جامعة نايف العربية للعلوم الأمنية
Naif Arab University for Security Sciences

Naif Arab University for Security Sciences
Arab Journal of Forensic Sciences & Forensic Medicine
www.nauss.edu.sa
http://ajfsfm.nauss.edu.sa

الجمعية العربية لعلوم الأدلة الجنائية والطب الشرعي
Arab Society for Forensic Sciences and Forensic Medicine

Review Article

CrossMark

Biomarkers in Forensic Diagnosis of Sudden Cardiac Death (SCD)

المعِلمات (المؤشرات) الحيوية المستخدمة في تشخيص الطب الشرعي لموت القلب المفاجئ.

Saikat Das [1], Soumeek Chowdhuri [2,*], Ritwik Ghosh [1]

[1] *Final Professional MBBS, Calcutta National Medical College, India.*
[2,*] *Department of Forensic Medicine and Toxicology, Calcutta National Medical College, India.*

Received 26 Nov. 2018; Accepted 14 Apr. 2019; Available Online 23 May 2019

Abstract

Diagnosis of sudden cardiac death (SCD) is challenging for medical professionals. For this reason, to make diagnosis easier for forensic pathologists, there is a pressing need for the use of biomarkers. This article highlights biomarkers that can be used in the postmortem diagnosis of SCD.

Cardiac troponins, high-sensitivity C-reactive protein, and creatine kinase-MB have proven to be very useful for this purpose. Lactate dehydrogenase, myoglobin and tumor necrosis factor α, although useful, are not efficient enough to be included in the list of biomarkers for the diagnosis of SCD.

Previous studies have shown both positive and negative results for natriuretic peptides as a biomarker, and further studies are required to confirm its use as a biomarker for diagnosis of SCD in autopsy cases. In living subjects, a multi-marker strategy is useful in predicting risk of cardiovascular deaths. It is suggested that for the diagnosis of SCD, a multi-marker strategy may be more efficient. However, more studies are required to confirm this.

Keywords: *Forensic Science, Sudden Cardiac Death, Biomarkers, Forensic Diagnosis.*

المستخلص

يُعد تشخيص الموت القلبي المفاجئ (SCD) تحديًا للمختصين الطبيين. لهذا السبب، ولجعل التشخيص أسهل لأخصائيي الطب الشرعي، هناك حاجة إلى استخدام المُعلمات الحيوية لتشخيص تلك الحالات. وعليه تركز هذه المقالة على المعلمات الحيوية المحتملة التي يمكن أن تستخدم في تشخيص وفيات القلب المفاجئة (SCD).

أثبتت بعض المُعلمات الحيوية مثل تروبنونات القلب Cardiac troponins وبروتين سي التفاعلي عالي الحساسية high-sensitivity C-reactive protein والكرياتين كيناز-أم بي creatine kinase-MB أنها مفيدة جدا لهذا الغرض. أما إنزيمات اللاكتات Lactate dehydrogenase، والميوغلوبين myoglobin، وعامل النخر الورمي ألفا tumor necrosis factor α، رغم أنها مفيدة إلا أنها غير فعالة بما يكفي لإدراجها في قائمة المُعلمات الحيوية لتشخيص وحالات موت القلب المفاجئ SCD.كما أعطت الدراسات السابقة نتائج إيجابية وأخرى سلبية لببتيدات الناتريوتريك natriuretic peptides، كمؤشر حيوي وهناك حاجة إلى المزيد من الدراسات لتأكيد استخدامه كمؤشر حيوي لتشخيص SCD في حالات التشريح. للأشخاص الذين على قيد الحياة فإن استراتيجية استخدام عدة معلمات تكون مفيدة في التنبؤ بخطر وفيات القلب والأوعية الدموية. ومن المقترح أنه من أجل تشخيص حالات موت القلب المفاجئ SCD، قد تكون استراتيجية استخدام معلمات متعددة أكثر كفاءة ولذا فإن المزيد من الدراسات مطلوبة لتأكيد ذلك.

الكلمات المفتاحية: علوم الأدلة الجنائية، الموت القلبي المفاجئ، المُعلِّمات الحيوية، تشخيص الطب الشرعي.

Production and hosting by NAUSS

* Corresponding Author: Soumeek Chowdhuri
Email: smk.kgp@gmail.com

doi: 10.26735/16586794.2019.011

1. Introduction

Annually, 30% of global mortality, or 17 million deaths, occur due to cardiovascular death. This makes it the leading cause of deaths worldwide. Previous studies show that about 40-50% of all cardiovascular deaths are sudden cardiac deaths (SCD) [1]. In daily practice, SCD is a diagnostic challenge for forensic pathologists. SCD is defined as sudden, natural, and unexpected death due to cardiac or unknown causes that occurs, if witnessed, within 1 hour of symptom onset and if unwitnessed, 24 hours after the deceased was last seen alive and functioning normally [2,3]. Pathologists do not find it difficult to detect myocardial lesions using conventional methods. However, in the very early stage of infarction, positive pathological evidence cannot be detected in cases of sudden death. Diagnosis then depends on negative findings excluding other causes of death [4-6]. For these reasons, there is a pressing need for the use of biomarkers in the diagnosis of sudden cardiac death.

Here we have systematically reviewed the different articles published from 2000 to 2018 for this purpose from different databases of PubMed, Cochrane Library, etc. Keywords related to the study aim and included in the search string were: sudden cardiac death, biomarkers, forensic diagnosis. However, this article does not focus on potential biomarkers that predict SCD, but precisely highlights biomarkers that can be used in postmortem diagnosis of SCD needed for newer non-invasive technologies.

2. Cardiac Troponins (cTn)

Troponins are contractile muscle proteins. Cardiac troponins are specific for myocardium found in human cardiac muscle tissue. These are of two types: cardiac troponin T (cTnT) and cardiac troponin I (cTnI). Normally, traces of cTnT and cTnI are not found in blood. Their level rises very high for a long duration after myocardial injury: cTnI for 7-10 days and cTnT for 10-14 days [7]. Cardiac troponins are proven to be biomarkers for myocardial injury and so are expected to be indicative of sudden death due to cardiac complications.

Variation in the level of peripheral blood cTnT is seen in different postmortem cases. A rise in the level of peripheral cTnT depends on the survival period after the onset of an ischemic heart attack. Poor sensitivity of cardiac troponin is demonstrated within the first hours of chest pain [8,9], raising the doubt of its usefulness in cases of SCD due to early myocardial ischemia. However, blood cTnT levels rise more rapidly in cases of sudden death, because of greater myocardial damage than in long survival cases. Studies suggest that elevation in postmortem blood and pericardial cTnT levels in sudden cardiac death may depend on the severity of ischemic myocardial damage which involves multiple interstitial hemorrhages and necrosis, and also postmortem period for heart and pericardial levels [10]. In 2012, Reichlin et al. [11] developed more sensitive arrays for cardiac troponins (cTnTs), which allow detection of myocardial damage as early as 3-4 hours after the onset of chest pain [12].

The percentage of cTnT degradation shows a pseudo-linear relationship with the time since death and can be used to estimate postmortem interval [13]. Degradation of cTnT is not massive and can be used for other forensic purposes.

In recent years, studies have proven that in living subjects cTnT and cTnIare are very useful in the diagnosis of cardiac injury. These gave rise to the thought of investigating the efficiency of cardiac troponins in the postmortem diagnosis of SCD. Later postmortem levels have also proven to be very useful to support SCD diagnosis in the first 12 hours after death [14]. However, a similar rise in the level of cTnT is seen in cases of death due to multiple trauma and mechanical asphyxia [12]. Troponin is also detectable in many normal individuals, for instance after strenuous exercise [15]. This leads to a lack of specificity of cTnT in cases of SCD, which can be improved if other causes of death can be ruled out and data extensively analyzed from circumstantial settings and clinical data [16].

3. High-sensitivity C-reactive protein (hs-CRP)

In blood, an increased level of CRP is seen in virtually all types of pathological situations which lead to acute inflammation [17-19]. 12 hours after the onset of inflammatory reactions, elevated levels of CRP can be determined in blood [20]. Quantification of CRP in post-mortem blood has been done sporadically [23]. Postmortem CRP values were compared with CRP values analyzed within 24 hours before death. It was found that elevated levels of CRP in post-mortem blood are a good marker for ongoing inflammation processes before death [24]. These suggest that levels of CRP in blood can be used as a biomarker in forensic diagnosis.

Sudden unexplained cardiac death (SUCD) can occasionally occur in nonelderly patients with epilepsy, psychiatric disorders, or no medical history. Studies were conducted to analyze if values of biomarkers for heart failure are associated with that in SUCD [25]. Serum hs-CRP levels were found to be useful in diagnosis of SCD.

4. Creatine Kinase-MB (CK-MB)

CK-MB is obtained mainly from cardiac muscles and extra-cardiac tissue in significant amounts. Myocardial damage shows a considerable rise in levels of CK-MB. It has two forms: CK-MB2 is a myocardial form while CK-MB1 is an extracardiac form. A ratio of CK-MB2 and CK-MB1 above 1.5 is sensitive for diagnosis of acute MI [7]. It is a potential biomarker for the diagnosis of SCD.

Compared with death due to other causes, the highest levels of CK-MB were reported in cases of SCD due to ischemic heart disease [26]. CK-MB was found to be significantly higher (at least 2.4-fold) compared to the sudden cardiac death with death due to other non-cardiac origins [Unclear] [27]. CK-MB proved to be useful in the diagnosis of SCD in autopsy cases.

5. Lactate Dehydrogenase (LDH)

Amongst two forms, LDH-1 is myocardial-specific. A ratio of LDH-1 and LDH2 above 1 is indicative of MI. LDH levels rise after 24 hours, reach their peak in 3-6 days and return to normal after 14 days [7]. Commonly, LDH and its isoenzymes are productive in the diagnosis of acute MI [27-29]. Studies have shown the usefulness of LDH in post-mortem diagnosis of MI in different body fluids [30-39]. These indicate that LDH can be a potential biomarker for diagnosis of SCD in autopsy cases.

Studies showed that LDH is not useful in post-mortem diagnosis in SCD due to ischemic heart disease [26]. Also, in autopsy cases, no significant difference was found between the level of LHD in SCD and other cases of death [14]. In spite of being an efficient biomarker in diagnosis of MI, LDH did not prove to be useful in diagnosis of SCD in autopsy cases.

6. Atrial and Brain Natriuretic Peptides (ANP and BNP)

Myocardial natriuretic peptides play an important role as markers of cardiac strain. The biological response of natriuretic peptides is observed in heart diseases and can be used to demonstrate cardiac dysfunction even after death [40].

In 2014, Hashimoto et al. [41] presented a case of SCD in a patient with epilepsy. The postmortem serum showed an extremely high N-terminal pro brain natriuretic peptide (NT-proBNP) level (3650 pg/mL vs. normal b125 pg/mL). This was the first report on a high serum NTproBNP level in sudden unexpected death in epilepsy (SUDEP), indicating heart failure.

However, later studies showed that natriuretic peptides were inefficient as a biomarker in diagnosis of SCD. Between 2006 and 2014, Kentaroet al. [25] conducted a case-control study aiming to analyze whether values of biomarkers for heart failure are associated with SUCD, and so serum concentrations of NT-proBNP were analyzed. However, the level of N-terminal pro brain natriuretic peptide did not show any significance. These suggest further investigations are required to find the role of natriuretic peptides as a biomarker in the diagnosis of SCD.

7. Myoglobin

Myoglobin is the first cardiac marker to shoot up [Unclear] after MI. Within 24 hours of attack of acute MI, myoglobin reduces to normal levels, as it rapidly gets eliminated through urine [7]. Although myoglobin proved to be a useful biomarker of MI, studies give negative results about its efficiency as a marker in diagnosis of SCD [14].

8. Tumor Necrosis Factor α (TNF-α)

Tumor necrosis factor is a mediator of acute inflammation and is seen in high levels before death while the inflammatory reactions are going on. TNF-α is a potential biomarker for diagnosis of SCD. However, studies suggested it is not as efficient for diagnosis of SCD [25]. Amongst a number of biomarkers, TNF-α is readily available in the laboratory and stable in post-mortem samples. However, it was not found to be significantly different in SCD and deaths due to other causes.

9. Blood Culture

Several studies have described the presence of systemic infections in life-threatening complications, including SCD [42, 43]. It is suggested that sudden unexpected death in infancy (SUDI) is caused by common bacterial toxins produced mainly by upper respiratory tract bacteria [44].

Studies have shown that positive blood culture, isolating pathogens, is a marker of systemic infection and indicates a high risk of SCD in chronic patients. In 2017, López-Amador et al. [45] conducted a study to explore the bacteriological profile of postmortem blood cultures in sudden cardiac deaths compared to other non-cardiac origin deaths. In the SCD group (n=20), cultures were positive to Escherichia coli (50%; 10/20), Staphylococcus aureus (20%; 4/20), Klebsiellapneumoniae (20%; 4/20), Candida albicans (15%; 3/20) and Staphylococcus epidermis (10%; 2/20). The non-cardiac origin death group (n=8) were positive to E. coli (25%; 2/8). This indicates that positive blood culture can be used as a biomarker in diagnosis of SCD. More research is needed to prove its efficiency.

10. Discussion

Inference obtained from laboratory findings of these markers greatly depends on the site of sampling from which the biomarkers are taken using syringes. Because of its close proximity to the myocardium, pericardial fluid is considered to be the most reliable source. It also has higher levels of cardiac biomarkers than cerebrospinal fluid, femoral blood or iliac venous blood [46].

There is no gender-related difference for the markers. No significant difference has been found in the levels of biomarkers between cases with and without cardiopulmonary resuscitation [47].

At the time of death, there is breakdown of vital activities due to the effect of autolysis [48], microbial degra-

Table 1- *Different biomarkers obtained from cadaveric fluids that are useful for diagnosis of SCD; biomarkers useful for detection of myocardial ischemia, but not useful for diagnosis of SCD; potential biomarkers with unconfirmed usefulness.*

BIOMARKERS	Cardiac troponins	Useful for diagnosis of SCD
	High-sensitivity C-reactive protein	
	Creatine kinase-MB	
	Lactate dehydrogenase	Useful for detection of myocardial ischemia, but not useful for diagnosis of SCD
	Myoglobin	
	Tumor necrosis factor α	
	Natriuretic peptides	Usefulness is not confirmed
	Dephosphorylated connexin 43 and Jun B	

dation and hemolysis. Redistribution of the cellular components takes place along the concentration gradient. For this reason, it is important to note the postmortem interval. Likewise, stability of cardiac troponins is maintained for up to 48 hours [49,50].

Poor sensitivity is noted for biomarkers for the first few hours, after which there is an elevation of the biomarkers. This depends upon the severity of infarction and thus does not have the limitation of being dependent upon using immunohistochemistry and histology. In the very early stage of infarction, positive pathological evidence cannot be detected in cases of sudden death. New technologies using biomarkers such as cTnThs (a new generation of highly sensitive arrays for cardiac troponins) can aid in the diagnosis of SCD due to early myocardial infarction more effectively than using conventional histological methods. However, the need to perform histology is still crucial and by far the most important method to detect a myocardial infarction in the forensic field. Both methods must be employed to confirm the diagnosis of SCD.

11. Conclusion

Diagnosis of sudden cardiac death (SCD) is difficult. To make the diagnosis easier for forensic pathologists, there is a pressing need for the use of biomarkers. Cardiac troponins, high-sensitivity C-reactive protein, and creatine kinase-MB have proven to be useful for this purpose. Lactate dehydrogenase, myoglobin and tumor necrosis factor α, although useful, are not efficient enough to be included in the list of biomarkers for the diagnosis of SCD. Previous studies gave both positive and negative results for natriuretic peptides as a biomarker, and further studies are required to confirm its use as a biomarker for diagnosis of SCD in autopsy cases. Early markers of myocardial ischemia such as dephosphorylated connexin 43 and Jun B [54] are seen to have rising concentration with progressive ischemia; however, their concentration levels in autopsy cases have not been studied.

In living subjects, a multi-marker strategy is useful in predicting risk of cardiovascular deaths [51-53]. A combination of several biomarkers is used for this purpose. This strategy proved to be successful in living subjects; however, their use in cadavers has not yet been examined. It is suggested that for the diagnosis of SCD, a multi-marker strategy would be more efficient. More studies are required to confirm this.

References

1. Mehra R. Global public health problem of sudden cardiac death. J Electrocardiol. 2007;40(6):S118-22. https://doi.org/10.1016/j.jelectrocard.2007.06.023
2. World Health Organization. 1992. International Classification of Diseases, 10th Revision (ICD-10). http://www.who.int/classifications/icd/en/.
3. Priori SG, Wilde AA, Horie M, Cho Y, Behr ER, Berul C, Blom N, Brugada J, Chiang CE, Huikuri H, Kannankeril P. HRS/EHRA/APHRS expert consensus statement on the diagnosis and management of patients with inherited primary arrhythmia syndromes: document endorsed by HRS, EHRA, and APHRS in May 2013 and by ACCF, AHA, PACES, and AEPC in June 2013. Heart rhythm. 2013;10(12):1932-63. https://doi.org/10.1016/j.hrthm.2013.05.014
4. Schoen FJ. Ischemic heart disease. In: Cotran RS, Kumar V, Robbins SL, editors. Robbins pathologic basis of disease. 5th ed. Phiadelphia: Saunders; 1994. p. 524–41.
5. Knight B. In: The pathology of sudden death. Forensic pathology. London: Edward Arnold; 1991. p. 444–72.
6. Zhu BL, Ishikawa T, Michiue T, Li DR, Zhao D, Oritani S, Kamikodai Y, Tsuda K, Okazaki S, Maeda H. Postmortem cardiac troponin T levels in the blood and pericardial fluid. Part 1. Analysis with special regard to traumatic causes of death. Leg Med. 2006;8(2):86-93. https://doi.org/10.1016/j.legalmed.2005.10.004
7. Mohan H. Textbook of Pathology. 7th ed. The Health Science Publishers; 1992. 414 p.
8. Jaffe, A. S., & Ordonez-Llanos, J. (2010). High sensitivity troponin in chest pain and acute coronary syndromes. A step forward?. Revista espanola de cardiologia, 63(07), 763-769. https://doi.org/10.1016/S0300-8932(10)70178-0

9. Reichlin T, Hochholzer W, Bassetti S, Steuer S, Stelzig C, Hartwiger S, Biedert S, Schaub N, Buerge C, Potocki M, Noveanu M. Early diagnosis of myocardial infarction with sensitive cardiac troponin assays. N Engl J Med. 2009;361(9):858-67. https://doi.org/10.1056/NEJMoa0900428
10. Zhu BL, Ishikawa T, Michiue T, Li DR, Zhao D, Kamikodai Y, Tsuda K, Okazaki S, Maeda H. Postmortem cardiac troponin T levels in the blood and pericardial fluid. Part 2: analysis for application in the diagnosis of sudden cardiac death with regard to pathology. Leg Med. 2006;8(2):94-101. https://doi.org/10.1016/j.legalmed.2005.10.003
11. Reichlin T, Twerenbold R, Reiter M, Steuer S, Bassetti S, Balmelli C, Winkler K, Kurz S, Stelzig C, Freese M, Drexler B. Introduction of high-sensitivity troponin assays: impact on myocardial infarction incidence and prognosis. Am J Med. 2012;125(12):1205-13. https://doi.org/10.1016/j.amjmed.2012.07.015
12. González-Herrera L, Valenzuela A, Ramos V, Blázquez A, Villanueva E. Cardiac troponin T determination by a highly sensitive assay in postmortem serum and pericardial fluid. Forensic Sci Med Pathol. 2016;12(2):181-188. doi:10.1007/s12024-016-9749-1. https://doi.org/10.1007/s12024-016-9749-1
13. Kumar S, Ali W, Singh US, Kumar A, Bhattacharya S, Verma AK, Rupani R. Temperature-Dependent Postmortem Changes in Human Cardiac Troponin-T (cTnT): An Approach in Estimation of Time Since Death. J Forensic Sci. 2016;61:S241-5. https://doi.org/10.1111/1556-4029.12928
14. Carvajal-Zarrabal O, Hayward-Jones PM, Nolasco-Hipolito C, Barradas-Dermitz DM, Calderón-Garcidueñas AL, López-Amador N. Use of cardiac injury markers in the postmortem diagnosis of sudden cardiac death. J Forensic Sci. 2017;62(5):1332-5. https://doi.org/10.1111/1556-4029.13397
15. Brush JE, Kaul S, Krumholz HM. Troponin Testing for Clinicians. J Am Coll Cardiol. 2016;68(21):2365-2375. https://doi.org/10.1016/j.jacc.2016.08.066
16. Barberi C, van den Hondel KE. The use of cardiac troponin T (cTnT) in the postmortem diagnosis of acute myocardial infarction and sudden cardiac death: A systematic review. Forensic sci int. 2018. https://doi.org/10.1016/j.forsciint.2018.09.002
17. Du Clos TW. Function of C-reactive protein. Ann med. 2000;32(4):274-8. https://doi.org/10.3109/07853890009011772
18. Young B, Gleeson M, Cripps AW. C-reactive protein: a critical review. Pathol. 1991;23(2):118-24. https://doi.org/10.3109/00313029109060809
19. Kushner I. C-reactive protein and the acute-phase response. Hospital practice (Office ed.). 1990;25(3A):13-6.
20. Peltola H, Jaakkola M. C-reactive protein in early detection of bacteremic versus viral infections in immunocompetent and compromised children. J pediatr. 1988;113(4):641-6. https://doi.org/10.1016/S0022-3476(88)80372-X
21. Benjamin DR, Siebert JR. C-Reactive protein and prealbumin in suspected sudden infant death syndrome. Pediatr pathol. 1990;10(4):503-7. https://doi.org/10.3109/15513819009067139
22. Rambaud C, Guibert M, Briand E, Grangeot-Keros L, Coulomb-L'Herminé A, Dehan M. Microbiology in sudden infant death syndrome (SIDS) and other childhood deaths. FEMS Immunol Med Microbiol. 1999;25(1-2):59-66. https://doi.org/10.1111/j.1574-695X.1999.tb01327.x
23. Quan L, Fujita MQ, Zhu BL, Ishida K, Maeda H. Immunohistochemical distribution of C-reactive protein in the hepatic tissue in forensic autopsy. Forensic sci int. 2000;113(1-3):177-82. https://doi.org/10.1016/S0379-0738(00)00259-0
24. Uhlin-Hansen L. C-reactive protein (CRP), a comparison of pre-and post-mortem blood levels. Forensic sci int. 2001;124(1):32-5. https://doi.org/10.1016/S0379-0738(01)00558-8
25. Kentaro S, Kimiharu I, Sari M, Shojiro T, Yuko K. Exploration of Predictive Biomarkers for Sudden Unexplained Cardiac Death in Nonelderly People: A Case-Control Study With Biochemical Parameters Related to Heart Failure. Am J Forensic Med Pathol.

2018;39(1):41-5.

26. Ghormade PS, Kumar NB, Tingne CV, Keoliya AN. Distribution & diagnostic efficacy of cardiac markers CK-MB & LDH in pericardial fluid for postmortem diagnosis of ischemic heart disease. J Forensic Leg Med. 2014;28:42-6. https://doi.org/10.1016/j.jflm.2014.09.011
27. Antman E, Bassand JP, Klein W, Ohman M, Sendon JL, Rydén L, Simoons M, Tendera M. Myocardial infarction redefined—a consensus document of the Joint European Society of Cardiology/American College of Cardiology committee for the redefinition of myocardial infarction: the Joint European Society of Cardiology/American College of Cardiology Committee. J Am Coll Cardiol. 2000;36(3):959-69. https://doi.org/10.1016/S0735-1097(00)00804-4
28. Lewandrowski K, Chen A, Januzzi J. Cardiac markers for myocardial infarction: a brief review. Pathol Patterns Reviews. 2002;118(suppl_1):S93-9.
29. Henderson AR, Moss DW. Enzymes. In: Burtis CA, Ashwood ER, editors. Tietz textbook of clinical chemistry. 5th ed. Philadelphia: Saunders Co.; 1986. p. 356e66.
30. Osuna E, Pérez-Cárceles MD, Vieira DN, Luna A. Distribution of biochemical markers in biologic fluids: application to the postmortem diagnosis of myocardial infarction. The Am J Forensic Med Pathol. 1998;19(2):123-8. https://doi.org/10.1097/00000433-199806000-00005
31. Carceles-Perez DM, Noeguera J, Jimenez JL, Martinez P, Luna A, Osuna E. Diagnostic efficacy of biochemical markers in diagnosis post-mortem of ischemic heart disease. Forensic Sci Int. 2004;142(1):1-7. https://doi.org/10.1016/j.forsciint.2004.02.007
32. Luna A, Villanueva E, Castellano MA, Jimenez G. The determination of CK, LDH and its isoenzymes in pericardial fluid and its application to the post-mortem diagnosis of myocardial infarction. Forensic Sci Int. 1982;19(1):85-91. https://doi.org/10.1016/0379-0738(82)90154-2
33. Luna A, Carmona A, Villanueva E. The postmortem determination of CK isozymes in the pericardial fluid in various causes of death. Forensic Sci Int. 1983;22(1):23-30. https://doi.org/10.1016/0379-0738(83)90116-0
34. Stewart RV, Zumwalt RE, Hirsch CS, Kaplan L. Postmortem diagnosis of myocardial disease by enzyme analysis of pericardial fluid. Am J Clin Pathol. 1984;82(4):411-7. https://doi.org/10.1093/ajcp/82.4.411
35. Osuna E, Perez MC, Jakobsson SW, Luna A. Biochemical and morphological markers in the post mortem diagnosis of ischemic heart distress. Acta Med Leg Soc (Liege). 1990;40:275-83.
36. Burns J, Milroy CM, Hulewicz B, West CR, Walkley SM, Roberts NB. Necropsy study of association between sudden death and cardiac enzymes. J Clin Pathol. 1992;45(3):217-20. https://doi.org/10.1136/jcp.45.3.217
37. Perez-Carceles MD, Osuna E, Vieira DN, Martinez A, Luna A. Biochemical assessment of acute myocardial ischaemia. J Clin Pathol. 1995;48(2):124-8. https://doi.org/10.1136/jcp.48.2.124
38. Batalis NI, Marcus BJ, Papadea CN, Collins KA. The role of postmortem cardiac markers in the diagnosis of acute myocardial infarction. J Forensic Sci. 2010;55(4):1088-91. https://doi.org/10.1111/j.1556-4029.2010.01368.x
39. Barabas B. CK-MB study in myocardial infarction and sudden cardiac death. Rom J Leg Med. 2009;17(2):156-62. https://doi.org/10.4323/rjlm.2009.156
40. Chen JH, Michiue T, Ishikawa T, Maeda H. Pathophysiology of sudden cardiac death as demonstrated by molecular pathology of natriuretic peptides in the myocardium. Forensic Sci Int. 2012;223(1-3):342-8. https://doi.org/10.1016/j.forsciint.2012.10.018
41. Hashimoto M, Nakajima M, Kuroda R, Yamaguchi R, Maeda H, Nagai H, Ikegaya H, Yoshida KI. Sudden unexpected death in a patient with epilepsy presenting with high N-terminal probrain natriuretic peptide level, cardiac lesions, and pulmonary edema. Int J cardiol. 2014;172(2):e265-7. https://doi.org/10.1016/j.ijcard.2013.12.069

42. Rae N, Finch S, Chalmers JD. Cardiovascular disease as a complication of community-acquired pneumonia. Curr Opin Pulm Med. 2016;22(3):212-8. https://doi.org/10.1097/MCP.0000000000000261
43. Brown AO, Millett ER, Quint JK, Orihuela CJ. Cardiotoxicity during invasive pneumococcal disease. Am J Respir Crit Care Med. 2015;191(7):739-45. https://doi.org/10.1164/rccm.201411-1951PP
44. Morris JA, Harrison LM, Biswas J, Telford DR. Transient bacteraemia: a possible cause of sudden life threatening events. Med hypotheses. 2007;69(5):1032-9. https://doi.org/10.1016/j.mehy.2007.02.039
45. López-Amador N, Alfonso-Bravo JA, Ortíz-Valdez K, Carvajal-Zarrabal O. Postmortem Blood Culture Profile in Sudden Cardiac Death: An Exploratory Study. J Forensic Sci. 2017; 5(2).
46. Palmiere C, Tettamanti C, Bonsignore A, De Stefano F, Vanhaebost J, Rousseau G, Scarpelli MP, Bardy D. Cardiac troponins and NT-proBNP in the forensic setting: Overview of sampling site, postmortem interval, cardiopulmonary resuscitation, and review of the literature. Forensic Sci Int. 2018;282:211-8. https://doi.org/10.1016/j.forsciint.2017.11.034
47. Chen JH, Inamori-Kawamoto O, Michiue T, Ikeda S, Ishikawa T, Maeda H. Cardiac biomarkers in blood, and pericardial and cerebrospinal fluids of forensic autopsy cases: A reassessment with special regard to postmortem interval. Leg med. 2015;17(5):343-50. https://doi.org/10.1016/j.legalmed.2015.03.007
48. Luna A. Is postmortem biochemistry really useful? Why is it not widely used in forensic pathology?. Leg Med. 2009;11:S27-30. https://doi.org/10.1016/j.legalmed.2009.02.040
49. Vanhaebost J, Ducrot K, de Froidmont S, Scarpelli MP, Egger C, Baumann P, Schmit G, Grabherr S, Palmiere C. Diagnosis of myocardial ischemia combining multiphase postmortem CT-angiography, histology, and postmortem biochemistry. La radiol medica. 2017;122(2):95-105. https://doi.org/10.1007/s11547-016-0698-2
50. Palmiere C, Tettamanti C, Bonsignore A, De Stefano F, Vanhaebost J, Rousseau G, Scarpelli MP, Bardy D. Cardiac troponins and NT-proBNP in the forensic setting: Overview of sampling site, postmortem interval, cardiopulmonary resuscitation, and review of the literature. Forensic Sci Int. 2018;282:211-8. https://doi.org/10.1016/j.forsciint.2017.11.034
51. St-Pierre AC, Cantin B, Bergeron J, Pirro M, Dagenais GR, Després JP, Lamarche B. Inflammatory markers and long-term risk of ischemic heart disease in men: a 13-year follow-up of the Quebec Cardiovascular Study. Atherosclerosis. 2005;182(2):315-21. https://doi.org/10.1016/j.atherosclerosis.2005.02.009
52. Wang TJ, Gona P, Larson MG, Tofler GH, Levy D, Newton-Cheh C, Jacques PF, Rifai N, Selhub J, Robins SJ, Benjamin EJ. Multiple biomarkers for the prediction of first major cardiovascular events and death. N Engl J Med. 2006;355(25):2631-9. https://doi.org/10.1056/NEJMoa055373
53. Kim HC, Greenland P, Rossouw JE, Manson JE, Cochrane BB, Lasser NL, Limacher MC, Lloyd-Jones DM, Margolis KL, Robinson JG. Multimarker prediction of coronary heart disease risk: the Women's Health Initiative. J Am Coll Cardiol. 2010;55(19):2080-91. https://doi.org/10.1016/j.jacc.2009.12.047
54. Sabatasso S, Mangin P, Fracasso T, Moretti M, Docquier M, Djonov V. Early markers for myocardial ischemia and sudden cardiac death. Int J Legal Med. 2016;130(5):1265-80. https://doi.org/10.1007/s00414-016-1401-9