



CardioCarePack – personalized medicine system for TDM of cardiological drugs based on LC-MS/MS analysis of samples collected at home with VAMS.

Rafał Szewczyk^{1, 2}; Adrianna Radulska³, Anna Lenartowicz²; Julia Mironenka²; Adrian Soboń^{1, 2}; Katarzyna Krupczyńska-Stopa^{1, 2}; Maciej Stopa^{1, 2}; Tomasz Borkowski³; Ewelina Marciniak³, Leszek Kalinowski³

¹Bioanalytic Sp. z o.o., Gdansk, Poland; ²LabExperts Sp. z o.o., Gdansk, Poland; ³ Medical University of Gdansk Gdansk, Poland

AGENDA

01

INTRODUCTION

Cardiac arrhythmia in numbers
Cardiac arrhythmia treatment
CardioCarePack principles

02

MATERIALS AND METHODS

Method development and validation
Clinical study

03

RESULTS

Method reproducibility
Serum to VAMS results correlation
QTc, therapeutic range, ICD & arrhythmia cases
IT system functionality

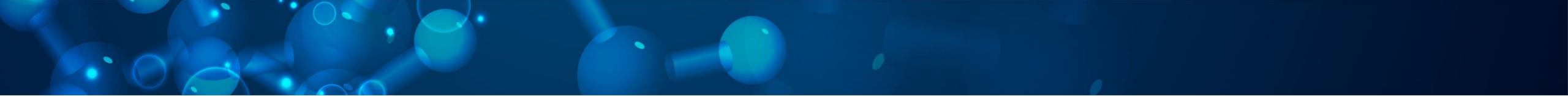
04

SUMMARY

CardioCarePack features

05

Q&A



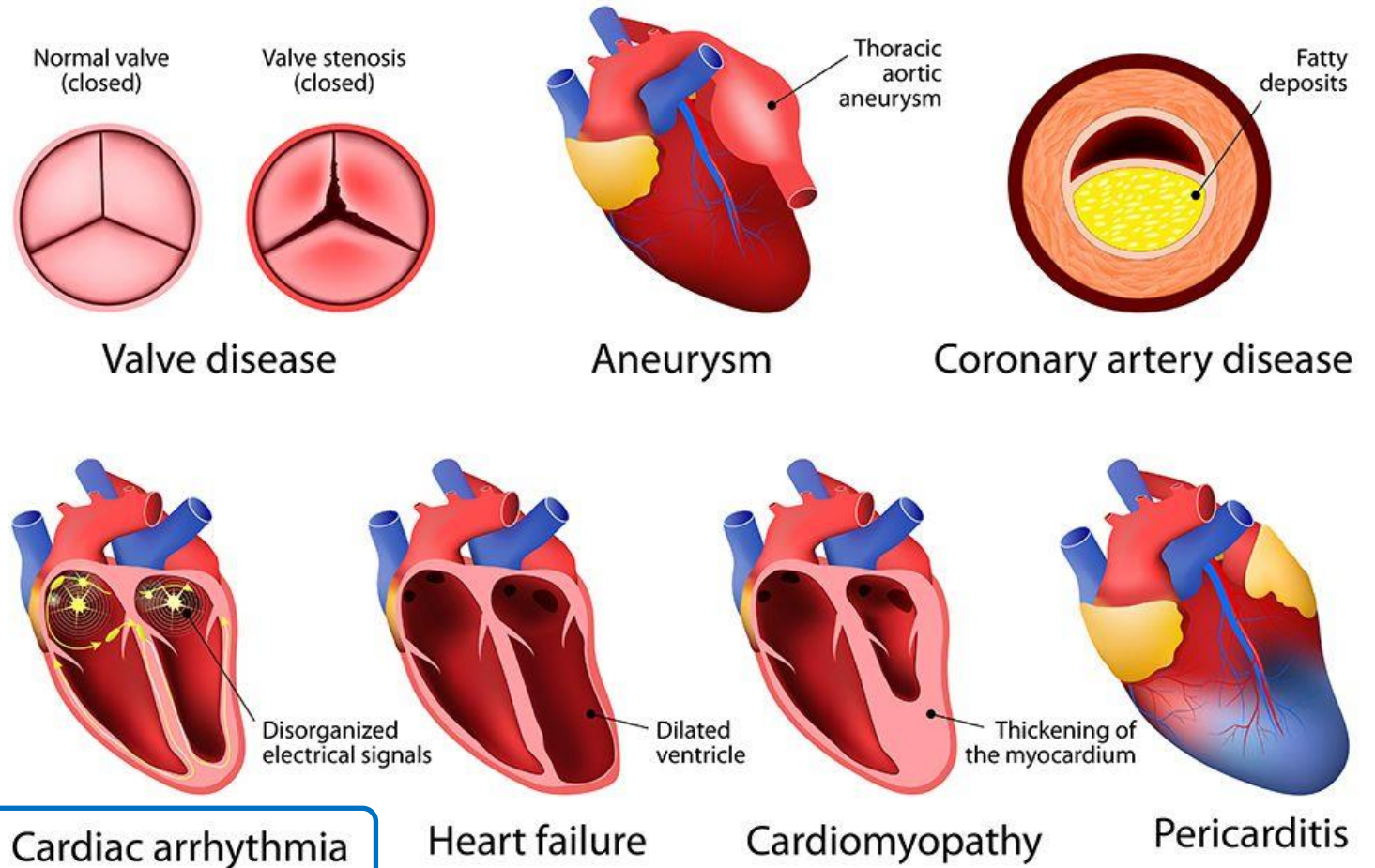
INTRODUCTION

Arrhythmia in numbers:

Bradycardia < 60 → 60-80 (normal) → 100 < Tachycardia

- Affects 2-3% of Europeans & Americans
- In Poland arrhythmia affects 1,8-2% of population (ca. 0,7-0,8 mln people)
- 80% of sudden cardiac deaths are caused by ventricular arrhythmias.
- In Europe there are ca. 13,3 mln of people, which should be effectively and safely treated.
- Due to the aging of the populations: in 2030 the number of patients with arrhythmia will increase to 14-17 million, while in 2040 it may be doubled.

Types of heart disease



Arrhythmia treatment

Typical therapeutic procedures



ABLATION

Low invasive procedure - 2-3 days in hospital. Effective in about 80% of cases. It is currently the treatment method of personal choice.



CARDIOVERTER OR DEFIBRILLATOR

These methods are used in cases of direct threat to life. Sometimes they require permanent implantation of such a device.



PHARMACOLOGICAL TREATMENT

The most common treatments are based on beta-blockers, glycosides or antagonists of specific enzymes and receptors.

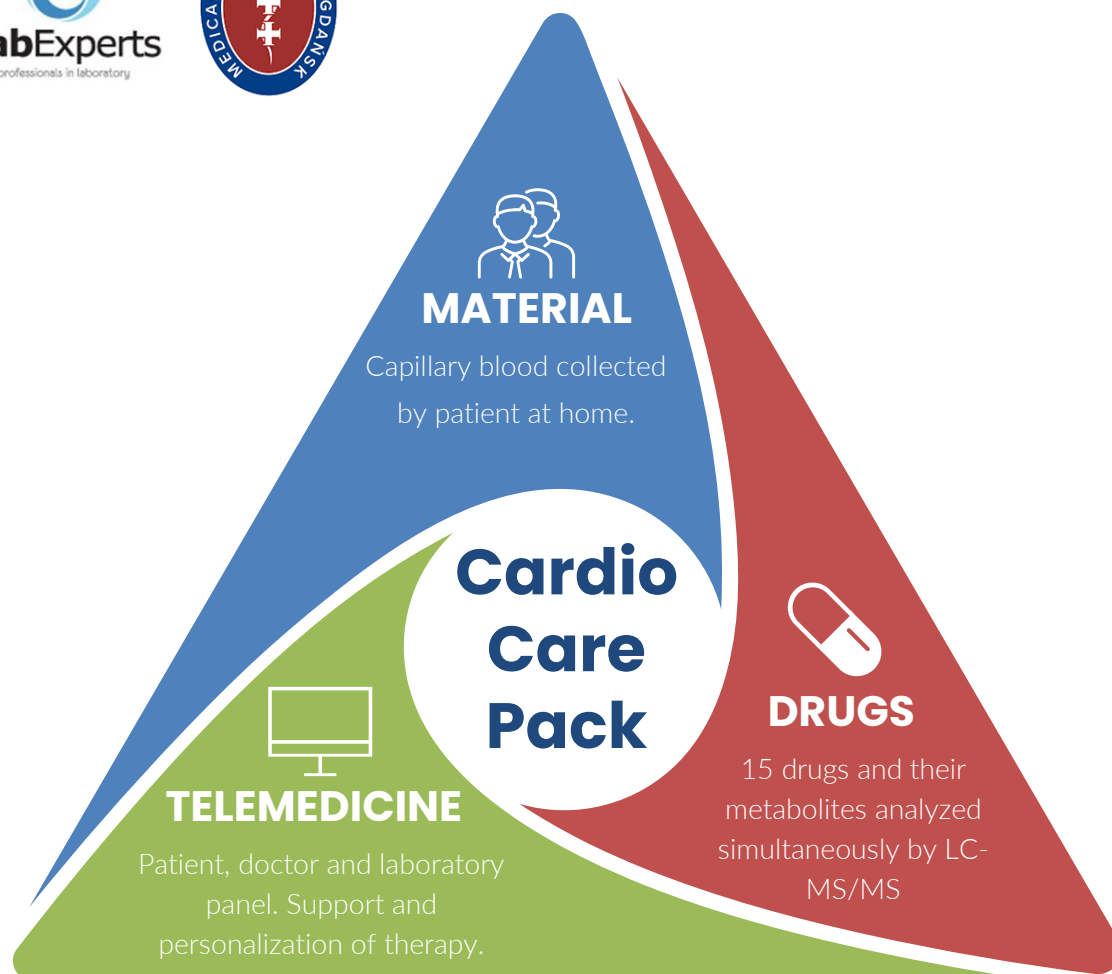


POIR.01.01.01-00-1196/19

CardioCarePack - A package of modern drug-monitored therapy solutions for patients with cardiac arrhythmias and development of personalized medicine.



Time frame – 3 years (2020-2023)



CAPILLARY BLOOD

Highly available service - the use of dry capillary blood, which the patient will collect at home.

DRUGS GROUP

Monitoring the concentration of 15 drugs and their metabolites in the blood during therapy of patients with cardiac arrhythmia

EXPERTS TELEMEDIC SYSTEM

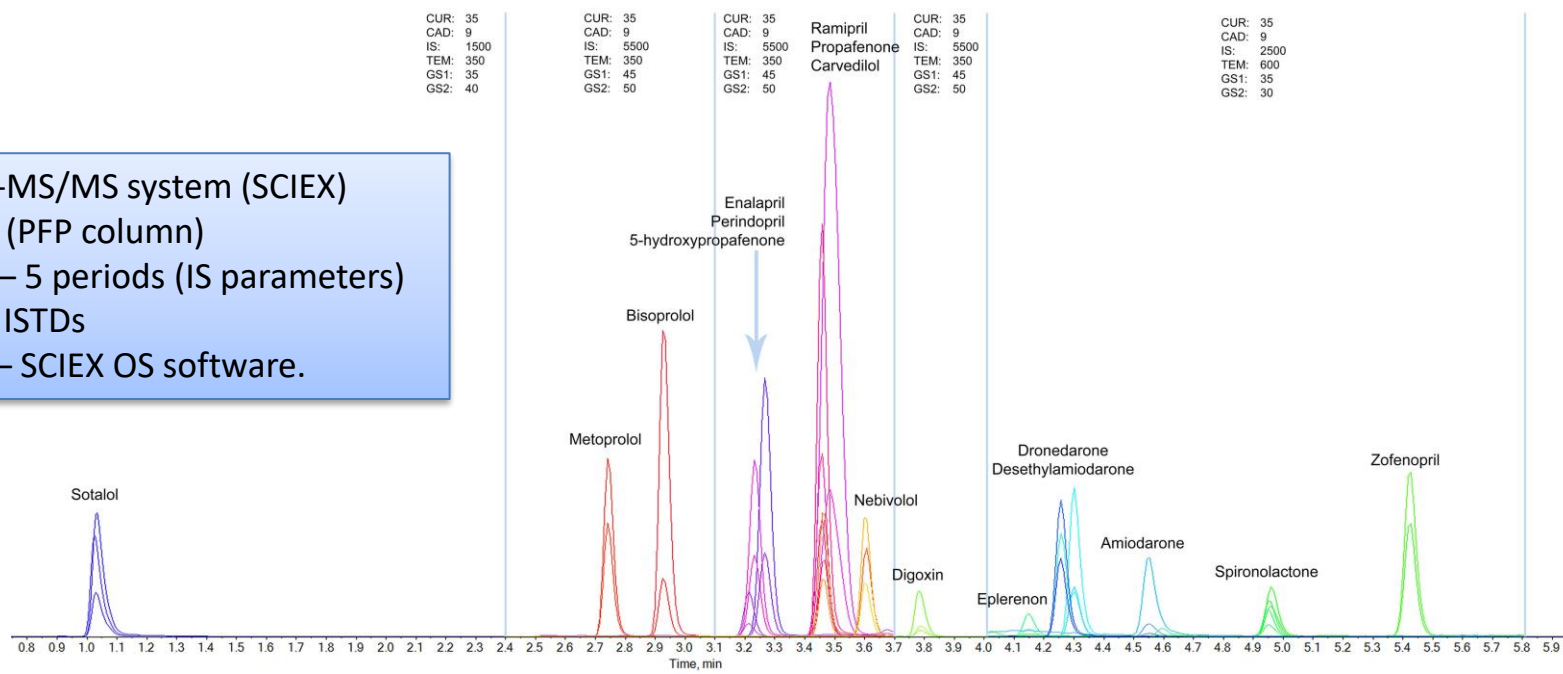
A tool that integrates all data between a doctor, patient and laboratory and supports the therapy process.



MATERIALS AND METHODS



QTRAP 5500+ LC-MS/MS system (SCIEX)
 RP-LC conditions (PFP column)
 MRM+ scanning – 5 periods (IS parameters)
 17 compounds + ISTDs
 Data processing – SCIEX OS software.



Parameter	Acceptance criteria	LC-MS/MS	Serum	Blood	Blood (Mitra®)
MRM pairs per compound	2	✓	-	-	-
MRM pairs per ISTD	1	✓	-	-	-
Sampling rate per chromatography peak	≥ 10	✓	-	-	-
LOD	S/N ≥ 3	-	✓	✓	✓
LLOQ (1 - 80 µg/L)	S/N ≥ 6	-	✓	✓	✓
Linearity	R ≥ 0.995	-	✓	✓	✓
Recovery (p < 0.05)	80-120%	-	✓	✓	✓
Precision (p < 0.05)	±10%	-	✓	✓	✓
Accuracy (p < 0.05)	100±10%	-	✓	✓	✓

Recommended transport conditions for Mitra samples: 20°C, desiccator, up to 5 days



Patients were divided into groups:

Finally, 324 persons included in the study

I - HF_rEF (heart failure with reduced ejection fraction) with ICD implant (100 persons):

- with Amiodaron based therapy (94 persons),
- 6 persons in control group (no amiodaron therapy).

II - patients with paroxysmal atrial fibrillation (206 persons):

- with the main anti-arrhythmic drug: amiodaron, propafenon, sotalol and digoxin (50 persons in each subgroup), where additional ADD could also be administered.
- 6 persons in control group (no pharmacological therapy).

Finger → Blood

Mitra[®] (20 μl)

Dry for 2-4h,
Pack and send to the lab

Sample preparation

Water with 0.1% formic acid
Sonication, 10 min., 37°C
ACN/MetOH (1:1) + ISTD, shaking for 20 min.
Centrifugation 15000 rpm
Evaporate under nitrogen, 50°C
Resuspension in 10% ACN with 0.1% formic acid
Sonicate, vortex and centrifuge 15000 rpm

LC-MS/MS

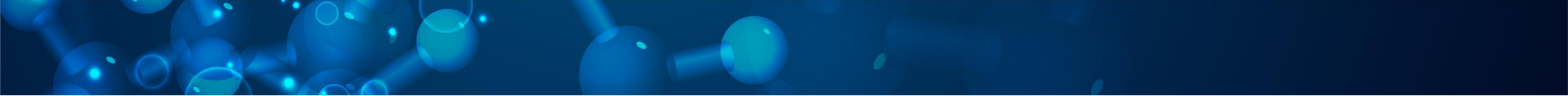
IT System

Medical facility – every 6 months – venous blood, dried blood (Mitra[®]), EKG, ASP, ALT, bilirubine, creatinine
At home – 1, 2 & 4 months after visit in medical facility – dried blood (Mitra[®])

2 years sample collection and analysis
Parralell IT system development

Data processing and interpretation
IT system integration

CardioCarePack ready for commercialization



RESULTS

Reproducibility - 80-120% ($p < 0.05$) determined on the basis of data obtained in an interlaboratory study.

Analyte	Venous blood (serum)			Capillary blood (Mitra®)		
	Reproducibility [%]	CV (%)	$p < 0.05$	Reproducibility [%]	CV (%)	$p < 0.05$
Amiodarone	108	2.27	✓	106	11.6	✓
5-OH-Propafenone	98.07	14.28	✓	94.56	22.09	✓
Bisoprolol	119.5	7.8	✓	115.4	17.97	✓
Carvedilol	98.17	5.88	✓	100.6	9.66	✓
Desethylamiodarone	101	6.73	✓	111.9	11.79	✓
Digoxin	104	4.79	✓	103.8	19.89	✓
Dronedarone*	N/A	N/A	N/A	N/A	N/A	N/A
Enalapril*	N/A	N/A	N/A	N/A	N/A	N/A
Eplerenone	115.7	7.8	✓	108.2	18.11	✓
Metoprolol	136.4	5.96	✓	132.8	20.05	✓
Nebivolol	103.8	2.89	✓	101.6	8.57	✓
Perindopril	112.6	17.46	✓	109.2	15.3	✓
Propafenone	99.9	6.43	✓	100.9	11.48	✓
Ramipril	87.92	19.68	✓	103.5	16.39	✓
Sotalol	99.28	4.06	✓	99.55	10.12	✓
Spironolactone	91.65	12.29	✓	96.45	19.13	✓
Zofenopril	75.49	34.27	✗	110.6	31.91	✗

QTRAP 5500+ vs 4500+
Different laboratory equipment
Samples analyzed „at once” vs
banking and queuing

* - no patients treated with this drugs during 2-years clinical study (N/A – not acquired)

Summarized data for anty-arrhythmic TDM with samples collected by VAMS.

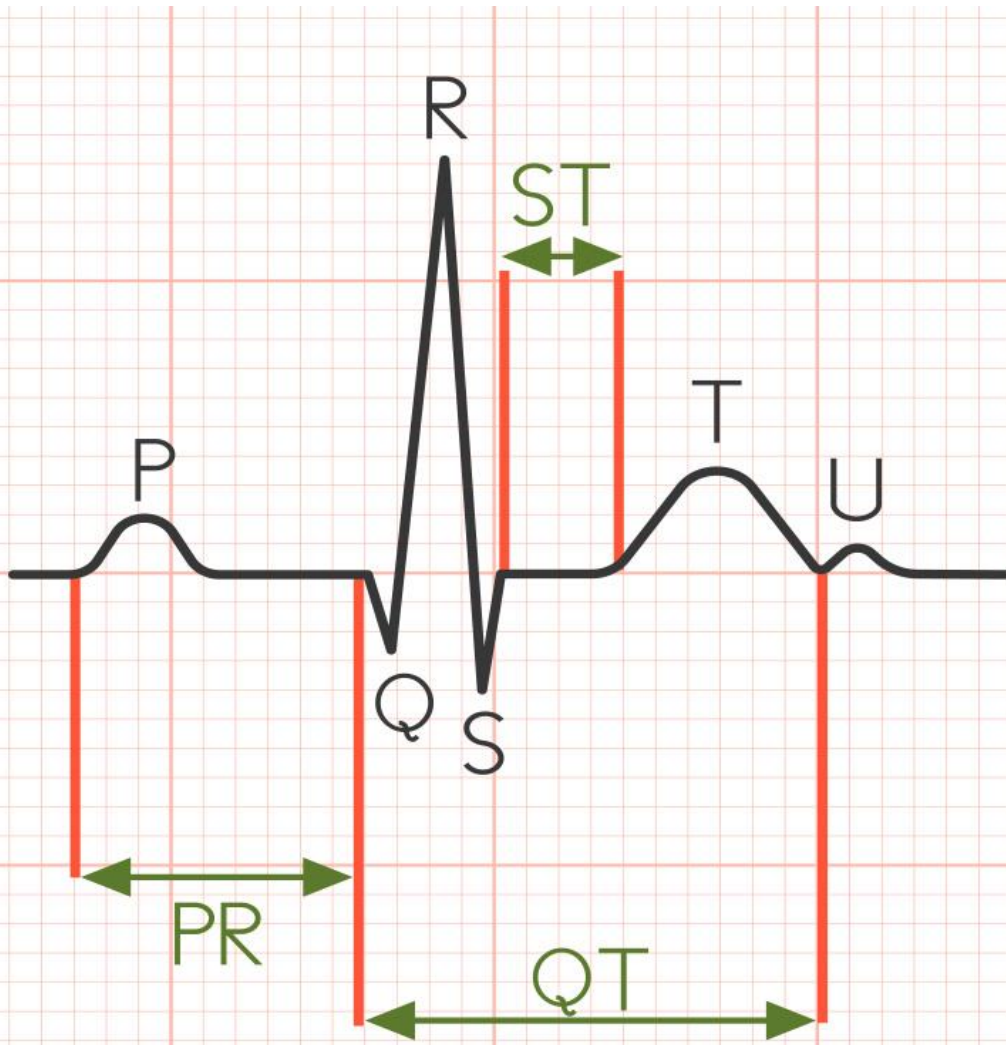
Analyte	Therapeutic index [µg/L]	Working range [µg/L]	LLOQ		Serum to Mitra ratio* (p<0.05)	
			[µg/L]	Signal to Noise	S/M ratio	%CV
Sotalol	1400 - 1700	10-2500	< 10	3406	0.94	6.05
5-OH Propafenone	153 - 337	50 -2500	< 10	2677	1	7.33
Amiodarone	1000 - 2500	10-2500	< 10	438	2.1	2.95
Bisoprolol	18323	2.5 - 250	< 1	438	1.06	4.35
Carvedilol	6.93 - 77	2.5 - 250	< 1	298	1.86	4.89
Desethylamiodarone	200 - 1000	10-2500	< 10	456	1.28	12.43
Digoxin	0.5 - 2	0.25 - 25	0.25	7	0.69	1.22
Dronedarone**	80 - 170	2.5 -250	< 1	127	N/A	N/A
Enalapril**	10 - 100	2.5 - 250	< 1	561	N/A	N/A
Eplerenone	200 - 1700	10-2500	< 1	95	1.78	5.12
Metoprolol	3 - 270	1 - 250	< 1	261	0.87	10.81
Nebivolol	0.5 - 1.5	0.25 - 25	0.1	6	1.04	1.71
Perindopril	80 - 150	10 -250	< 1	811	1.86	4.91
Propafenone	100 - 1000	1 - 250	< 1	1136	1.82	9.88
Ramipril	10 - 10000	2.5 - 250	< 1	435	1.15	7.90
Spironolactone	10 - 300	2.5 - 250	< 1	33	1.76	5.19
Zofenopril	50 - 170	2.5 - 250	< 1	57	2.38	34.43

S/M ratio is stable for all drug concentrations that we measured during the study.

S/M ratio can be used as a factor to estimate serum concertation.

* - S/M ratio is calculated on the basis of paralell analysis of serum and VAMS collected blood from patients in medical facility (four control tests every half a year during 2-years study)

** - no patientstreated with this drugs during 2-years clinical study



Correlation on QTc elongation in relation to drug concentration (within therapeutic range).

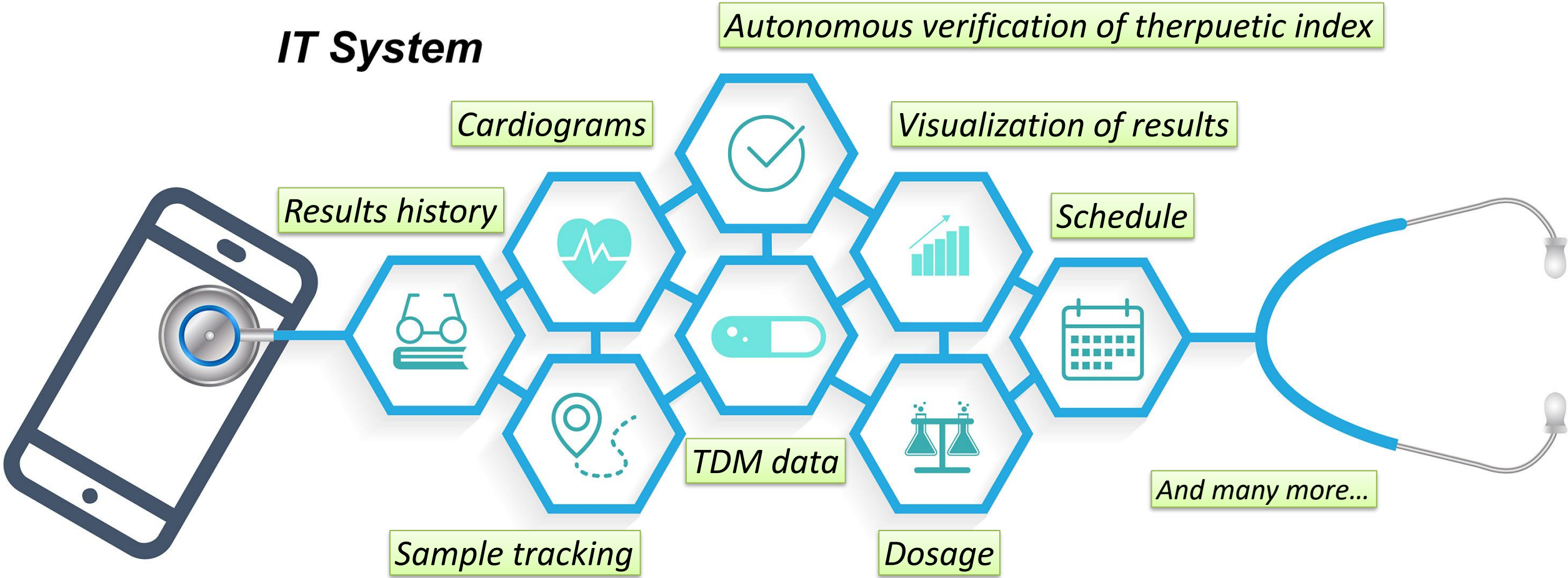
Antiarrhythmic drug & group	R	P – value (p < 0.05)
Amiodarone GR I	0,23	✓
Amiodarone GR II	0,19	✓
Sotalol GR II	0,29	✓
Propafenone + OH-propafenone GR II	0,43	✓
Digoxin GR II	-0,01	✗
Other tested compounds	±0.01	

The concentration of the compounds were maintained within the therapeutic range and never exceeded the upper limit of the norm.

Only:

- 11 patients with ICD or CRD-T interventions (defibrillations or stimulations) – 3.6% of the tested group,
- 31 patients with arrhythmia episodes – 10.1% of the tested group

IT System



Navigation: Karta pacjenta, Dokumenty

Truskawkowa Barbara

PESEL:

Umów wizytę

Zmień hasło, Wyloguj

Tab: Dane osobowe i medyczne, Przyjmowane leki, Wizyty, Komentarze lekarza, Załączniki, Próbk, Komunikaty

perpuetic index

ults

Lista pacjentów (0)

Wyszukaj pacjenta: Wpisz dane pacjenta

Otwórz filtry

Dodaj nowego pacjenta

ID	NAZWISKO I IMIĘ	GRL	DANE KONTAKTOWE	ADRES ZAMIESZKANIA	OSTATNIA WIZYTA	NASTĘPNA WIZYTA	KARTA PACJENTA
----	-----------------	-----	-----------------	--------------------	-----------------	-----------------	----------------

Lista pracowników (5)

Wyszukaj pracownika: Wpisz dane pracownika

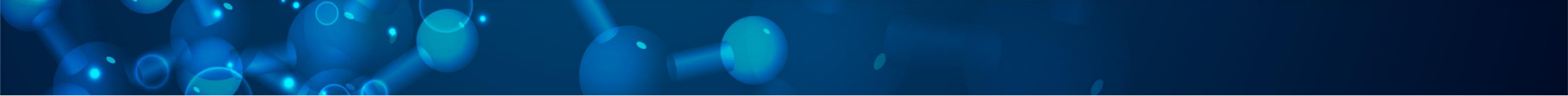
Dodaj nowego pracownika

ID	NAZWISKO I IMIĘ	ADRES E-MAIL	PLACÓWKA	TELEFON	OPERACJE
5	Mrozik Marta	martamrozik@gmail.com	12-346, Gdańsk ul. Kościuszki	+48 523147856	Zarządzaj,
4	Radulska Ada	adrianna.radulska@gumed.edu.pl	12-346, Gdańsk ul. Kościuszki	+48 485004545	Zarządzaj,
3	Muszak Monika	muszakmonika@gumed.edu.pl	12-346, Gdańsk ul. Kościuszki	+48 662150603	Zarządzaj,
2	Marciniak Ewelina	ewelina.marciniak@gumed.edu.pl	20-210, Gdańsk Niedźwiedzia	+48 536179279	Zarządzaj,
1	Pelechaty Grzegorz	gp@netcorelabs.pl	12-346, Gdańsk ul. Kościuszki	+48 730955565	Przywróć

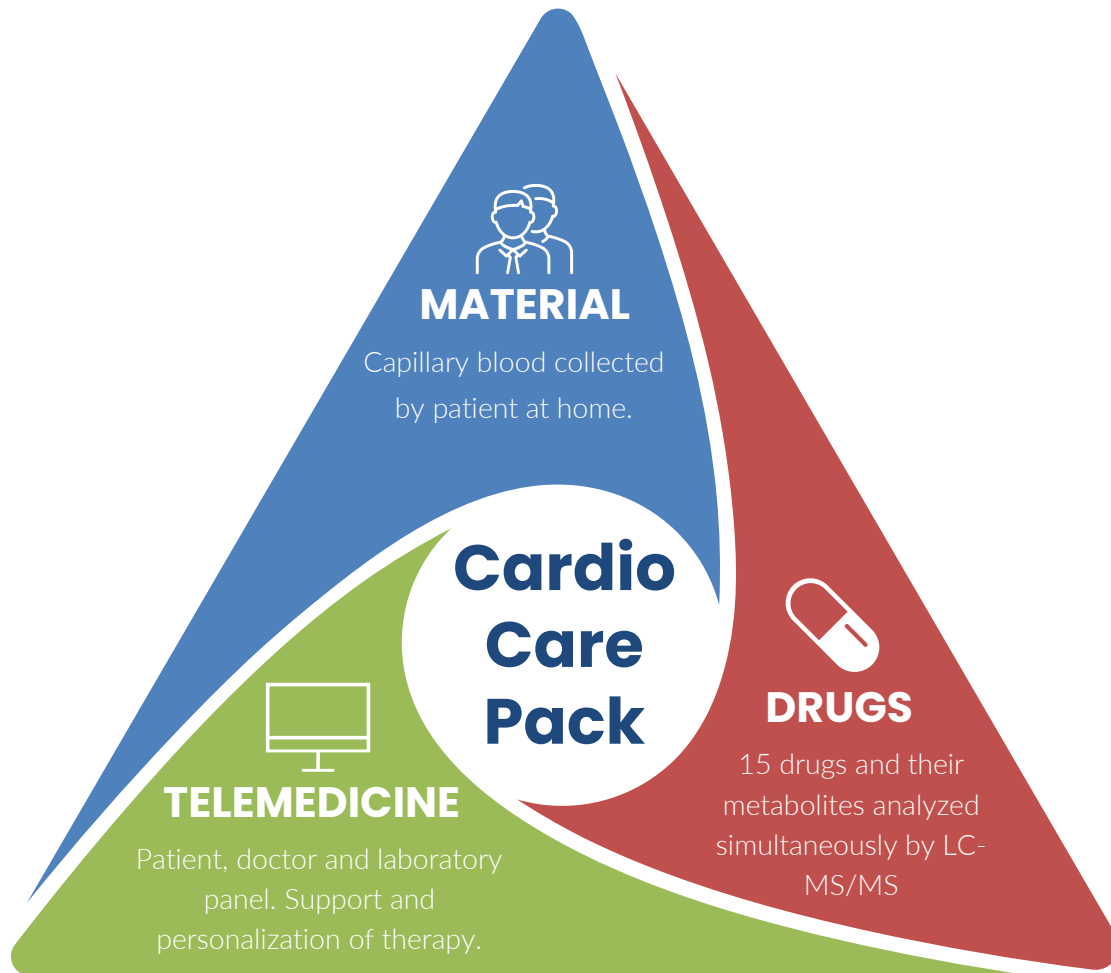
Wiersze na stronie: 20

1-5 z 5

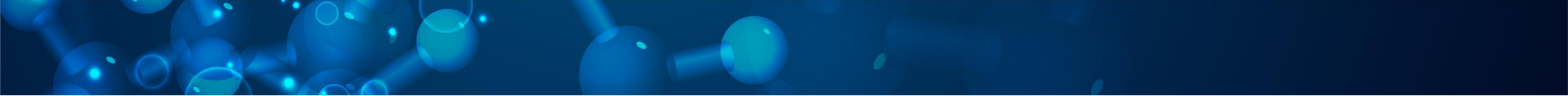




SUMMARY



- The developed procedures and methods for selected drugs determination in blood, serum and VAMS-collected blood met all validation criteria
- Recommended transport conditions for Mitra® samples: 20°C, desiccator, up to 5 days
- 324 persons included in a 2-year clinical study – serum & Mitra-based TDM, EKG and other tests
- There are no clinical data for Dronedarone and Enalapril (no patients with these drugs applied in therapy)
- Interlaboratory study showed excellent reproducibility of the assay (80-120% ($p < 0.05$), excluding Zofenopril)
- The concentration of the compounds were maintained within the therapeutic range and never exceeded the upper limit of the norm.
- Only 11 patients with ICD or CRD-T interventions (defibrillations or stimulations) – 3.6% of the tested group and only 31 patients with arrhythmia episodes – 10.1% of the tested group
- A server-based telemedical system where history, doses, therapeutic index flagging, cardiograms and other diagnostic results and data are available for patient, doctor and laboratory staff, respectively.



THANK YOU
Q&A