID-19**

Multiple meta-analyses had been undertaken to find the significance of various laboratory parameters in COVID-19. Soraya et al. had concluded thrombocyte count to have a crucial role in the diagnosis and prognosis of COVID-19. Further, lymphocyte count, D-dimer and CRP levels helped to assess the severity of the disease (86). Henry et al. had observed that markers of inflammation, coagulation markers and organ damage to be significantly elevated severe and fatal COVID-19 patients. In patients with severe disease, interleukins 6 (IL-6) and 10 (IL-10) and serum ferritin were found to be predominantly elevated (87). Interestingly, in a pooled analysis of Laboratory Parameters paediatric COVID-19 patients, contrary to adult patients, leukocyte indices showed inconsistent trends (88). The elevated levels of the neutrophil count, D-dimer, prothrombin time (PT), fibrinogen erythrocyte sedimentation rate, procalcitonin, IL-6, and IL-10 were found to be better predictors for severe COVID-19 disease (89, 90). Further, high IL-6, CRP, D-dimer and neutrophils were found to be better predictors of mortality in COVID-19 (89). A similar meta-analysis also observed severe or critical COVID-19 to be associated with innate immune response and tissue damage (91).

## **6. CONCLUSION**

In summary, the crucial role that clinical laboratory plays in the management of diseases has never been more evident than today. Validating

various assays for diagnosing COVID-19 helps in early diagnosis and initiation of treatment as well as prevent transmission. The assessment of the clinical utility of tests in different scenarios in COVID-19 and ensuring its accuracy adds to the efforts to treatment of the disease as well as predicting complications. This review has emphasised the importance of laboratory in the COVID-19 crisis. The emergence of diagnostic assays with better sensitivity and specificity equips the laboratories with an enhanced ability to identify COVID-19 cases early and prevents transmission (92). The routine laboratory parameters have been shown to be able to distinguish between positive and negative patients, have the capacity to predict prognosis & complications and have usefulness in monitoring treatment response. Further studies in this arena would lead to validation of better assays for precise diagnosis and newer biomarkers for monitoring treatment and disease progression. Decision-makers should not underestimate the role of the laboratory as it plays a pivotal role in patient-centred and sustainable future of health care.



### **Funding**

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.



## **REFERENCES**

1. Zhou P, Yang X-L, Wang X-G, Hu B, Zhang L, Zhang W, et al. A pneumonia outbreak associated with a new coronavirus of probable bat origin. *Nature*. 2020 Mar;579(7798):270–3.
2. Wu JT, Leung K, Leung GM. Nowcasting and forecasting the potential domestic and international spread of the 2019-nCoV outbreak originating in Wuhan, China: a modelling study. *The Lancet*. 2020 Feb;395(10225):689–97.

3. WHO Director-General's opening remarks at the media briefing on COVID-19.
4. Specific Primers and Probes for Detection 2019 Novel Coronavirus; China National Institute For Viral Disease Control and Prevention: Beijing, 2020.
5. Detection of 2019 Novel Coronavirus (2019-nCoV) in Suspected Human Cases by RT-PCR; School of Public Health, Hong Kong University: Hong Kong, 2020.
6. Corman VM, Landt O, Kaiser M, Molenkamp R, Meijer A, Chu DK, et al. Detection of 2019 novel coronavirus (2019-nCoV) by real-time RT-PCR. *Eurosurveillance* [Internet]. 2020 Jan 23 [cited 2020 May 31];25(3). Available from: <https://www.eurosurveillance.org/content/10.2807/1560-7917.ES.2020.25.3.2000045>
7. Naganori, N.; Shirato, K.; et al. Detection of Second Case of 2019-nCoV Infection in Japan; Department of Virology III, National Institute of Infectious Diseases, Japan, 2020.
8. Diagnostic Detection of Novel Coronavirus 2019 by Real Time RT-PCR; Department of Medical Sciences, Ministry of Public Health, Thailand, 2020.
9. CDC 2019-Novel Coronavirus (2019-nCoV) Real-Time RT-PCR Diagnostic Panel; Division of Viral Diseases, U.S. Centers for Disease Control and Prevention: Atlanta, GA, 2020.
10. Coronavirus Disease 2019 (COVID-19); Centers for Disease Control and Prevention.
11. Miller S, Chiu C, Rodino KG, Miller MB. Point-Counterpoint: Should We Be Performing Metagenomic Next-Generation Sequencing for Infectious Disease Diagnosis in the Clinical Laboratory? Ledebner N, editor. *J Clin Microbiol* [Internet]. 2019 Oct 16 [cited 2020 May 31];58(3). Available from: <https://jcm.asm.org/content/58/3/e01739-19>
12. Report of the WHO-China Joint Mission on Coronavirus Disease 2019 (COVID-19); WHO: Geneva, Switzerland, 2020.
13. Lu R, Zhao X, Li J, Niu P, Yang B, Wu H, et al. Genomic characterisation and epidemiology of 2019 novel Coronavirus: implications for virus origins and receptor binding. *The Lancet*. 2020 Feb;395(10224):565–74.
14. World Health Organization (WHO) (2020) Advice on the use of point-of-care immunodiagnostic tests for COVID-19 <https://www.who.int/news-room/commentaries/detail/advice-on-the-use-of-point-of-care-immunodiagnostic-tests-for-covid-19>.
15. Centers for Disease Prevention and Control (CDC) (2020) Interim guidelines for collecting, handling, and testing clinical specimens from persons for coronavirus disease 2019 (COVID-19). <https://www.cdc.gov/coronavirus/2019-nCoV/lab/guidelines-clinical-specimens.html>.
16. Indian Council of Medical Research (ICMR) (2020) [https://icmr.nic.in/sites/default/files/upload\\_documents/Validation\\_of\\_Commercial\\_Kits\\_02042020.pdf](https://icmr.nic.in/sites/default/files/upload_documents/Validation_of_Commercial_Kits_02042020.pdf).
17. Rodino KE, Buckwalter SP, Walchak RC, Germer JJ, Fernholz E, Boerger A, Schuetz AN, Yao JD, Binnicker MJ. 30 March 2020. Evaluation of saline, phosphate-buffered saline, and minimum essential medium as potential alternatives to viral transport media for SARS-CoV-2 testing. *J Clin Microbiol* doi:10.1128/JCM.00590-20.
18. Perchetti GA, Huang ML, Peddu V, Jerome KR, Greninger AL.J. Stability of SARS-CoV-2 in Phosphate-Buffered Saline for Molecular Detection. *Clin Microbiol*. 2020 Jul 23;58(8):e01094-20. doi: 10.1128/JCM.01094-20. Print 2020 Jul 23.
19. Wang W, Xu Y, Gao R, Lu R, Han K, Wu G, Tan W. Detection of SARS-CoV-2 in Different Types of Clinical Specimens. *JAMA*. 2020 May 12;323(18):1843-1844. doi: 10.1001/jama.2020.3786. PMID: 32159775; PMCID: PMC7066521.
20. Chu DKW, Pan Y, Cheng SMS, Hui KPY, Krishnan P, Liu Y, Ng DYM, Wan CKC, Yang P, Wang Q, Peiris M, Poon LLM. Molecular Diagnosis of a Novel Coronavirus (2019-nCoV) Causing an Outbreak of Pneumonia. *Clin Chem*. 2020 Apr 1;66(4):549-555. doi: 10.1093/clinchem/hvaa029.
21. Chantal B F et al. Analytical sensitivity and efficiency comparisons of SARS-CoV-2 RT-qPCR primer-probe sets. *Nat Microbiol*. 2020 Oct;5(10):1299-1305. doi: 10.1038/s41564-020-0761-6. Epub 2020 Jul 10.
22. R. Konrad, U. Eberle, A. Dangel, B. Treis, A. Berger, K. Bengs, V. Fingerle, B. Liebl, N. Ackermann, A. Sing, Rapid establishment of laboratory diagnostics for the novel coronavirus SARS-CoV-2 in Bavaria, Germany, February 2020, *Euro Surveill* 25 (9) (2020)
23. Smyrliki I et al, Massive and rapid COVID-19 testing is feasible by extraction-free SARS-CoV-2 RT-PCR. *Nature Communications* volume 11, Article number: 4812 (2020)
24. Morehouse, Z.P., Proctor, C.M., Ryan, G.L. et al. A novel two-step, direct-to-PCR method for virus detection off swabs using human coronavirus 229E. *Virology* 17, 129 (2020). <https://doi.org/10.1186/s12985-020-01405-y>
25. Wee SK, Sivalingam SP, Yap EPH. Rapid Direct Nucleic Acid Amplification Test without RNA Extraction for SARS-CoV-2 Using a Portable PCR Thermocycler. *Genes*. 2020 Jun;11(6). DOI: 10.3390/genes11060664.
26. Laboratory testing for coronavirus disease (COVID-19) in suspected human cases : WHO Interim guidance 19 March 2020.
27. Lamb LE, Bartolone SN, Ward E, Chancellor MB. Rapid Detection of Novel Coronavirus (COVID-19) by

Reverse Transcription- Loop-Mediated Isothermal Amplification. :17.

28. Yu L, Wu S, Hao X, Dong X, Mao L, Pelechano V, et al. Rapid Detection of COVID-19 Coronavirus Using a Reverse Transcriptional Loop-Mediated Isothermal Amplification (RT-LAMP) Diagnostic Platform. *Clin Chem [Internet]*. 2020 May 12 [cited 2020 May 31]; Available from: <https://academic.oup.com/clinchem/advance-article/doi/10.1093/clinchem/hvaa102/5823294>

29. Zhang Y, Odiwuor N, Xiong J, Sun L, Nyaruaba RO, Wei H, et al. Rapid Molecular Detection of SARS-CoV-2 (COVID-19) Virus RNA Using Colorimetric LAMP [Internet]. *Infectious Diseases (except HIV/AIDS)*; 2020 Feb [cited 2020 May 31]. Available from: <http://medrxiv.org/lookup/doi/10.1101/2020.02.26.20028373>

30. Yan C, Cui J, Huang L, Du B, Chen L, Xue G, et al. Rapid and visual detection of 2019 novel coronavirus (SARS-CoV-2) by a reverse transcription loop-mediated isothermal amplification assay. *Clin Microbiol Infect*. 2020 Jun;26(6):773–9.

31. Lu R, Wu X, Wan Z, Li Y, Jin X, Zhang C. A Novel Reverse Transcription Loop-Mediated Isothermal Amplification Method for Rapid Detection of SARS-CoV-2. *Int J Mol Sci*. 2020 Apr 18;21(8):2826.

32. Park G.S., Ku K., Baek S.H., Kim S.J., Kim S.I., Kim B.T., Maeng J.S. Development of reverse transcription loop-mediated isothermal amplification assays targeting SARS-CoV-2. *J. Mol. Diagn.: J. Mod. Dynam*. 2020;22(6):729–735.

33. Yan C., Cui J., Huang L., Du B., Chen L., Xue G., Li S., Zhang W., Zhao L., Sun Y., Yao H., Li N., Zhao H., Feng Y., Liu S., Zhang Q., Liu D., Yuan J. Rapid and visual detection of 2019 novel coronavirus (SARS-CoV-2) by a reverse transcription loop-mediated isothermal amplification assay. *Clin. Microbiol. Infect*. 2020;26(6):773–779.

34. Gootenberg JS, Abudayyeh OO, Lee JW, Essletzbichler P, Dy AJ, Joung J, et al. Nucleic acid detection with CRISPR-Cas13a/C2c2. *Science*. 2017 Apr 28;356(6336):438–42.

35. Kellner MJ, Koob JG, Gootenberg JS, Abudayyeh OO, Zhang F. SHERLOCK: nucleic acid detection with CRISPR nucleases. *Nat Protoc*. 2019 Oct;14(10):2986–3012.

36. Hou T, Zeng W, Yang M, Chen W, Ren L, Ai J, et al. Development and Evaluation of A CRISPR-based Diagnostic For 2019-novel Coronavirus [Internet]. *Infectious Diseases (except HIV/AIDS)*; 2020 Feb [cited 2020 May 31]. Available from: <http://medrxiv.org/lookup/doi/10.1101/2020.02.22.20025460>

37. Broughton JP, Deng X, Yu G, Fasching CL, Servellita V, Singh J, et al. CRISPR–Cas12-based detection of SARS-CoV-2. *Nat Biotechnol [Internet]*. 2020 Apr 16 [cited 2020 May 31]; Available from: <http://www.nature.com/articles/s41587-020-0513-4>

38. Hou T, Zeng W, Yang M, Chen W, Ren L, Ai J, Wu J, Liao Y, Gou X, Li Y, Wang X, Su H, Gu B, Wang J, Xu T Development and evaluation of a rapid CRISPR-based diagnostic for COVID-19.

*PLoS Pathog*. 2020 Aug 27;16(8):e1008705. doi: 10.1371/journal.ppat.1008705. eCollection 2020 Aug.

39. Xiong Ding 1, Kun Yin 1, Ziyue Li 1, Rajesh V Lalla 2, Enrique Ballesteros 3, Maroun M Sfeir 3, Changchun Liu 4 Ultrasensitive and visual detection of SARS-CoV-2 using all-in-one dual CRISPR-Cas12a assay. *Nat Commun* . 2020 Sep 18;11(1):4711. doi: 10.1038/s41467-020-18575-6.

40. Zhang W, Du R-H, Li B, Zheng X-S, Yang X-L, Hu B, et al. Molecular and serological investigation of 2019-nCoV infected patients: implication of multiple shedding routes. *Emerg Microbes Infect*. 2020 Jan 1;9(1):386–9.

41. Zhong L, Chuan J, Gong B, Shuai P, Zhou Y, Zhang Y, et al. Detection of serum IgM and IgG for COVID-19 diagnosis. *Sci China Life Sci*. 2020 May;63(5):777–80.

42. Jia X, Zhang P, Tian Y, Wang J, Zeng H, Wang J, et al. Clinical significance of IgM and IgG test for diagnosis of highly suspected COVID-19 infection [Internet]. *Infectious Diseases (except HIV/AIDS)*; 2020 Mar [cited 2020 May 31]. Available from: <http://medrxiv.org/lookup/doi/10.1101/2020.02.28.20029025>

43. Evaluations of serological test in the diagnosis of 2019 novel coronavirus (SARS-CoV-2) infections during the COVID-19 outbreak. :16.

44. Ma H, Zeng W, He H, Zhao D, Yang Y, Jiang D, et al. COVID-19 diagnosis and study of serum SARS-CoV-2 specific IgA, IgM and IgG by chemiluminescence immunoanalysis [Internet]. *Infectious Diseases (except HIV/AIDS)*; 2020 Apr [cited 2020 May 31]. Available from: <http://medrxiv.org/lookup/doi/10.1101/2020.04.17.20064907>

45. L. Guo, et al., Profiling early humoral response to diagnose novel coronavirus disease (COVID-19), *Clin. Infect. Dis.* (2020).

46. R. Zhao, et al., Early detection of SARS-CoV-2 antibodies in COVID-19 patients as a serologic marker of infection, *Clin. Infect. Dis.* (2020).

47. Pollán M, Pérez-Gómez B, Pastor-Barriuso R, Oteo J, Hernán MA, Pérez-Olmeda M, et al. Prevalence of SARS-CoV-2 in Spain (ENE-COVID): a nationwide, population-based seroepidemiological study. *The Lancet [Internet]*. 2020 Jul [cited 2020 Jul 21]; Available from: <https://linkinghub.elsevier.com/retrieve/pii/S0140673620314835>

48. Flannery DD, Gouma S, Dhudasia MB, et al. SARS-CoV-2 Seroprevalence Among Parturient Women. *Preprint. medRxiv*. 2020;2020.07.08.20149179. Published 2020 Jul 10. doi:10.1101/2020.07.08.20149179

49. Takita M, Matsumura T, Yamamoto K, Yamashita E, Hosoda K, Hamaki T, et al. Geographical Profiles of COVID-19 Outbreak in Tokyo: An Analysis of the Primary Care Clinic-Based Point-of-Care Antibody Testing. *J Prim Care Community Health*. 2020 Jan;11:215013272094269.
50. Wölfel, et al., Virological assessment of hospitalised patients with COVID-2019, *Nature* 581 (2020) 465–469.
51. B. Atkinson, E. Petersen, SARS-CoV-2 shedding and infectivity, *Lancet* 395 (2020) 1339–1340.
52. Bailey D, Konforte D, Barakauskas VE, Yip PM, Kulasingam V, Abou El Hassan M, Beach LA, Blasutig IM, Catomeris P, Dooley KC, Gong Y, Kavsak P, Randell EW, Robinson JL, Shaw J, Taher J, White-AI Habeeb N. Canadian society of clinical chemists (CSCC) interim consensus guidance for testing and reporting of SARS-CoV-2 serology. *Clin Biochem*. 2020 Oct 6:S0009-9120(20)30844-4.
53. Fan BE, Chong VCL, Chan SSW, Lim GH, Lim KGE, Tan GB, et al. Hematologic parameters in patients with COVID-19 infection. *Am J Hematol* [Internet]. 2020 Jun [cited 2020 May 25];95(6). Available from: <https://onlinelibrary.wiley.com/doi/abs/10.1002/ajh.25774>
54. Chen X, Yang Y, Huang M, Liu L, Zhang X, Xu J, et al. Differences between COVID-19 and suspected then confirmed SARS-CoV-2-negative pneumonia: a retrospective study from a single center. *J Med Virol* [Internet]. 2020 Apr 1 [cited 2020 May 25]; Available from: <http://doi.wiley.com/10.1002/jmv.25810>
55. Di Micco P, Russo V, Carannante N, Imperato M, Rodolfi S, Cardillo G, et al. Clotting Factors in COVID-19: Epidemiological Association and Prognostic Values in Different Clinical Presentations in an Italian Cohort. *J Clin Med*. 2020 May 7;9(5):1371.
56. Li Q, Ding X, Xia G, Chen H-G, Chen F, Geng Z, et al. Eosinopenia and elevated C-reactive protein facilitate triage of COVID-19 patients in fever clinic: a retrospective case-control study. *EClinicalMedicine*. 2020 May;100375.
57. Ferrari D, Motta A, Strollo M, Banfi G, Locatelli M. Routine blood tests as a potential diagnostic tool for COVID-19. *Clin Chem Lab Med CCLM* [Internet]. 2020 Apr 16 [cited 2020 May 25];0(0). Available from: <https://www.degruyter.com/view/journals/cclm/ahead-of-print/article-10.1515-cclm-2020-0398/article-10.1515-cclm-2020-0398.xml>
58. Liu R, Ma Q, Han H, Su H, Liu F, Wu K, et al. The value of urine biochemical parameters in the prediction of the severity of coronavirus disease 2019. *Clin Chem Lab Med CCLM* [Internet]. 2020 Apr 14 [cited 2020 May 25];0(0). Available from: <https://www.degruyter.com/view/journals/cclm/ahead-of-print/article-10.1515-cclm-2020-0220/article-10.1515-cclm-2020-0220.xml>
59. Gao Y, Li T, Han M, Li X, Wu D, Xu Y, et al. Diagnostic Utility of Clinical Laboratory Data Determinations for Patients with the Severe COVID-19. *J Med Virol* [Internet]. 2020 Mar 17 [cited 2020 May 25]; Available from: <https://onlinelibrary.wiley.com/doi/abs/10.1002/jmv.25770>
60. Lingeswaran M, Goyal T, Ghosh R, Suri S, Mitra P, Misra S, et al. Inflammation, Immunity and Immunogenetics in COVID-19: A Narrative Review. *Indian J Clin Biochem* [Internet]. 2020 Jun 6 [cited 2020 Jun 10]; Available from: <http://link.springer.com/10.1007/s12291-020-00897-3>
61. Zhu Z, Cai T, Fan L, Lou K, Hua X, Huang Z, et al. Clinical value of immune-inflammatory parameters to assess the severity of coronavirus disease 2019. *Int J Infect Dis*. 2020 Jun;95:332–9.
62. Han H, Xie L, Liu R, Yang J, Liu F, Wu K, et al. Analysis of heart injury laboratory parameters in 273 COVID-19 patients in one hospital in Wuhan, China. *J Med Virol* [Internet]. 2020 Apr 15 [cited 2020 May 25]; Available from: <http://doi.wiley.com/10.1002/jmv.25809>
63. Wu Y, Huang X, Sun J, Xie T, Lei Y, Muhammad J, et al. Clinical Characteristics and Immune Injury Mechanisms in 71 Patients with COVID-19. Rosenberg HF, editor. *mSphere* [Internet]. 2020 Jul 15 [cited 2020 Jul 21];5(4). Available from: <https://msphere.asm.org/content/5/4/e00362-20>
64. Zhang D, Zhou X, Yan S, Tian R, Su L, Ding X, et al. Correlation between cytokines and coagulation-related parameters in patients with coronavirus disease 2019 admitted to ICU. *Clin Chim Acta*. 2020 Nov;510:47–53.
65. Zhang B, Zhang J, Chen H, Chen L, Chen Q, Li M, et al. Novel coronavirus disease 2019 (COVID-19): relationship between chest CT scores and laboratory parameters. *Eur J Nucl Med Mol Imaging* [Internet]. 2020 May 12 [cited 2020 May 25]; Available from: <http://link.springer.com/10.1007/s00259-020-04854-3>
66. Ibañez C, Perdomo J, Calvo A, Ferrando C, Reverter JC, Tassies D, et al. High D dimers and low global fibrinolysis coexist in COVID19 patients: what is going on in there? *J Thromb Thrombolysis* [Internet]. 2020 Jul 15 [cited 2020 Jul 21]; Available from: <http://link.springer.com/10.1007/s11239-020-02226-0>
67. Zhang Y, Cao W, Jiang W, Xiao M, Li Y, Tang N, et al. Profile of natural anticoagulant, coagulant factor and anti-phospholipid antibody in critically ill COVID-19 patients. *J Thromb Thrombolysis* [Internet]. 2020 Jul 9 [cited 2020 Jul 21]; Available from: <http://link.springer.com/10.1007/s11239-020-02182-9>
68. Yuan X, Huang W, Ye B, Chen C, Huang R, Wu F, et al. Changes of hematological and immunological parameters in COVID-19 patients. *Int J Hematol* [Internet]. 2020 Jul 12 [cited 2020 Jul 21]; Available from: <http://link.springer.com/10.1007/s12185-020-02930-w>



69. Tan C, Huang Y, Shi F, Tan K, Ma Q, Chen Y, et al. C-reactive protein correlates with computed tomographic findings and predicts severe COVID-19 early. *J Med Virol* [Internet]. 2020 Apr 25 [cited 2020 May 25]; Available from: <https://onlinelibrary.wiley.com/doi/abs/10.1002/jmv.25871>
70. Fu J, Kong J, Wang W, Wu M, Yao L, Wang Z, et al. The clinical implication of dynamic neutrophil to lymphocyte ratio and D-dimer in COVID-19: A retrospective study in Suzhou China. *Thromb Res*. 2020 Aug;192:3–8.
71. Tang N, Li D, Wang X, Sun Z. Abnormal coagulation parameters are associated with poor prognosis in patients with novel coronavirus pneumonia. *J Thromb Haemost*. 2020 Apr;18(4):844–7.
72. Chen X, Yan L, Fei Y, Zhang C. Laboratory abnormalities and risk factors associated with in-hospital death in patients with severe COVID-19. *J Clin Lab Anal* [Internet]. 2020 Jul 12 [cited 2020 Jul 21]; Available from: <https://onlinelibrary.wiley.com/doi/abs/10.1002/jcla.23467>
73. Liao D, Zhou F, Luo L, Xu M, Wang H, Xia J, et al. Haematological characteristics and risk factors in the classification and prognosis evaluation of COVID-19: a retrospective cohort study. *Lancet Haematol* [Internet]. 2020 Jul [cited 2020 Jul 21]; Available from: <https://linkinghub.elsevier.com/retrieve/pii/S2352302620302179>
74. Yuan J, Zou R, Zeng L, Kou S, Lan J, Li X, et al. The correlation between viral clearance and biochemical outcomes of 94 COVID-19 infected discharged patients. *Inflamm Res*. 2020 Jun;69(6):599–606.
75. Li Y, Zhao K, Wei H, Chen W, Wang W, Jia L, et al. Dynamic relationship between D-dimer and COVID-19 severity. *Br J Haematol* [Internet]. 2020 May 18 [cited 2020 May 25]; Available from: <https://onlinelibrary.wiley.com/doi/abs/10.1111/bjh.16811>
76. Pilotto A, Odolini S, Stefano Masciocchi S, Comelli A, Volonghi I, Gazzina S, et al. Steroid-responsive encephalitis in Covid-19 disease. *Ann Neurol* [Internet]. 2020 May 17 [cited 2020 May 25]; Available from: <https://onlinelibrary.wiley.com/doi/abs/10.1002/ana.25783>
77. Liu Y, Sun W, Guo Y, Chen L, Zhang L, Zhao S, et al. Association between platelet parameters and mortality in coronavirus disease 2019: Retrospective cohort study. *Platelets*. 2020 May 18;31(4):490–6.
78. Fan Z, Chen L, Li J, Cheng X, Yang J, Tian C, et al. Clinical Features of COVID-19-Related Liver Functional Abnormality. *Clin Gastroenterol Hepatol*. 2020 Jun;18(7):1561–6.
79. Han H, Xie L, Liu R, Yang J, Liu F, Wu K, Chen L, Hou W, Feng Y, Zhu C. Analysis of heart injury laboratory parameters in 273 COVID-19 patients in one hospital in Wuhan, China. *J Med Virol*. 2020 Jul;92(7):819–823. doi: 10.1002/jmv.25809.
80. Cui S, Chen S, Li X, Liu S, Wang F. Prevalence of venous thromboembolism in patients with severe novel coronavirus pneumonia. *J Thromb Haemost* [Internet]. 2020 May 6 [cited 2020 May 25]; Available from: <https://onlinelibrary.wiley.com/doi/abs/10.1111/jth.14830>
81. Chen L, Huang S, Yang J, Cheng X, Shang Z, Lu H, et al. Clinical characteristics in patients with SARS-CoV-2/HBV co-infection. *J Viral Hepat* [Internet]. 2020 Jul 15 [cited 2020 Jul 21]; Available from: <https://onlinelibrary.wiley.com/doi/abs/10.1111/jvh.13362>
82. Roberts MB, Izzy S, Tahir Z, Al Jarrah A, Fishman JA, El Khoury J. COVID-19 in solid organ transplant recipients: dynamics of disease progression and inflammatory markers in ICU and non-ICU admitted patients. *Transpl Infect Dis* [Internet]. 2020 Jul 12 [cited 2020 Jul 21]; Available from: <https://onlinelibrary.wiley.com/doi/abs/10.1111/tid.13407>
83. Duan K, Liu B, Li C, Zhang H, Yu T, Qu J, et al. Effectiveness of convalescent plasma therapy in severe COVID-19 patients. *Proc Natl Acad Sci*. 2020 Apr 28;117(17):9490–6.
84. Wright FL, Vogler TO, Moore EE, Moore HB, Wohlauer MV, Urban S, et al. Fibrinolysis Shutdown Correlates to Thromboembolic Events in Severe COVID-19 Infection. *J Am Coll Surg* [Internet]. 2020 May [cited 2020 May 25]; Available from: <https://linkinghub.elsevier.com/retrieve/pii/S1072751520304002>
85. Lin A, He Z-B, Zhang S, Zhang J-G, Zhang X, Yan W-H. Early risk factors for the duration of SARS-CoV-2 viral positivity in COVID-19 patients. *Clin Infect Dis* [Internet]. 2020 Apr 27 [cited 2020 May 25]; Available from: <https://academic.oup.com/cid/advance-article/doi/10.1093/cid/ciaa490/5825508>
86. Soraya GV, Ulhaq ZS. Crucial laboratory parameters in COVID-19 diagnosis and prognosis: An updated meta-analysis. *Med Clin (Barc)*. 2020 Aug 28;155(4):143–151. doi: 10.1016/j.medcli.2020.05.017. Epub 2020 Jun 5. PMID: 32586670; PMCID: PMC7274591.
87. Henry BM, de Oliveira MHS, Benoit S, Plebani M, Lippi G. Hematologic, biochemical and immune biomarker abnormalities associated with severe illness and mortality in coronavirus disease 2019 (COVID-19): a meta-analysis. *Clin Chem Lab Med*. 2020 Jun 25;58(7):1021–1028. doi: 10.1515/cclm-2020-0369. PMID: 32286245.
88. Henry BM, Benoit SW, de Oliveira MHS, Hsieh WC, Benoit J, Ballout RA, Plebani M, Lippi G. Laboratory abnormalities in children with mild and severe coronavirus disease 2019 (COVID-19): A pooled analysis and review. *Clin Biochem*. 2020 Jul;81:1–8. doi: 10.1016/j.clinbiochem.2020.05.012. Epub 2020 May 27. PMID: 32473151; PMCID: PMC7251358.
89. Elshazli RM, Toraih EA, Elgaml A, El-Mowafy M, El-Mesery M, Amin MN, Hussein MH, Killackey MT, Fawzy

MS, Kandil E. Diagnostic and prognostic value of hematological and immunological markers in COVID-19 infection: A meta-analysis of 6320 patients. PLoS One. 2020 Aug 21;15(8):e0238160. doi: 10.1371/journal.pone.0238160. PMID: 32822430; PMCID: PMC7446892.

90. Henry B, Cheruiyot I, Vikse J, Mutua V, Kipkorir V, Benoit J, Plebani M, Bragazzi N, Lippi G. Lymphopenia and neutrophilia at admission predicts severity and mortality in patients with COVID-19: a meta-analysis. Acta Biomed. 2020 Sep 7;91(3):e2020008. doi: 10.23750/abm.v91i3.10217. PMID: 32921706.

91. Moutchia J, Pokharel P, Kerri A, McGaw K, Uchai S, Nji M, Goodman M. Clinical laboratory parameters associated with severe or critical novel coronavirus disease 2019 (COVID-19): A systematic review and meta-analysis. PLoS One. 2020 Oct 1;15(10):e0239802. doi: 10.1371/journal.pone.0239802. PMID: 33002041; PMCID: PMC7529271.

92. Mitra P, Misra S, Sharma P. COVID-19 Pandemic in India: What Lies Ahead. Indian J Clin Biochem [Internet]. 2020 Apr 20 [cited 2020 Jun 10]; Available from: <http://link.springer.com/10.1007/s12291-020-00886-6>

# Broadening the list of differential diagnosis for acute abdomen – a case report from Nepal

Vivek Pant<sup>1</sup>, Keyoor Gautam<sup>2</sup>, Devish Pyakurel<sup>2</sup>, Aabha Shrestha<sup>2</sup>, Santosh Pradhan<sup>1</sup>, Neeraj Joshi<sup>3</sup>

<sup>1</sup> Biochemistry, Department of Clinical Biochemistry, Samyak Diagnostic, Jawalakhel, Lalitpur, Nepal

<sup>2</sup> Pathology, Department of Pathology, Samyak Diagnostic, Jawalakhel, Lalitpur, Nepal

<sup>3</sup> Department of Internal Medicine, Nidan Hospital, Pulchowk, Lalitpur, Nepal

---

## ARTICLE INFO

### **Corresponding author:**

Vivek Pant, MD  
Department of Clinical Biochemistry  
Samyak Diagnostic, Jawalakhel  
P.O.Box: 11708, Lalitpur  
Nepal  
E-mail: [drv pant@gmail.com](mailto:drv pant@gmail.com)

### **Key words:**

ayurvedic medicine, lead toxicity,  
acute abdomen, blood lead level

---

## ABSTRACT

When a patient has an acute abdominal pain, it is important to identify if the underlying cause is life threatening. To that end, a thorough medical history and relevant investigation will be pivotal. Here we report a case of lead toxicity where the patient presented with an acute abdomen following intake of Ayurvedic medicines. The baseline blood lead level was 82.3 µg/dl. The Ayurvedic medicines when analyzed for its lead content, revealed high lead concentration. We observed that the cessation of Ayurvedic medication along with D-penicillamine therapy was beneficial in reducing the blood lead level and in alleviating abdominal pain. Our findings implicate the need of awareness program regarding the potential health hazards associated with the use Ayurvedic medicines.

## INTRODUCTION

Lead poisoning has been recognized as a major public health problem, particularly in developing nations like Nepal. (1) Air, dust, soil, paints, cosmetics, dietary and herbal supplements, and soiled parental work clothing are potential sources of exposure to lead. Lead can have a wide range of biological effects depending on the level and duration of exposure, including effects on heme synthesis, the central nervous system, kidneys, alimentary tract, and other organs. (2) The effect is mediated through increased oxidative stress, ionic mechanisms, and apoptosis. (3)

Lead colic is a rare cause of abdominal pain. (4) The diagnosis is most often reached in a context of professional exposure or in populations at risk of contact with lead. Due to previously reported cases of lead colic in Ayurvedic medicine user, this cause is important to be considered in abdominal pain of unknown origin. (5, 6)

Products used in Ayurvedic medicine contain herbs, metals, minerals, or other materials that may be harmful if used improperly or without the direction of a trained practitioner. Doses of metals in Ayurvedic medicine in practice are based on recommendations given in ancient Ayurvedic texts. Nearly half of the medicines used in the Ayurvedic formulary intentionally contain at least one metal to enhance potency of the drug. (7) The addition of lead is believed to have fungicidal properties and improve shelf-life of the medicine. These medicines can have drug interaction with the allopathic medicine.

Nepalese people have a growing fascination with natural remedies and traditional medicines. (8) Practitioners of Ayurveda in Nepal undergo recognized institutional education and training and are licensed by the government body. Though traditional medicines have been used in Nepal, there is little quality control or trials. Some unscrupulous drug manufacturers

mix allopathic medicines in Ayurvedic drugs, usually steroids and since the patient feels temporary relief; he ascribes it to the Ayurvedic medicine. (9) Uncontrolled use of herbs, use of heavy metals, lack of quality control and adding steroids damages the quality of Ayurvedic medicine. (9) In Nepal, patient with chronic diseases like arthritis, asthma, hemorrhoids, insomnia, autoimmune diseases and skin diseases have more tendency to use ayurvedic medicines. Acute clinical presentation in patients using Ayurvedic medicine adulterated with heavy metal like gold and alkaloids has been reported from Nepal previously. (10) To the best of our knowledge, acute clinical manifestation due to lead toxicity after consumption of Ayurvedic medicine has not been reported from Nepal earlier.

## CLINICAL DIAGNOSTIC CASE

A 38-year-old man presented to a gastroenterologist with a one month history of progressive epigastric pain without radiation. The pain had increased in intensity in the last two days. He reported dark and hard stool, decreased appetite, tiredness, and nausea. He did not have any significant medical or surgical history. However, he had been taking Ayurvedic medication for three months to increase his sperm count, which was prescribed to him by a licensed Ayurvedic practitioner in Kathmandu. He is in the army by profession. He consumed alcohol occasionally and was a non-smoker.

His vital signs were stable. On physical examination, no signs of peritonitis were observed. Physical examination was remarkable for abdominal tenderness in the epigastric area. Testicular examination and per rectal examination were normal.

Laboratory evaluation revealed a hemoglobin level of 9.7 g/dL (Reference range - 13.5 – 16.9 gm %) with a mean corpuscular volume (MCV)

of 87.1 fL (Reference range - 81.8 – 95.5 fL) and a reticulocyte count of 3.8% (Reference range - 0.5 – 2%). Hemoglobin and MCV were measured using the Sysmex automated hematology analyzer XN 330 (Sysmex, Milton Keynes, UK). Reticulocyte count was measured by microscopy. The liver, pancreas and kidney function tests were normal except for a mild increase in transaminase level. Imaging included a CT scan and an abdominal ultrasound, neither of which revealed any abnormalities. In addition, an upper gastrointestinal endoscopy and colonoscopy revealed no abnormalities either.

The serum iron chemistry, antinuclear antibody screening, vitamin B12, folate and thyroid-stimulating hormone were in normal range. His peripheral blood smear showed anisocytosis with normochromia. There was no evidence of hemolysis. The hemoglobin electrophoresis was normal too. Subsequently, blood lead level (BLL)

was measured and the result showed an elevated level of lead at 82.3 µg/dL (normal <10 µg/dL). Measurement of BLL was performed using the Lead Care II instrument (Magellan Diagnostics Inc., N. Billerica, Massachusetts, USA) based on the principle of anodic stripping voltametry. The zinc protoporphyrin level was 310 mg/dL (normal <40 mg/dL) and was measured using hematofluorometer (Helena Laboratories, Beaumont, Texas, USA).

The patient's history showed no other potential sources of lead exposure than the intake of Ayurvedic medicine. He lived in a modern house. None of his family members had similar symptoms. He is an army by profession and he reported that he performs most of his work using gloves and protective clothing in order to minimize exposure if any.

Extracts from the seven Ayurvedic medicines that the patient was using, were evaluated

**Table 1** Lead level in the Ayurvedic drugs the index patient was using

Name of Ayurvedic drug	Lead concentration (In parts per million)
Prawal Pishti	11.18
Siddha Makara Dhvaja Gutti	102.53
Chankrashekhar ras	12.89
Shatawari Granules	4.53
Musli pak (Laghu)	11.54
Vanari	13.74
Vanga Bhasma	209.70

**Note:** Prescribed limit of lead in Ayurvedic medicine is less than 10 ppm (22).

using an atomic absorption spectrophotometer. The test showed a high concentration of lead in six out of the seven medicines. (Table 1)

Patient was managed on outpatient basis. He was prescribed with D-Penicillamine 250 mg one hour before meal, initially once a day for one month then twice a day for another two months and thrice a day for the fourth month. Patient's BLL alleviated over time (Figure 1). His symptoms were resolved after 2 weeks of treatment.

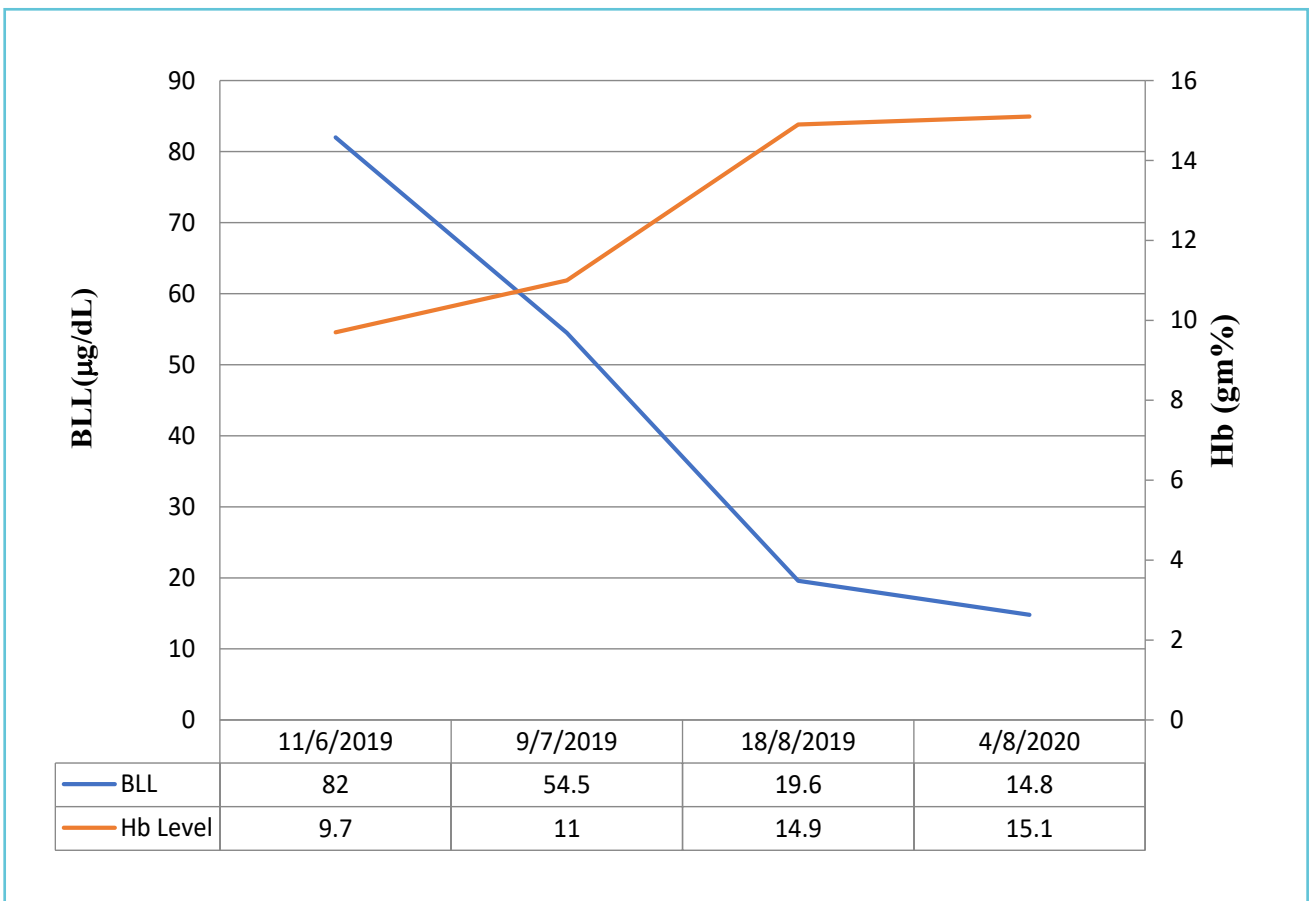
### DISCUSSION

Acute lead toxicity that results from short-term, high dose lead absorption causes normocytic or microcytic anemia, abdominal pain and constipation, arthralgias and myalgias, and central

nervous system impairment including headache, mood disorder and encephalopathy. (11) Our patient manifested many of the known signs and symptoms of acute lead toxicity, including abdominal pain, constipation, anemia and abnormal liver enzymes.

The exact pathogenesis of lead induced abdominal colic is unknown. However, proposed mechanisms include alterations in the visceral smooth muscle tone due to action of lead on visceral autonomic nervous system, changes in the sodium transport in small intestinal mucosa, porphyrinopathy and lead induced interstitial pancreatitis. (12, 13) Abnormal liver enzyme is possible due to the depletion of the antioxidants savings of the cells in acute lead toxicity. (14) Lead interferes with a variety of heme

**Figure 1** Patient blood lead level (BLL) versus hemoglobin (Hb) trended over time



biosynthetic enzymes, including delta-aminolevulinic acid that conjugates levulinic acid to form porphobilinogen and ferrochelatase which incorporates ferrous iron into protoporphyrin IX ring. This results in anemia, hypersideraemia, reticulocytosis and basophilic stippling, due to the persistence of cytoplasmic proteins. (15, 16) Basophilic stippling of erythrocytes is typical but not specific for lead poisoning. (15, 17) In our patient, the basophilic stippling was not seen.

Several cases of lead intoxication associated with Ayurvedic medicines have been reported worldwide. (18 -21) Patients taking these medicines are often overlooked and are usually not evaluated for lead exposure until serious manifestations have occurred. Clinical practitioner in geographical area with Ayurvedic medication users should have a high index of suspicion of lead toxicity among persons with characteristic signs and symptoms in the absence of occupational exposure.

The World Health Organization has prescribed a limit for lead contents in herbal medicine at 10 ppm. (22) Six Ayurvedic medicine out of seven, that patient was using had higher lead concentration. (Table 1) The Vanga Bhasma named Ayurvedic medicine which the index case was using contained the highest amount of lead (209.70 ppm) out of these six Ayurvedic medicines, when analyzed through atomic absorption spectrophotometer. The lead concentration in all seven Ayurvedic medicines is shown in Table 1. Heavy metals are commonly incorporated into Ayurvedic preparations as ashes or bhasmas. Experts in this field claim that role of bhasmas is to enhance the herbal products potency via facilitating the entry into the relevant cells and if adequately prepared are safe for administration. Use of bhasmas in Ayurvedic medicine leading to lead toxicity has been reported previously. (23, 24)

In adults, the decision to use chelation therapy is ultimately clinical but may be guided by the BLL. The two chelating agents most commonly used to treat adults are oral succimer [meso-2, 3-dimercaptosuccinic acid (DMSA)] and edetate calcium disodium (CaEDTA). (25) D-penicillamine was used in our patient since this is the only available treatment option in Nepal.

The pace of improvement may be highly variable, ranging from weeks to years, depending on the magnitude of intoxication. (26) It has been found that chelation therapy reduces blood lead concentrations acutely, but the levels rebound within weeks to months after treatment due to redistribution from bone, requiring repeated courses of treatment. Our patient recovered quickly and the BLL decreased linearly. The acute high dose intake of Ayurvedic medicine in our patient might be the cause for diminished lead distribution to the bone and linear decrease in BLL. However, our patient is advised for an annual blood lead and zinc protoporphyrin level examination and, avoidance of exposure to lead by preventing use of improperly prepared contaminated Ayurvedic drugs.

## LEARNING POINTS

- The adulterated Ayurvedic medicine due to its easy availability and lack of focused scientific research has potential to cause more cases of lead toxicity.
- Clinicians should consider lead toxicity secondary to Ayurvedic medicine intake in their differential diagnoses of anemia, with abdominal pain.
- Health risks posed by the Ayurvedic medicine should be discussed among health-care providers and awareness should be increased among general public.



### Consent

Written informed consent was obtained from the patient for publication of this case report.

### Acknowledgement

We appreciate the kindness and technical help from Mr. Ram Charitra Shah and Mr. Sunil Babu Khatri.

### Author contributions

VP conceptualized and designed the study. This manuscript is written by VP. Data collection and laboratory analysis was performed by SP and AS. NJ was the physician involved in patient management. KG, DP and NJ revised and approved the final version of this manuscript.



### REFERENCES

1. Gautam K, Pant V, Pradhan S, Pyakurel D, Bhandari B, Shrestha A. Blood lead levels in rag-pickers of Kathmandu and its association with hematological and biochemical parameters. *EJIFCC*. 2020 Jun; 31(2):125.
2. Papanikolaou NC, Hatzidaki EG, Belivanis S, Tzanakakis GN, Tsatsakis AM. Lead toxicity update. A brief review. *Medical science monitor*. 2005 Oct 1; 11(10):RA329-36.
3. Sakai T. Biomarkers of lead exposure. *Industrial health*. 2000; 38(2):127-42.
4. Van Vonderen MG, Klinkenberg-Knol EC, Craanen ME, Touw DJ, Meuwissen SG, De Smet PA. Severe gastrointestinal symptoms due to lead poisoning from Indian traditional medicine. *The American journal of gastroenterology*. 2000 Jun 1; 95(6):1591.
5. Raviraja A, Babu GV, Sehgal A, Saper RB, Jayawardene I, Amarasiriwardena CJ, Venkatesh T. Three cases of lead toxicity associated with consumption of ayurvedic medicines. *Indian Journal of Clinical Biochemistry*. 2010 Jul 1; 25(3):326-9.
6. Sood A, Midha V, Sood N. Pain in abdomen--do not forget lead poisoning. *Indian journal of gastroenterology: official journal of the Indian Society of Gastroenterology*. 2002; 21(6):225-6.
7. Gogtay NJ, Bhatt HA, Dalvi SS, Kshirsagar NA. The use and safety of non-allopathic Indian medicines. *Drug safety*. 2002 Dec 1; 25(14):1005-19.
8. Kafle G, Bhattarai I, Shrestha AK, Siwakoti M. Why do patients choose to consume Ayurvedic Medicines in Nepal? An exploratory study. *International Journal of Ayurvedic Medicine*. 2018 Oct 1; 9(4):250-7.
9. Ernst E. Toxic heavy metals and undeclared drugs in Asian herbal medicines. *Trends in pharmacological sciences*. 2002 Mar 1; 23(3):136-9.
10. Paudyal B, Thapa A, Sigdel KR, Adhikari S, Basnyat B. Adverse events with ayurvedic medicines-possible adulteration and some inherent toxicities. *Wellcome Open Research*. 2019; 4.
11. Centers for Disease Control and Prevention (CDC). Adult blood lead epidemiology and surveillance--United States, 2008-2009. *MMWR. Morbidity and mortality weekly report*. 2011 Jul 1; 60(25):841.
12. Karmakar N, Anand S. Study of the inhibitory effect of lead acetate on duodenal contractility in rat. *Clinical and experimental pharmacology and physiology*. 1989 Sep; 16(9):745-50.
13. Yoshinari O, Makoto K. An association between increased porphyrin precursors and onset of abdominal symptoms in lead poisoning. *Toxicology letters*. 1984 May 1; 21(2):219-23.
14. Matović V, Buha A, Đukić-Čosić D, Bulat Z. Insight into the oxidative stress induced by lead and/or cadmium in blood, liver and kidneys. *Food and Chemical Toxicology*. 2015 Apr 1; 78:130-40.
15. Albahary C. Lead and hemopoiesis: the mechanism and consequences of the erythropathy of occupational lead poisoning. *The American journal of medicine*. 1972 Mar 1; 52(3):367-78.
16. Menezes G, D'souza HS, Venkatesh T. Chronic lead poisoning in an adult battery worker. *Occupational Medicine*. 2003 Oct 1; 53(7):476-8.
17. JENSEN WN, MORENO GD, BESSIS MC. An electron microscopic description of basophilic stippling in red cells. *Blood*. 1965 Jun; 25(6):933-43.
18. Ernst E. Heavy metals in traditional Indian remedies. *European journal of clinical pharmacology*. 2002 Feb 1; 57(12):891-6.
19. Centers for Disease Control and Prevention (CDC). Lead poisoning associated with ayurvedic medications--five states, 2000-2003. *MMWR Morb Mortal Wkly Rep*. 2004; 53:582-584.
20. Muzi G, Dell'Omo M, Murgia N, Curina AN, Ciabatta SI, Abbritti G. Lead poisoning caused by Indian



ethnic remedies in Italy. *La Medicina del lavoro*. 2005; 96(2):126.

21. Gunturu KS, Nagarajan P, McPhedran P, et al. Ayurvedic herbal medicine and lead poisoning. *J Hematol Oncol*. 2011; 4:51.

22. World Health Organization. Quality control methods for medicinal plant materials. World Health Organization; 1998.

23. Raviraja A, Babu GV, Sehgal A, Saper RB, Jayawardene I, Amarasiriwardena CJ, Venkatesh T. Three cases of lead toxicity associated with consumption of ayurvedic medicines. *Indian Journal of Clinical Biochemistry*. 2010 Jul 1; 25(3):326-9.

24. Mehta V, Midha V, Mahajan R, Narang V, Wander P, Sood R, Sood A. Lead intoxication due to ayurvedic medications as a cause of abdominal pain in adults. *Clinical Toxicology*. 2017 Feb 7; 55(2):97-101.

25. Porru S, Alessio L. The use of chelating agents in occupational lead poisoning. *Occupational Medicine*. 1996 Feb 1; 46(1):41-8.

26. Kosnett MJ, Wedeen RP, Rothenberg SJ, Hipkins KL, Materna BL, Schwartz BS, Hu H, Woolf A. Recommendations for medical management of adult lead exposure. *Environmental health perspectives*. 2007 Mar; 115(3): 463-71.

# Galactorrhoea and hyperprolactinaemia in a non-pregnant female with burns

Marli van Heerden, Dineo Mabuza

*Department of Chemical Pathology, University of the Witwatersrand and National Health Laboratory Service,  
Johannesburg, South Africa*

---

## ARTICLE INFO

### **Corresponding author:**

Dr. Dineo Mabuza,  
Department of Chemical Pathology  
University of Witwatersrand  
Wits Medical School  
7 York Road, Parktown, 2193  
Johannesburg  
South Africa  
Phone: +27(11) 489-8762  
Fax: +27(11) 489-8451  
E-mail: [dineo.mabuza@nhls.ac.za](mailto:dineo.mabuza@nhls.ac.za)

### **Key words:**

chest burns, galactorrhoea,  
prolactin, hyperprolactinaemia

---

## ABSTRACT

Galactorrhoea is the presence of breast milk secretion in non-pregnant or non-breastfeeding females. In females with galactorrhoea and amenorrhoea, hyperprolactinaemia is the most likely cause. Chest wall pathology is a known cause of hyperprolactinaemia, however there are few documented reports on galactorrhoea following burn injury to the chest wall. We report a case of an adult female patient who sustained hot water burns to her torso. Following the second skin graft failure, galactorrhoea was noted and investigated. Hyperprolactinaemia was found with normal renal and thyroid function. There were no abnormalities detected on the hormones in the hypothalamic pituitary axis and MRI. Hyperprolactinaemia was attributed to chest wall injuries causing pathological stimulation of the neural suckling reflex. The patient was treated with dopamine agonists for one month and the galactorrhoea disappeared. Treatment was discontinued on discharge and the prolactin levels remained normal upon follow-up visit to the clinic.

## INTRODUCTION

Galactorrhoea indicates the presence of milk secretion from the breast in the absence of pregnancy or more than 6 months postpartum in a woman not breastfeeding (1). Milk production is under the influence of the hormone prolactin, secreted by the anterior hypothalamus; its secretion is mainly controlled by the inhibitory effect of dopamine (1,2). Factors that inhibit dopamine secretion and clearance of prolactin have an effect on prolactin levels. These include diet, stress, exercise, renal & liver disease, hypothyroidism, high oestrogen states, medication and chest wall pathology (2). High prolactin levels result in decreased luteinizing hormone (LH) and follicle stimulating hormone (FSH) by inhibition of release of gonadotropin releasing hormone and may result in amenorrhoea (3).

Although chest wall pathology is a known cause of galactorrhoea with hyperprolactinaemia, there are few reported cases of patients with chest wall burns. We report a case of galactorrhoea with hyperprolactinaemia in an adult female with chest wall burns.

## CLINICAL-DIAGNOSTIC CASE

A 28-year old female, presented to the emergency unit, in May 2018 with hot water burns to her chest following a domestic dispute. She was 31 weeks pregnant and subsequently progressed to premature labour for which a caesarean section was performed. The baby demised one month later. There was no significant past medical or surgical history.

A skin graft to the chest area was performed (Figure 1) and she was discharged to follow up at the outpatient clinic for wound care. She was readmitted in September for graft failure due to sepsis and her treatment included antibiotics and wound dressings. The second skin graft was performed in October and subsequently

became septic. She presented with pus on both breasts and Methicillin Resistant Staphylococcus was cultured and found to be sensitive to Vancomycin. She was treated with Vancomycin and discharged home for wound care at the outpatient clinic. Despite the antibiotic therapy the graft failed.

In February 2019, she was readmitted for wound sepsis. Galactorrhoea was noted and endocrinologists were consulted. She reported occasional headaches and absent menses but no visual disturbances. Her last menstrual cycle was prior to the pregnancy and she was not on any contraceptives. She denied illicit drug use and was not on any dopamine antagonists. On examination there was no evidence of hirsutism, acne and hyperpigmentation.

Pregnancy was excluded by means of a urine human chorionic gonadotropin (hCG) point of care test in the ward. Serum prolactin was measured and found to be markedly elevated (353.7 µg/L, reference interval (RI), 4.8-23.3 µg/L). Macroprolactin was excluded in the laboratory by polyethylene glycol (PEG) precipitation. This procedure is performed with PEG solution (25% w/v), prepared by dissolving PEG 6000 in phosphate buffered saline (PBS) stock solution and PBS buffer. An equal volume of the patient sample and PEG solution was mixed and allowed to incubate at room temperature for ten minutes. The PEG was precipitated by centrifugation, after which the supernatant was analysed. The post-PEG monomeric prolactin was 249.5 µg/L (RI, 3.5-18 µg/L) which confirmed true hyperprolactinaemia. The renal function and other hormones in the hypothalamic pituitary axis were assessed (Table 1).

Levels of creatinine, estimated glomerular filtration rate (eGFR), thyroid stimulating hormone (TSH), free T4 (fT4), human growth hormone (hGH), insulin-like growth factor 1 (IGF-1) and

**Figure 1** The patient with severe burn wounds post skin graft



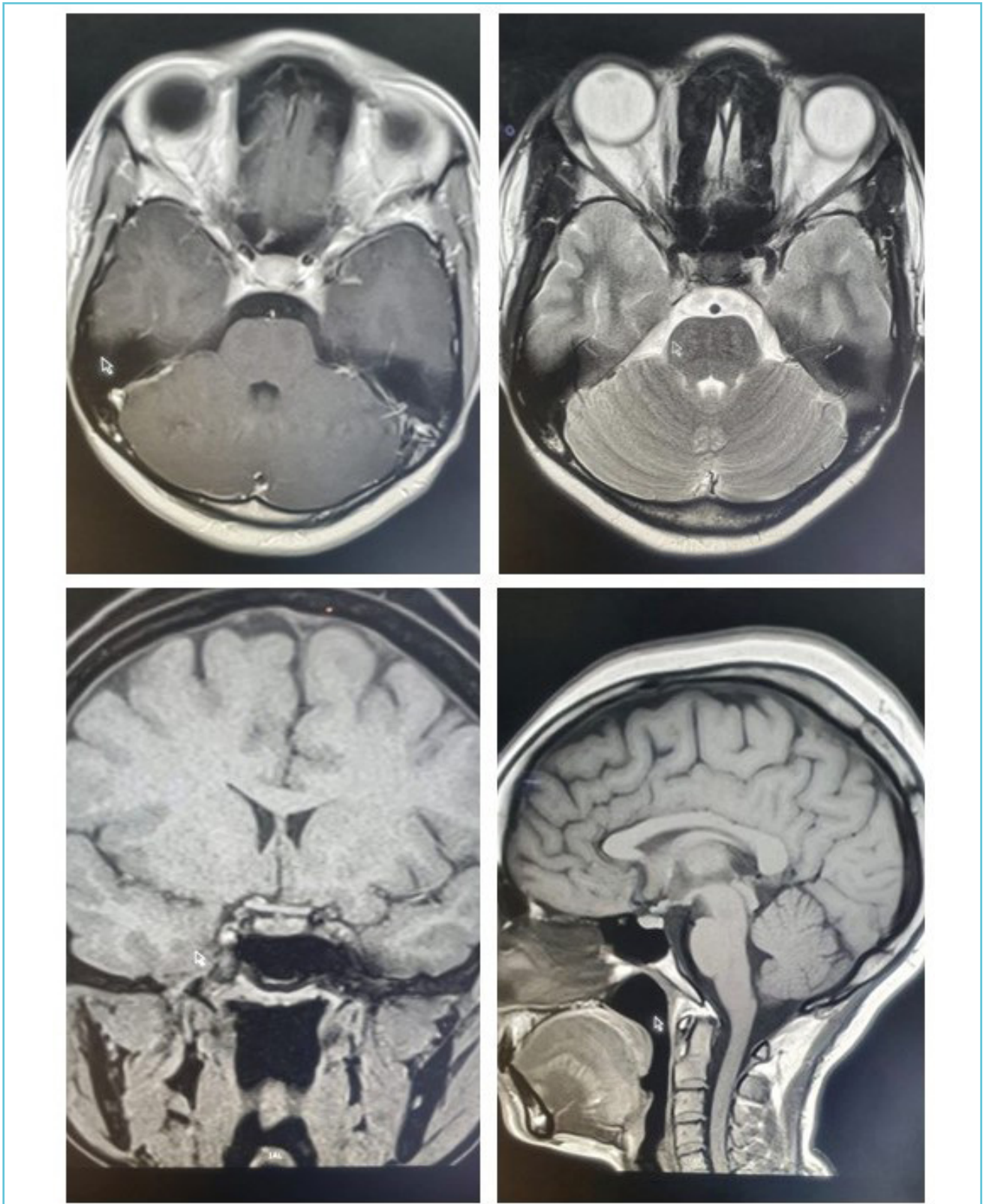
**Table 1** Selected biochemical results

Test	Reference interval	Test results (19/02/2019)
Creatinine	49-90 µmol/L	59
eGFR (MDRD)	> 60 ml/min/1.73m <sup>2</sup>	> 60
TSH	0.27-4.20 mIU/L	2.1
ft4	12.0-22.0 pmol/L	12.6
FSH	Follicular phase: 3.5 -12.5 IU/L Ovulation phase: 4.7 - 21.5 IU/L Luteal phase: 1.7 - 7.7 IU/L Postmenopausal: 25.8 - 134.8 IU/L	6.3
LH	Follicular phase: 2.4 - 12.6 IU/L Ovulation phase: 14.0 - 95.6 IU/L Luteal phase: 1.0 - 11.4 IU/L Postmenopausal: 7.7 - 58.5 IU/L	16.4
Estradiol	Follicular phase: 45 – 854 pmol/L Ovulation phase: 151 – 1461 pmol/L Luteal phase: 82 – 1251 pmol/L Postmenopausal: (<18 – 505 pmol/L	107
hGH	0.13 - 9.88 µg/L	0.5
IGF-I	88-537 µg/L	140
ACTH	1.6-13.9 pmol/L	2.6
Random Cortisol	Morning (06:00-10:00): 133 – 537 nmol/L Afternoon (16:00-20:00): 68 – 327 nmol/L	205

adrenocorticotrophic hormone (ACTH) were all within the reference intervals. A Magnetic resonance imaging (MRI) scan (Figure 2) revealed a normal pituitary gland and stalk with no evidence of infiltratory disease or a tumour.

The patient was treated with a dopamine agonist for one month and the galactorrhoea disappeared. Her prolactin levels were within the reference interval on discharge and at 1 week follow up (Table 2).

Figure 2 The patient's MRI of the brain that shows no pituitary pathology



**Table 2** Prolactin results

Reference interval	15/02/2019	19/02/2019*	21/02/2019	04/03/2019#	25/03/2019*
4.8 – 23.3 µg/L	353.7	20.3	21.6	1.2	0.9

\*post initiation of bromocriptine; #2 weeks on therapy; \*1 week post discharge.

## DISCUSSION

There are three forms of prolactin in the circulation: monomeric (23kDa), dimeric and the biologically inactive polymeric prolactin (>100kDa), also known as macroprolactin (3,4). Most immunoassays detect biologically active and inactive forms and results may not reflect the true biological activity (4). Thus, macroprolactin must be excluded to avoid unnecessary investigations or interventions (4,5). In our patient the monomeric prolactin remained elevated post PEG precipitation. Due to the fact that a small amount of monomeric prolactin can be precipitated, post-PEG reference ranges were used (6). Other methods that can remove macroprolactin include gel filtration chromatography, ultrafiltration and antibody binding techniques (4). PEG precipitation is routinely used in most laboratories as it is the least expensive and shows the best correlation with gel-filtration chromatography which is regarded as the gold standard (4,6). Dynamic function testing is not recommended (5).

Once it is confirmed that there is true hyperprolactinaemia, pregnancy should be excluded first (7). The high oestrogen concentration found in pregnancy stimulates prolactin synthesis and results in hyperprolactinaemia (1,2). In addition, the patient was also not on any oestrogen containing contraceptives. The use of dopamine antagonists such as antipsychotics, antihypertensives, antidepressants and metoclopramide was excluded. Illicit drugs and opioids, which are known for inhibition of dopamine release,

were also excluded (2,5,7). Decreased renal clearance of prolactin in chronic kidney disease causes hyperprolactinaemia (1,2,5). Primary hypothyroidism can result in diffuse pituitary enlargement, stimulation of prolactin release by thyrotropin releasing hormone and decreased prolactin clearance from the systemic circulation (2). However, our patient had normal renal and thyroid function.

Once all the above had been excluded, hypothalamic-pituitary disease may be considered (7). A prolactin secreting tumour is the most common pathologic cause of hyperprolactinaemia (1). Prolactin levels >250 µg/L, as seen in this patient, are suggestive of a prolactinoma (5). However, there were no hypothalamic-pituitary lesions noted on MRI.

With the exclusion of all the common causes, the most obvious cause of the galactorrhoea and hyperprolactinemia was the chest burns. The mechanism of hyperprolactinaemia in chest wall injuries is the pathological stimulation of the neural suckling reflex (1,8). The burn wounds are sensitive to touch and the stimuli pass along the intercostal nerves to the posterior column of the spinal cord and finally to the hypothalamus where dopamine release is inhibited (1,9). The patient did not breastfeed post-delivery and she received Bromocriptine to stop lactation. According to the patient, the galactorrhoea started in October but was not reported to the clinicians. The presence of galactorrhoea may explain the septic wounds and graft failure. A moist environment, due to

galactorrhoea, may inhibit incorporation of the graft into the wound bed and provide a favourable environment for bacterial growth. In addition, the presence of galactorrhoea may have caused wet dressings, which resulted in their early and inappropriate removal by the patient; thus, providing an opportunity for exposure of the wound to bacteria.

A study by Goyal et al in India found the incidence of amenorrhoea and/or galactorrhoea in female patients with chest burns to be 15.15%. According to the authors one of the reasons why this condition might be under reported is due to patients not volunteering the information unless prompted (9). Karimi and colleagues (10) reported a case of galactorrhoea that started three months after discharge and responded well to therapy. Their patient had developed hypertrophic scars and keloids despite preventative treatment and skin grafting. The authors claim that this time period correlates with collagen deposition and scar contraction and that the hyperprolactinemia observed during this anabolic period may be related to the development of refractory hypertrophic scars (10).

Galactorrhoea associated with amenorrhoea and hyperprolactinaemia requires treatment (7). The aim of therapy is to restore normal gonadal function and fertility as well as prevent osteoporosis (2). Prolactin levels were successfully reduced on medical therapy in our patient and she was discharged without any medication. It is imperative that she is monitored for recurrence of galactorrhoea and possibly measurement of prolactin at follow up visits.

This case demonstrates the importance of recognizing chest wall pathology as a cause of hyperprolactinemia. Clinicians should specifically ask all women with chest wall burns about galactorrhoea. The presence of galactorrhoea and amenorrhea following chest wall burns should

prompt clinicians to investigate for hyperprolactinemia which can be effectively treated with dopamine agonists.

### TAKE HOME MESSAGES/ LEARNING POINTS

1. In females with galactorrhoea and amenorrhoea, hyperprolactinaemia is the most likely cause.
2. Most immunoassays detect the various forms of prolactin, therefore, macroprolactin must be excluded before further investigations.
3. Chest wall pathology is not a common cause of hyperprolactinaemia, however it should be considered in burn patients presenting with galactorrhoea.
4. Chest wall pathology results in pathological stimulation of the neural suckling reflex which inhibits the release of dopamine by the hypothalamus.
5. Medical therapy, with dopamine agonists, is an effective management strategy for galactorrhoea with hyperprolactinaemia



### *Author disclosures & contributions*

1. Written informed consent was obtained from the patient for publication of this case report and any accompanying images.
2. Ethics was obtained from the Medical Human Research Ethics Committee. Protocol number M190296.
3. The authors declare that they have no competing interests.
4. M van Heerden and D Mabuza interviewed the patient, analysed and interpreted the patient data, and were the major contributors in writing the manuscript. All authors read and approved the final manuscript.



### **Acknowledgements**

We would like to thank Dr. Sarah Melissa Katzew for the clinical information and images of the burns and Dr. Nadege Ntumba for the MRI images.



### **REFERENCES**

1. Leung AKC, Pacaud D. Diagnosis and management of galactorrhea. *Am Fam Physician*. 2004;70(3):543-50.
2. Serri O, Chik CL, Ur E, Ezzat S. Diagnosis and management of hyperprolactinemia. *CMAJ*. 2003;169(6):575-81.
3. Bernard V, Young J, Chanson P, Binart N. New insights in prolactin: pathological implications. *Nat Rev Endocrinol*. 2015; 11:265-75.
4. Kavanagh L, McKenna TJ, Fahie-Wilson MN, Gibney J, Smith TP. Specificity and Clinical Utility of Methods for the Detection of Macroprolactin. *Clin Chem*. 2006;52(7): 1366-72.
5. Melmed S, Casanueva FF, Hoffman AR, Kleinberg DL, Montori VM, Schlechte JA, et al. Diagnosis and Treatment of Hyperprolactinemia: An Endocrine Society Clinical Practice Guideline. *J Clin Endocrinol Metab*. 2011;96(2): 273-88.
6. Beltran L, Fahie-Wilson MN, McKenna TJ, Kavanagh L, Smith TP. Serum total prolactin and monomeric prolactin reference intervals determined by precipitation with polyethylene glycol: Evaluation and validation on common immunoassay platforms. *Clin Chem*. 2008;54(10):1673-81.
7. Huang W, Molitch ME. Evaluation and management of galactorrhea. *Am Fam Physician*. 2012;85(11):1073-80.
8. Jones RE, Gulati M. Hypothalamic and Pituitary Function. In: Bishop ML, Fody EP, Schoeff LE, editors. *Clinical Chemistry: Principles, Techniques, and Correlations*. 7th ed. Philadelphia, PA: Lippincott Williams & Wilkins; 2013. p. 438-52.
9. Goyal N, Gore MA, Shankar R. Galactorrhea and amenorrhea in burn patients. *Burns*. 2008;34(6):825-8.
10. Karimi H, Nourizad S, Momeni M, Rahbar H, Momeni M, Farhadi K. Burns, hypertrophic scar and galactorrhea. *J Inj Violence Res*. 2013;5(2):117-9.

# Improved COVID-19 testing by extraction-free SARS-CoV-2 RT-PCR

Khelil Mohamed Mokhtar

*Pasteur Institute of Algeria, Department of Immunology, Annex of M'sila, Algeria*

---

## ARTICLE INFO

---

**Corresponding author:**

Khelil Mohamed Mokhtar  
Pasteur Institute of Algeria  
Department of Immunology  
Annex of M'sila  
Algeria  
Phone: +213781032119  
E-mail: [kh.immuno@gmail.com](mailto:kh.immuno@gmail.com)

**Key words:**

SARS-CoV-2, COVID-19, RT-PCR,  
heat treatment, nasopharyngeal swab samples

---

## LETTER TO THE EDITOR

---

The RNA extraction is an important checkpoint for the detection of SARS-CoV-2 in swab samples, but it is a major barrier to available and rapid COVID-19 testing. In this study, we validated the extraction-free RT-qPCR method by heat-treatment as an accurate option to nucleic acid purification in Algerian population.

Dear editor,

The new emergence of the novel human coronavirus, in December 2019, in Wuhan City (China), rapidly evolved into a global pandemic. The virus was confirmed to have spread to Algeria in February 2020, which put notable pressure on public and private health laboratories as they attempt to keep up with demands for SARS-CoV-2 testing despite shortage of reagents (1). Currently, the widely used protocol for SARS-CoV-2 detection is RT-qPCR assay preceded by purification of viral RNA from patient sample, typically from nasopharyngeal (NP) swab as described by CDC and WHO (2-4). However, nucleic acid purification step is not only laborious and time-consuming, but the additional steps requiring manual handling can result in experimental errors, especially false positive results due to specimen-to-specimen carryover (5). To address this issue, recent attempts have been made to circumvent RNA extraction in COVID-19 testing by performing RT-qPCR directly on heat-treated subject samples (65°C for 30 min or 95°C for 10min) or directly loading patient swab medium into RT-PCR reaction mix. Using heat-treatment approach the sensitivity ranged from 92 to 96% and specificity from 93 to 100% (6). Here, we tested the direct method of SARS-CoV-2 RT-qPCR on heat-treated (Hit-RT-PCR) nasopharyngeal swab samples and compared the results with RNA-extraction based RT-PCR results.

This study was conducted at the clinical laboratory of Institut Pasteur of M'sila, Algeria. Nasopharyngeal swabs (NP) from patients with high likelihood for COVID-19 were collected by medical infectiologists and deposited in viral transport medium at different healthcare institution of the city of M'sila. Arrived to the laboratory, samples were stored at -20°C until extracted and tested within 72h. For routine analysis, RNA was extracted from 140 µL of NP samples using the QIAamp Viral RNA Mini kit. Reverse

transcription and quantitative PCR were performed using the Biogerm® novel Coronavirus (2019-nCoV) nucleic acid kit following the manufacturer instructions: Total reactions of 25µl were obtained by mixing 20µl of master mix (primers and probe mix: ORF1ab, N and RNase P genes) and 5 µl of clinical sample to fill the reaction. The thermal cycling steps were: stage1: 50°C for 10 min, stage2: 95°C for 5 min, stage3: 95°C for 10 sec, 55°C for 40 sec, 40 cycles. The RT-qPCR was performed on a Rotor-Gen Q real time PCR machine (Qiagen®) using the Rotor-Gen Software v2.3.

We initially aimed to validate heat-treatment method to get an accurate view of its performance in a real world clinical diagnostic setting. We blindly heated a panel of aliquots from 60 NP samples representing intermediate (CT of 20 - 30) and low (CT of more than 30) viral RNA loads by direct RT-qPCR. The SARS-CoV-2 Ct levels (ORF1ab and N) in these samples were previously determined by RT-qPCR that included RNA extraction (Ct cutoff ≤38).

NP swab samples were thermally treated in water bath at 65°C for 30 min. Samples were then placed in room temperature for 15 min, vortexed for 10 seconds, centrifuged at 1000g for 1 min and 5µl of the supernatant was directly loaded into RT-qPCR reaction. Comparably, aliquots from 161 NP samples were subjected to heat-treatment but with increasing heating time to 60 min.

An agreement analysis (positive and negative percent agreement) were applied between diagnostic results of our experiment and results obtained by the conventional SARS-CoV-2 testing protocol. Diagnostic results were considered as categorical variables (1 for the presence of SARS cov2 infection and 0 for the absence of infection). All statistical analysis were performed using R version 3.6.0 (R Core Team, 2014) (7). In this work we used anonymized material from

samples that had been collected for clinical diagnostics of SARS-CoV-2.

We found a weak agreement when NP samples were heated for 30min (PPA: 58%, 95%CI: 45 to 69%). But, the agreement increased (PPA: 78%, 95%CI: 70 to 84%) when we increased the heating time to (60 min). We also found a substantial agreement between N gene results of extracted and heat-inactivated samples (overall agreement 78%, 95%CI 70 to 83%) but a weak agreement for ORF1ab gene (overall agreement 45%, 95%CI 37 to 52%). Ct values of N gene for hit-RT-qPCR samples were higher than for RNA eluates of the same samples (mean difference =1.9 Ct). Surprisingly, three samples were identified as COVID-19 positive by 60 min heat-treatment RT-qPCR (one sample positive for N and ORF1ab and two for only N) but were negative on extracted RNA. Figure 1 and 2 show the full results of this experiment while Table 1 provides a summary.

Clinical laboratories of the developing world are overwhelmed with COVID-19 testing demands. As a means to validate heat-treatment RT-PCR method in our clinical laboratory, we have shown that prior heating at 65°C for 30 min was less accurate compared to prior heating at 65°C for 60 min. Our observation were not corroborated by previous results which showed that prior heating at 65°C for 30 min was adequate to correctly identify 92 to 96% of screened samples. This could be explained by difference in the composition of viral transport medium used (Inhibitory agents from the swab and medium may inhibit RT-qPCR) or a mutations in the Algerian strain of SARS-cov2, rendering the virus more resistant to heat-treatment. Our improved protocol correctly identified 100% of clinical samples with viral load between (20 and 30 Ct). The only samples missed were those among lower Ct range (Ct> 30). Of the 2065 cases with a positive diagnosis at "Institut Pasteur of M'sila" by our clinical laboratory at the time of writing, only 27% would fall in this low Ct range,

which demonstrate that our improved protocol will accurately detect the majority of COVID-19 patients. Evidence that analytical sensitivity of heat-treatment RT-PCR was inferior (higher Ct values) compared to extraction-based RT-qPCR is that heating for long time may degrade RNA in presence of metal ions and/or RNases and that more RNA was loaded for eluates compared to Hit-RT-PCR. Furthermore, the higher performance of primers and probes targeting short amplicon (N, 110 bp) confirmed previous reports. Hence, short amplicons targets may be more suitable for Hit-RT-qPCR protocol.

A surprising finding was that heat-treatment RT-PCR identified three samples as COVID-19 positive while they had been identified as COVID-19 negative by conventional protocol. The Ct values of heat-treatment RT-PCR samples were high (> 30) suggesting one possible explanation of this phenomenon: NP samples may had very low viral RNA load that was below the limit of detection - i.e the lowest concentration level with a detection rate of 95% for positive results-of the RT-PCR kit (1000 copies/ml) (9). So, negative results in patients with typical symptoms of COVID-19 may become detectable by repeating the test. Unfortunately, we were unable to confirm COVID-19 positivity by collection of a new swab samples.

In summary, we have shown that screening for SARS-CoV-2 infection by RT-qPCR could be achieved through heat-treatment protocol (65°C for 60 min) without the use of RNA extraction kits, in the studied population. We hypothesize that each clinical laboratory should validate its own heat-treatment protocol which may be specific to the pre-analytical (viral transport medium composition) and environmental factors. Previous reports suggest that initial negative result by heat-treatment RT-PCR should be repeated by RNA extraction for: symptomatic patients, healthcare personnel, and others with a high suspicion of COVID-19 (8). However,

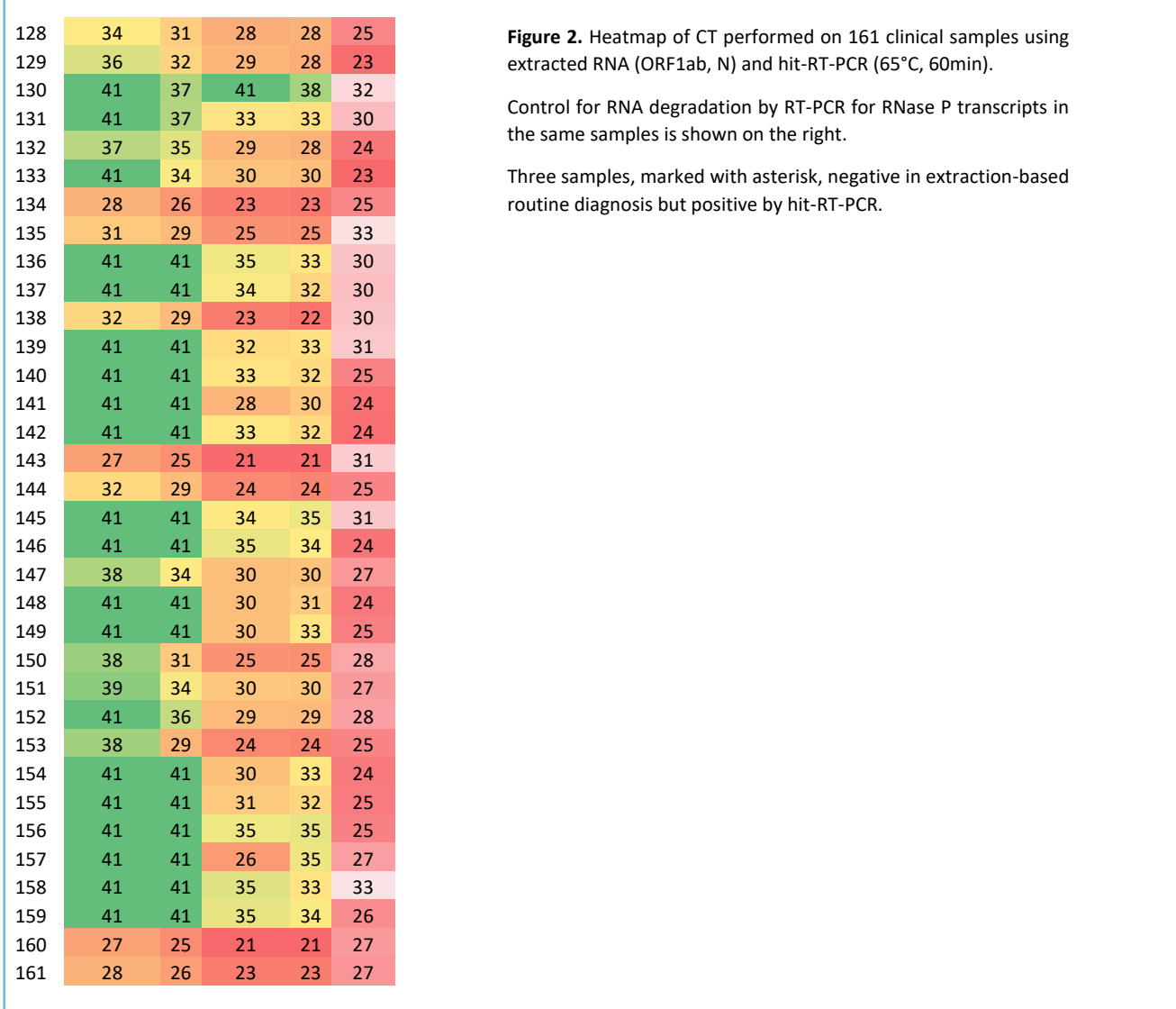
**Figure 1** Heat-map of ORF1ab and N Ct values for (65°C, 30min) protocol

ID	65°C,30min		Eluate		Rnase
	ORF1ab	N	ORF1ab	N	P
1	38	39	29	27	24
2	41	41	28	28	24
3	41	41	35	31	31
4	41	41	35	31	25
5	38	36	26	25	31
6	41	35	27	26	24
7	41	41	32	30	27
8	41	41	30	28	24
9	41	41	30	29	25
10	41	41	28	26	28
11	41	41	27	26	27
12	24	22	41	22	28
13	41	41	29	28	25
14	32	28	25	24	24
15	34	33	31	30	25
16	41	38	35	32	25
17	41	37	33	32	27
18	41	37	36	32	30
19	35	33	30	28	26
20	41	41	36	33	27
21	41	35	32	30	27
22	35	37	35	35	30
23	25	23	23	22	25
24	41	41	30	33	24
25	26	23	23	22	24
26	28	25	25	24	23
27	34	27	26	25	25
28	35	32	31	29	29
29	36	36	32	35	26
30	41	37	36	33	30
31	35	33	39	34	24
32	41	41	35	33	26
33	41	41	37	35	27
34	41	41	36	36	25
35	23	24	22	23	26
36	33	32	28	28	30
37	37	35	33	32	29
38	41	41	35	34	24
39	29	28	23	24	25
40	41	41	41	39	29
41	41	41	36	35	26
42	33	32	28	28	25
43	30	29	25	26	25
44	41	41	36	37	26
45	29	29	26	26	28
46	41	41	37	35	29
47	30	28	25	24	23
48	32	31	26	26	30
49	28	27	23	23	30
50	24	25	22	22	24
51	36	37	30	31	24
52	37	36	21	21	25
53	41	41	34	34	27
54	34	32	29	29	25
55	30	28	23	23	28
56	23	24	23	23	25
57	41	41	34	32	28
58	41	41	33	32	25
59	41	41	41	37	23
60	41	41	31	31	32

**Figure1.** Heatmap of CT performed on 60 clinical samples using extracted RNA (ORF1ab, N) and hit-RT-PCR (65°C, 30min). Control for RNA degradation by RT-PCR for RNase P transcripts in the same samples is shown on the right.

**Figure 2** Heat-map of ORF1ab and N Ct values for (65°C, 60min) protocol

ID	65°C,60min		Eluate		Rnase P	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58	59	60	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84*	85	86	87	88	89	90	91	92	93	94	95	96	97	98	99	100	101	102	103	104	105	106	107	108	109	110	111	112	113	114	115	116	117	118	119	120	121	122	123	124	125	126	127
	ORF1ab	N	ORF1ab	N																																																																																							
1	34	27	27	28	24	41	41	26	24	23	25	85	41	35	36	33	25																																																																										
2	41	35	32	31	24	41	41	26	24	25	25	86	41	41	36	33	25																																																																										
3	41	34	35	34	31	41	41	26	25	25	26	87	41	41	41	41	27																																																																										
4	41	36	36	35	25	41	41	35	33	33	28	88	31	25	27	26	33																																																																										
5	32	27	33	32	31	41	41	38	36	36	29	89	35	29	27	26	26																																																																										
6	28	24	23	24	24	41	41	22	22	22	23	90	41	35	34	32	27																																																																										
7	41	32	32	32	27	41	41	36	30	30	32	91	41	32	31	29	27																																																																										
8	27	23	23	23	24	41	41	41	36	34	30	92	27	23	22	22	30																																																																										
9	26	22	23	23	25	41	41	36	38	36	24	93	41	38	41	35	25																																																																										
10	41	31	30	30	28	41	41	32	32	31	24	94	41	32	31	29	24																																																																										
11	30	24	24	24	27	41	41	22	22	22	25	95	41	23	22	21	24																																																																										
12	29	23	24	24	28	41	41	23	22	22	27	96	41	37	35	31	23																																																																										
13	41	33	33	32	25	41	41	41	34	34	25	97	41	35	26	27	25																																																																										
14	37	30	28	29	24	41	41	24	24	24	28	98	41	29	36	34	29																																																																										
15	41	41	41	41	25	41	41	33	32	32	25	99	41	27	24	24	26																																																																										
16	41	41	41	41	25	41	41	36	36	36	28	100	33	28	27	26	35																																																																										
17*	36	32	41	41	27	41	41	27	26	25	25	101	34	28	27	26	24																																																																										
18	41	41	41	41	33	41	41	37	38	38	23	102	41	31	29	27	26																																																																										
19*	41	34	41	41	26	41	41	32	30	30	32	103	38	32	30	28	27																																																																										
20	41	31	41	36	27	41	41	29	26	27	30	104	41	36	27	26	25																																																																										
21	41	32	31	32	27	41	41	23	34	34	24	105	41	23	22	22	26																																																																										
22	32	27	41	37	30	34	29	25	24	23	23	106	41	41	37	35	30																																																																										
23	41	41	31	31	25	32	26	26	23	25	25	107	41	41	41	41	29																																																																										
24	41	30	27	27	24	38	29	26	25	33	33	108	41	38	36	35	24																																																																										
25	41	34	35	33	24	41	41	38	41	33	30	109	41	41	41	41	25																																																																										
26	28	23	23	23	23	41	41	28	25	24	30	110	41	22	22	22	29																																																																										
27	27	29	28	28	25	38	32	27	26	30	30	111	41	22	22	22	26																																																																										
28	41	36	33	33	29	41	41	28	25	24	31	112	41	27	22	23	25																																																																										
29	41	30	25	23	26	41	41	41	41	41	24	113	41	29	34	34	25																																																																										
30	41	36	33	33	30	41	41	41	41	41	24	114	41	41	35	35	26																																																																										
31	41	41	41	41	24	41	41	33	29	29	24	115	41	41	38	36	28																																																																										
32	41	39	41	41	26	33	26	24	23	31	31	116	25	24	22	23	29																																																																										
33	30	28	26	26	27	41	41	38	41	35	25	117	41	41	41	36	23																																																																										
34	41	41	41	41	25	41	41	31	34	32	31	118	41	41	41	41	32																																																																										
35	41	41	41	36	26	41	41	31	34	32	31	119	41	41	41	41	30																																																																										
36	41	41	41	37	30	41	41	41	37	33	24	120	41	41	41	41	30																																																																										
37	41	41	41	41	29	36	33	29	28	24	24	121	30	25	24	23	24																																																																										
38	41	30	28	28	24	41	41	41	41	41	25	122	30	25	24	23	24																																																																										
39	41	41	41	38	25	41	41	41	41	41	25	123	34	29	25	25	25																																																																										
40	41	37	41	37	29	41	41	41	41	36	28	124	33	31	28	27	27																																																																										
41	41	30	29	29	26	41	41	31	31	30	27	125	41	41	36	36	25																																																																										
84*	41	30	29	29	26	41	41	33	36	34	28	126	41	36	37	35	28																																																																										
						41	41	33	37	35	25	127	31	29	25	25	25																																																																										
						41	41	32	41	41	24	127	36	35	32	32	28																																																																										



**Figure 2.** Heatmap of CT performed on 161 clinical samples using extracted RNA (ORF1ab, N) and hit-RT-PCR (65°C, 60min).

Control for RNA degradation by RT-PCR for RNase P transcripts in the same samples is shown on the right.

Three samples, marked with asterisk, negative in extraction-based routine diagnosis but positive by hit-RT-PCR.

**Table 1** Detection sensitivity of Hit- RT-qPCR for 30 min versus Hit- RT-qPCR for 60 min on NP samples containing a range of SARS-CoV-2 viral RNA loads

Viral RNA load (Ct)	Heat-inactivation time	
	30 min	60 min
20 - 30	26/34 (76%)	74/74 (100%)
>30	9/26 (34%)	38/70 (54%)
<b>Total</b>	<b>35/60 (58%)</b>	<b>112/144 (78%)</b>

based on recent evidence showing the oddity of SARS-CoV-2 that can be cultured in respiratory samples 9 days after symptom onset, notably in patients with mild disease, it appears that re-testing in such patients may not be necessary (10). Such a strategy would drastically reduce the need for RNA extraction for a substantial portion of future COVID-19 tests.



### **Acknowledgements**

We are grateful to Mekki Oussama who participated in sample handling, organization and performed experiments.



### **REFERENCES**

1. Taipale J. and Linnarsson S. To stop COVID-19, test everyone, repeatedly. Medium (2020). Available: at <https://medium.com/@sten.linnarsson/to-stop-covid-19-test-everyone-373fd80eb03b>.
2. Centers for Disease Control and Prevention & Division of Viral Diseases. CDC 2019- Novel Coronavirus (2019-nCoV) Real-Time RT-PCR Diagnostic Panel. Available at: <https://www.fda.gov/media/134922/download>.
3. Chu, D.K.W., et al. Molecular Diagnosis of a Novel Coronavirus (2019-nCoV) Causing an Outbreak of Pneumonia. Clin Chem (2020).
4. Corman, V.M., et al. Detection of 2019 novel coronavirus (2019-nCoV) by real-time RT-PCR. Euro Surveill 25 (2020).
5. Susan Sefers and Jonathan E. Schmitz (2013). Molecular Contamination and Amplification Product Inactivation. Tang, Y-W and Stratton CW (Eds). Advanced techniques in diagnostic microbiology. (pp. 505-525).
6. Ioanna Smyrlaki et al. Massive and rapid COVID-19 testing is feasible by extraction free SARS-CoV-2 RT-qPCR. BioRxiv. Available at: <https://doi.org/10.1101/2020.04.17.20067348>
7. R Core Team (2013). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. Available at: <http://www.R-project.org/>.
8. Bruce, E. A. et al. Direct RT-qPCR detection of SARSCoV-2 RNA from patient nasopharyngeal swabs without an RNA extraction step. bioRxiv 2020.03.20.001008 (2020) doi:10.1101/2020.03.20.001008.
9. Xueliang Wang et al. Limits of Detection of 6 Approved RT-PCR Kits for the Novel SARS Coronavirus-2 (SARS-CoV-2). Clinical Chemistry 0:0 1–2 (2020).
10. WHO. Scientific Brief: Criteria for releasing COVID-19 patients from isolation. Available at: <https://www.who.int/news-room/commentaries/detail/criteria-for-releasing-covid-19-patients-from-isolation>



### **Editor-in-chief**

**János Kappelmayer**

Department of Laboratory Medicine  
University of Debrecen, Hungary

### **Assistant Editor**

**Harjit Pal Bhattoa**

Department of Laboratory Medicine  
University of Debrecen, Hungary

### **Editorial Board**

**Khosrow Adeli**, The Hospital for Sick Children, University of Toronto, Canada

**Borut Božič**, University Medical Center, Ljubljana, Slovenia

**Edgard Delvin**, CHU Sainte-Justine Research Center, Montréal, Québec, Canada

**Nilda E. Fink**, Universidad Nacional de La Plata, Argentina

**Ronda Greaves**, Biochemical Genetics, Victorian Clinical Genetics Services, Victoria, Australia

**Mike Hallworth**, Shrewsbury, United Kingdom

**Andrea R. Horvath**, Prince of Wales Hospital and School of Medical Sciences, University of New South Wales, Sydney, Australia

**Ellis Jacobs**, EJ Clinical Consulting, LLC, USA

**Allan S. Jaffe**, Mayo Clinic, Rochester, USA

**Bruce Jordan**, Roche Diagnostics, Rotkreuz, Switzerland

**Gábor L. Kovács**, University of Pécs, Hungary

**Evelyn Koay**, National University, Singapore

**Tamas Kószegi**, University of Pécs, Hungary

**Janja Marc**, University of Ljubljana, Slovenia

**Gary Myers**, Joint Committee for Traceability in Laboratory Medicine, USA

**Tomris Ozben**, Akdeniz University, Antalya, Turkey

**Maria D. Pasic**, Laboratory Medicine and Pathobiology, University of Toronto, Canada

**Maria del C. Pasquel Carrera**, College of Chemists, Biochemists and Pharmacists, Pichincha, Ecuador

**Oliver Racz**, University of Kosice, Slovakia

**Rosa Sierra Amor**, Laboratorio Laquims, Veracruz, Mexico

**Sanja Stankovic**, Institute of Medical Biochemistry, Clinical Center of Serbia, Belgrade, Serbia

**Danyal Syed**, Ryancenter, New York, USA

**Grazyna Sypniewska**, Collegium Medicum, NC University, Bydgoszcz, Poland

**Peter Vervaart**, LabMed Consulting, Australia

**Stacy E. Walz**, Arkansas State University, USA



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

Copyright © 2020 IFCC. All rights reserved.

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.

This is a Platinum Open Access Journal distributed under the terms of the *Creative Commons Attribution Non-Commercial License* which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

Produced by:

 **Insoft Digital**  
Web Solutions

[epub@insoftdigital.com](mailto:epub@insoftdigital.com)

Published by:

  
**IFCC**  
International Federation  
of Clinical Chemistry  
and Laboratory Medicine

[www.ifcc.org](http://www.ifcc.org)