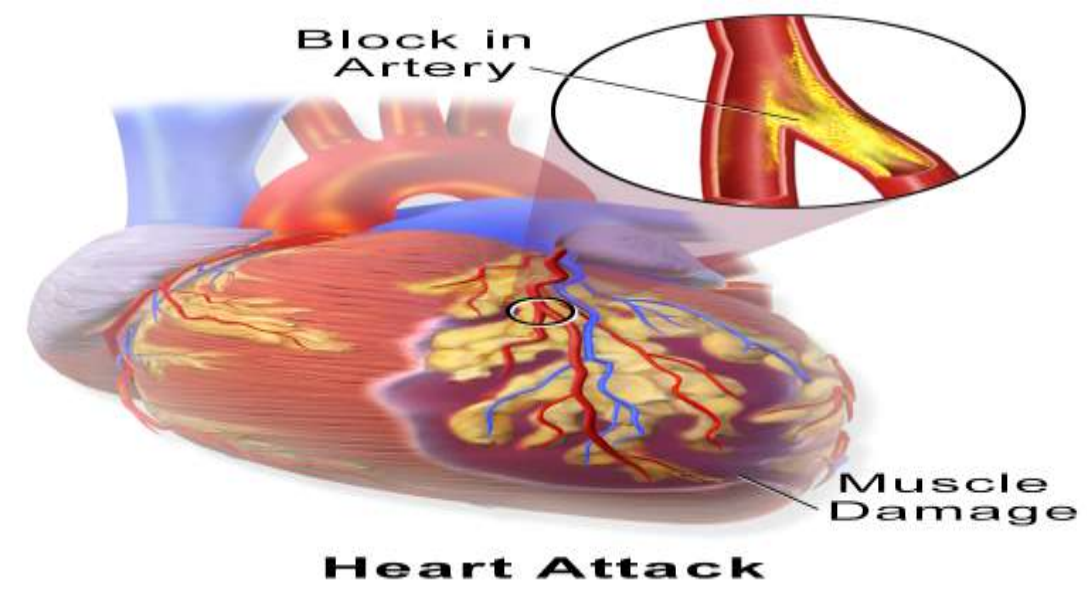
CARDIOVASCULAR SYSTEM

BIOCHEMICAL MARKERS FOR MI



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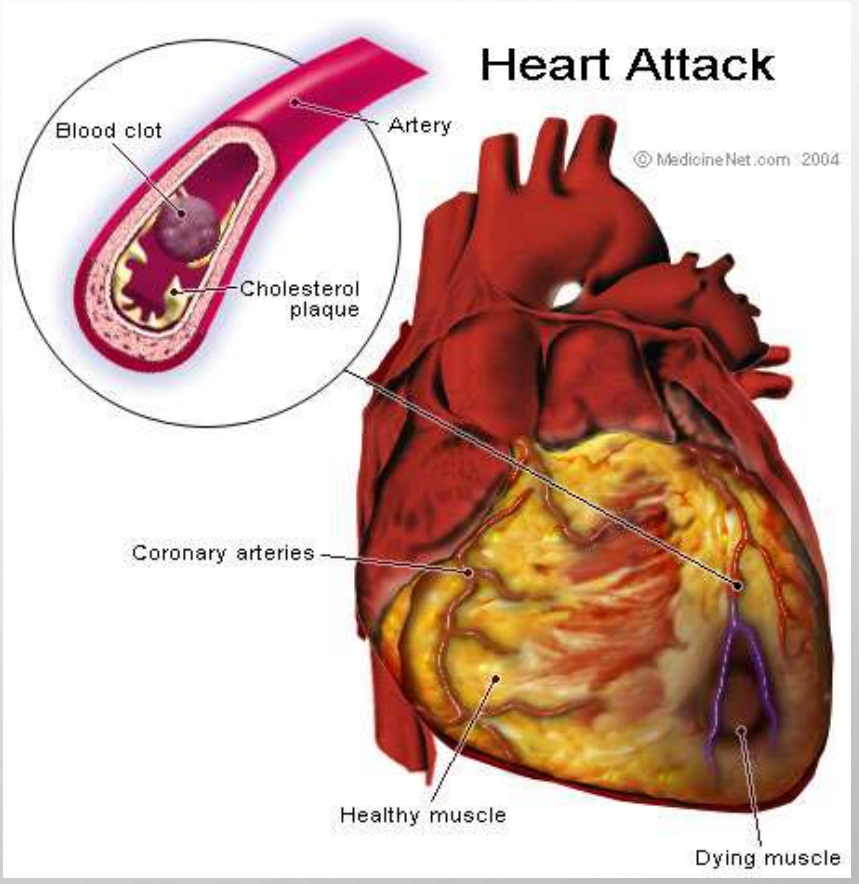
Acute Myocardial Infarction

- An imbalance between the supply of oxygen and the myocardial demand resulting in myocardial ischemia.

- A rapid development of myocardial necrosis caused by prolonged ischemia resulting in an irreversible myocardial injury.
- The development of infarction or ischemia will depend on the degree of occlusion or the presence of collateral blood flow.



Biochemical Changes

ischemia to myocardial muscles (with low O_2 supply)

↓

anaerobic glycolysis

↓

increased accumulation of Lactate

↓

decrease in pH

↓

activate lysosomal enzymes

↓

disintegration of myocardial proteins

↓

cell death & necrosis

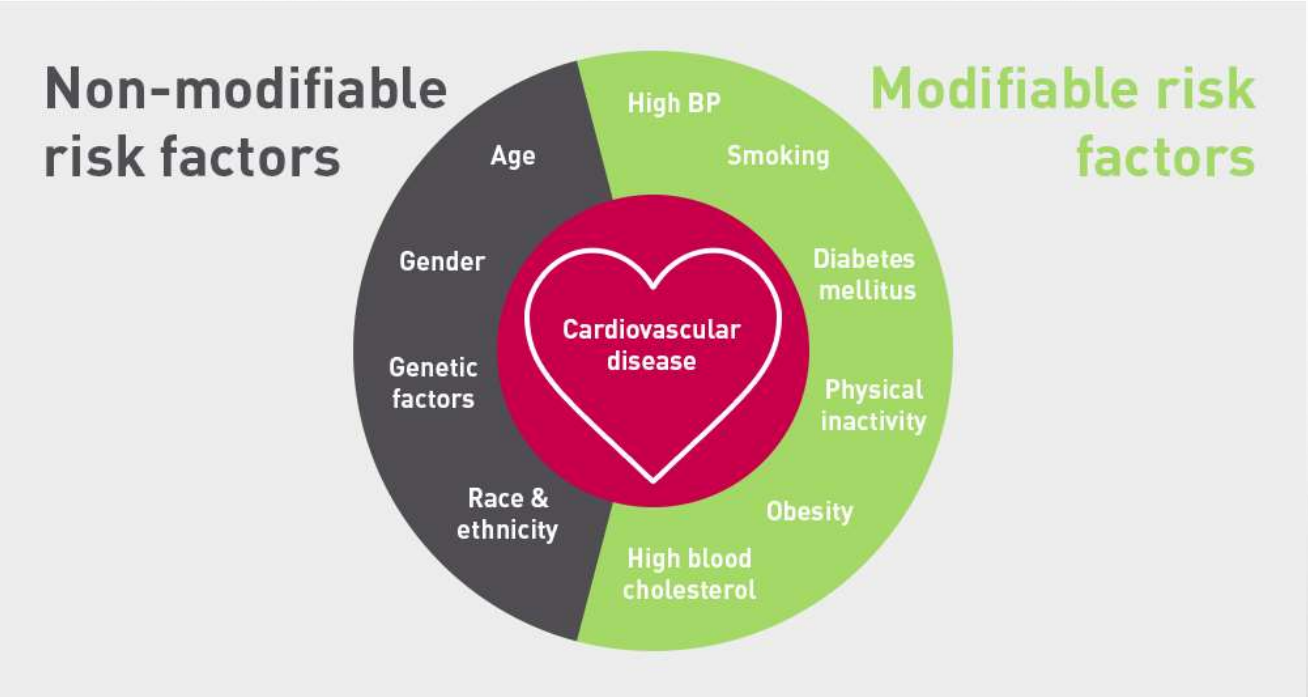
clinical manifestations (chest pain)

BIOCHEMICAL MARKERS

release of intracellular contents to blood

ECG changes

RISK FACTORS



- LDL-C is most important **atherogenic** particle.
- Apo B: Only apoprotein on LDL. ApoA1 is often used as a biomarker for prediction of CVD.
- ApoB100 / ApoA1 ratio is more effective at predicting heart attack risk, in patients who had had an acute MI, than either the ApoB100 or ApoA1 measure alone.

MYOCARDIAL INFARCTION

- Many patients with myocardial infarction have a typical history of crushing **central chest pain**, perhaps radiating to the arm or jaw, associated with typical ECG changes.
- myocardial infarction can, however, present **atypically**, or even be clinically silent, particularly in the elderly.
- The clinical evaluation often is limited by atypical symptoms, in most patients the initial ECG is non-diagnostic.
- The role of cardiac markers in the diagnosis and treatment of patients with chest pain and suspected AMI has evolved considerably.

WHAT ARE THE INVESTIGATIONS?

- ECG. - chest x-ray - coronary angiogram - Lipid profile
- serum cardiac enzymes & proteins.

WHO Diagnosis of Acute Myocardial Infarction (AMI)

Presence of two of the three criteria:

1. ***History*** of characteristic ***chest pain.***
2. ***Electrocardiographic changes.***
3. Typical ***pattern of serum cardiac enzyme & proteins*** rise, peak and return to reference range.

- **However, in 1999, European Society of Cardiology and the American College of Cardiology**
  - **Sensitive biomarkers** for the diagnosis of AMI
  - **Cardiac troponins (cTn) is the gold standard.**

IDEAL CARDIAC MARKER CHARACTERISTICS

- **Cardiac specific**. specific to myocardial muscle cells (no false positive).
- **Sensitive:** can detect minor damage. no miss of positive cases (no false negative)
- **Prognostic**: relation between plasma level & extent of damage
- Rises **soon** after plaque rupture.
- Elevated over a sustained period of time.
- **Easy to measure**, fast assay.
- Diagnostic utility verified by clinical studies.

QUESTIONS ANSWERED BY MARKERS OF CARDIAC DAMAGE

- **RULE IN/OUT AN ACUTE MI**
- **CONFIRM AN OLD MI (SEVERAL DAYS)**
- **MONITOR RE-INFARCTION**
- **MONITOR THE SUCCESS OF THROMBOLYSIS**

Biochemical markers in myocardial ischemia /necrosis

1. Cardiac Enzymes (isoenzymes):

- Total CK, CK-MB activity, CK-MB mass
- Aspartate aminotransferase (AST), Lactate dehydrogenase (LDH),.
- Glycogen phosphorylase BB (GPBB).

2. Cardiac proteins:

- Myoglobin & Troponins
- Ischemia Modified Albumin
- Heart-Fatty Acid binding protein (H-FABP).

3. Micro RNA (miRNA)

BIOCHEMICAL MARKERS IN MYOCARDIAL ISCHAEMIA /NECROSIS

OBSOLETE

- ASPARTATE AMINOTRANSFERASE -TOTAL CK - LACTATE DEHYDROGENASE

ESTABLISHED

- TROPONIN T - TROPONIN I - CK/MB - MYOGLOBIN

EMERGING

- MICRO RNA (MIRNA)

- HEART FATTY ACID-BINDING PROTEIN (H-FABP)

- ISCHEMIA-MODIFIED ALBUMIN

- GLYCOGEN PHOSPHORYLASE BB (GPBB)

- COPEPTIN - B-TYPE NATRIURETIC PEPTIDE

- GROWTH DIFFERENTIATION FACTOR 15 - PREGNANCY-ASSOCIATED PLASMA PROTEIN A

LABORATORY INVESTIGATIONS

SPECIMEN COLLECTION:

- ***SERUM*** IS THE SPECIMEN OF CHOICE
- ***HEPARINIZED PLASMA*** IS ACCEPTABLE
- ***VENOUS WHOLE BLOOD*** FOR RAPID CARDIAC TROPONIN T.
- ***SALIVA***

COLLECTION TIME:

- ***SERIAL SPECIMENS*** COLLECTED AT APPROPRIATE TIME INTERVALS.
- SERIAL MEASUREMENTS ARE MOST USEFUL
- SAMPLES ARE DRAWN **ON ADMISSION**

AT 2-4 HOURS

AT 6-8 HOURS

AT 12 HOURS

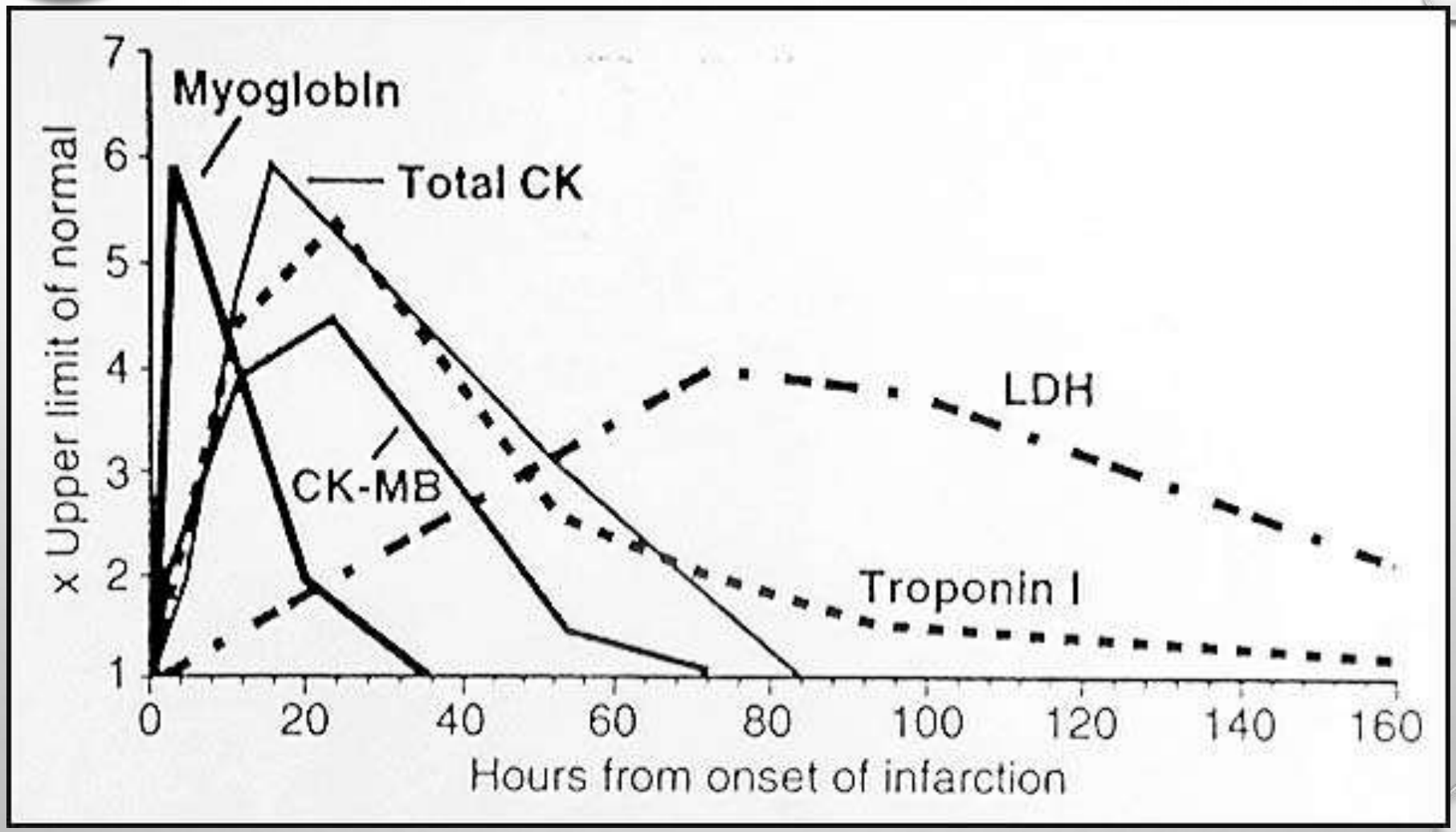
MYOGLOBIN

- O2-binding protein (heme-containing protein).
- Released from skeletal and heart muscle when damaged.
- Rapidly cleared by kidneys (not **long term** marker).
- Its level varies with gender, age, physical activity.
- More sensitive than CK, CK-MB activities.
- myoglobin is not cardiac specific, better used in conjunction with other markers. Increased in patients with skeletal muscle disease and chronic renal failure

MYOGLOBIN

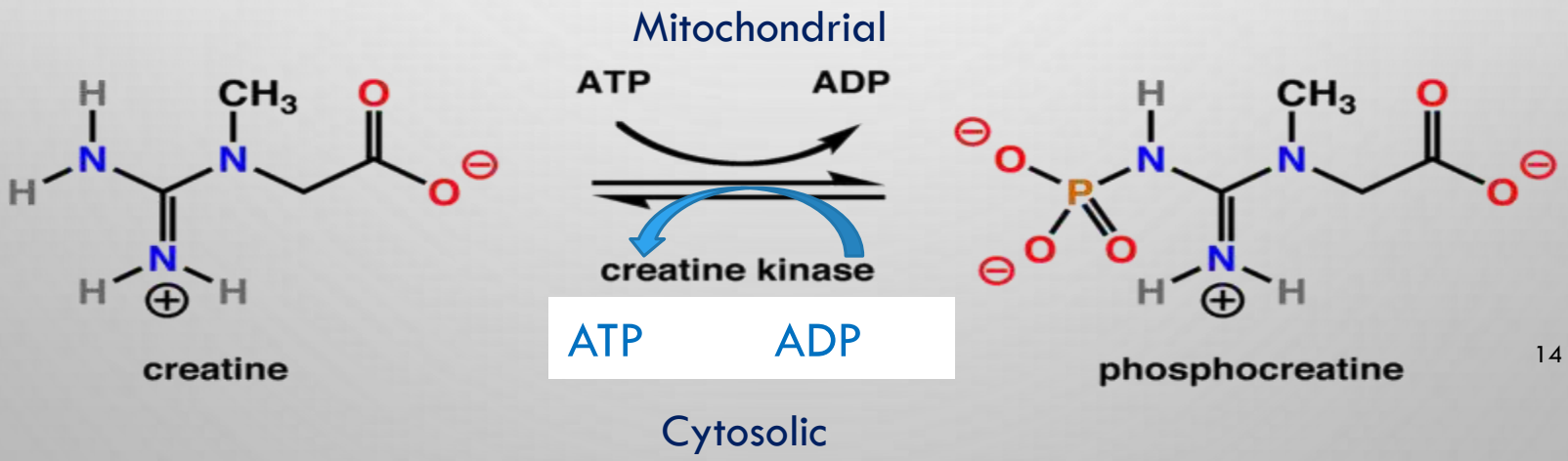
- IT **STARTS** TO RISE WITHIN **1-4 H**
- **DETECTED** BETWEEN **6-9 H** IN NEARLY **ALL** AMI PATIENTS FROM CHEST PAIN.
- **RETURNS** TO BASE LINE LEVELS WITHIN **18- 24 H.**
- IF MYOGLOBIN ARE NORMAL **8H** AFTER PAIN ……. AMI CAN BE **RULES OUT**.
- [CK-MB IS PREFERRED THAN MYOGLOBIN IN PATIENTS WHO ARE ADMITTED LATER THAN **10-12 H** AFTER PAIN].

Biochemical markers of MI



CREATINE KINASE (CK)

- **Creatine kinase acts as a regulator of high-energy phosphate production and utilization within contractile tissues.**
- **Cytoplasmic CK is a dimer, composed of M and/or B subunits, which associate forming CK-MM, CK-MB and CK-BB isoenzymes**
- **CK catalyses the conversion of creatine and consumes ATP to create phosphocreatine (PCr) and ADP.**
- **This CK enzyme reaction is reversible, such that also ATP can be generated from PCr and ADP.**



CREATINE KINASE (CK)

- **CK-MM is the main isoenzyme found in skeletal >> Cardiac muscles.**
- **CK-MB is found mainly in cardiac muscle Trace amounts of CK-MB are found in skeletal muscle.**
- **CK-BB is the predominant isoenzyme found in brain, colon, ileum, stomach and urinary bladder.**

CK- TOTAL

- A RAISED PLASMA ***TOTAL CK ACTIVITY***, DUE TO ENTIRELY **CK-MM** MAY FOLLOW:
  - SKELETAL MUSCLE DISEASE.
  - RECENT INTRAMUSCULAR INJECTION
  - EXERCISE
  - SURGERY.
- **(NON SPECIFIC)**
- LIMITED PROGNOSTIC VALUE.

CK-MB

- **High specificity. more specific than total CK BUT: less specific than troponin I.**
- **Gold standard as cardiac marker (was).**
- **It takes at least 4-6 h to increase.**
- **Peak levels at 12-24 h.**
- **Return 2-3 days.**
- **useful for early diagnosis of MI**
- **useful for diagnosis re-infarction**

- # CK-MB (MASS)

MASS ESTIMATION BETTER THAN ACTIVITY.

TO INCREASE SPECIFICITY, RATIO (RELATIVE INDEX)

***RELATIVE INDEX* = CK-MB MASS / CK ACTIVITY.**

CK-MB (MASS)

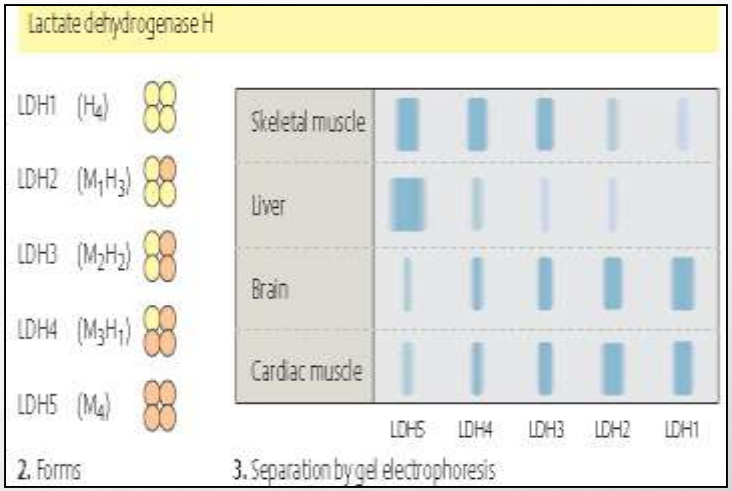
- If ratio >>> 3 indicative of AMI rather than skeletal muscle damage.
- CK/MB isoenzyme is not myocardium-specific occurring for instance in a small amount in skeletal muscle.
- Its use in the diagnosis AMI is considered acceptable only in cases where cTn assays are unavailable.
- The one advantage of CK-MB over the troponins is the early clearance that helps in the detection of re-infarction.

ASPARTATE TRANSAMINASE (AST)

- **HEPATIC CONGESTION** DUE TO RIGHT-SIDED HEART DYSFUNCTION MAY CONTRIBUTE TO THE ***RISE OF PLASMA AST*** ACTIVITY. **A NON-SPECIFIC** MARKER OF MI
- IF THERE IS **PRIMARY HEPATIC DYSFUNCTION**, PLASMA ***AST*** RISES WHEREAS ***LDH1*** ACTIVITY USUALLY REMAINS NORMAL.
- THE SEQUENCE OF CHANGES IN **PLASMA AST ACTIVITY IN MI** IS ***SIMILAR*** TO THOSE OF ***CK*** .
- **AST** AND **LDH** MEASUREMENTS ***ARE RARELY OF PRACTICAL VALUE*** IN THE MANAGEMENT OF PATIENTS WITH SUSPECTED MYOCARDIAL INFARCTION.
- ***EXCEPTIONALLY,*** WHEN A PATIENT WITH CHEST PAIN PRESENTS ***LATE***, MEASUREMENT OF **LDH** MAY BE HELPFUL AS THIS ENZYME **REMAINS ELEVATED IN THE PLASMA FOR SEVERAL DAYS** FOLLOWING MYOCARDIAL INFARCTION.

Lactate dehydrogenase (LDH)

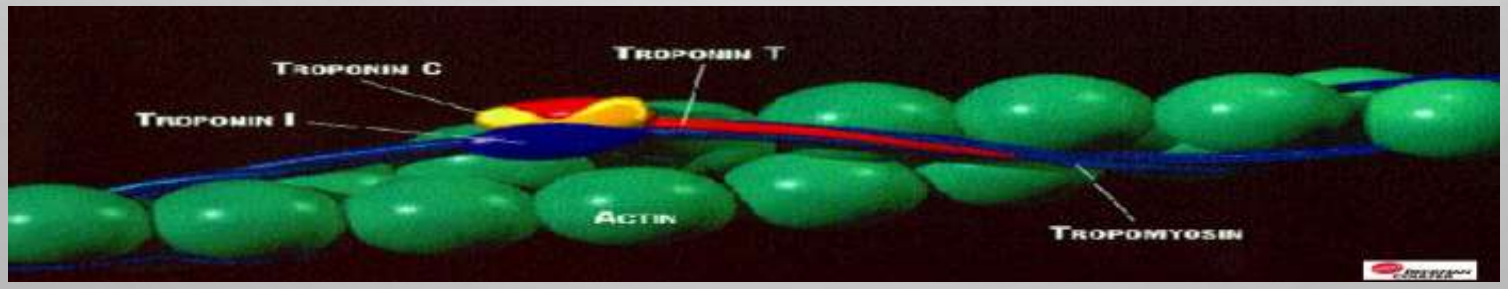
- LDH is a **tetramer**, each chain may be one of two types (H,M) where LDH1 is (H4) while LD5 is (M4)
- **LD1 & LD2** predominates in **heart**
- LDH increases **later** than CK-MB and Ck
- Reaches a **max. level** in **48 h**
- Remains elevated for **5-6 days** after the MI



- A **non-specific marker** of tissue injury: High levels are found in *liver, lung, kidney* and other diseases.
- Myocardial infarction resulting in insufficient oxygen delivery to that portion of cardiac muscle. This causes the affected muscle to rely on **anaerobic metabolism** for its energy supply with concomitant production of lactic acid.

TROPONIN

- TROPONIN IS A **PROTEIN.**
- PRESENT IN HIGH CONCENTRATION IN **MUSCLE & HEART.**
- REGULATES THE FORCE OF **MUSCULAR CONTRACTIONS**
- IS COMPOSED OF **3 SUB UNITS I, T AND C.**
- **TROPONIN C:** CA^{++} BINDING. (***NOT HEART-SPECIFIC).***
- THE **TROPONIN I AND TROPONIN T** FOUND IN HEART MUSCLE IS SIGNIFICANTLY **DIFFERENT** FROM TROPONINS FOUND IN NON-CARDIAC MUSCLE



TROPONIN T

- TROPOMYOSIN BINDING ELEMENT .
- ITS LEVEL INCREASES **WITHIN** **6 HRS** OF MI.
- **PEAKS** AT **72 HRS .**
- **REMAINS** ELEVATED **7-10 DAYS.**
- TROPONIN T MAY BE **ELEVATED** IN PATIENTS WITH ***CHRONIC RENAL FAILURE*** AND THUS MAY ***NOT BE SO CARDIAC-SPECIFIC***

TROPONIN I:

- IT IS RELEASED **WITHIN** **4-6 HRS** OF THE ONSET OF MI.
- **PEAKS** **14-24HRS.**
- **REMAINS** ELEVATED FOR **3-5 DAYS.**
- **DISAPPEARS** FROM BLOOD **AFTER ABOUT ONE WEEK**. SO, USEFUL FOR DIAGNOSIS OF DELAYED ADMISSION CASES.
- CARDIAC TROPONINS HAVE BEEN RECOMMENDED AS THE ***BIOCHEMICAL CARDIAC MARKER OF CHOICE***.

CARDIAC TROPONIN: TROPONIN I (CTN I)

- SERUM TROPONINS ARE ***NOT FOUND IN HEALTHY INDIVIDUALS*** (UNLIKE CK/MB).
- TROPONINS ARE BOTH **MORE SENSITIVE** (DIAGNOSE MINOR INFARCTION) AND **MORE SPECIFIC** THAN CK-MB IN TERMS OF ITS DIAGNOSTIC ABILITY WITH RESPECT TO MYOCARDIAL DAMAGE.
- **PROGNOSTIC MARKER** (RELATION BETWEEN LEVEL IN BLOOD & EXTENT OF CARDIAC DAMAGE). DETERMINATION OF SIZE OF INFARCT.
- DETERMINATION OF **SUCCESS OF REPERFUSION.**
- **TWO NEGATIVE TROPONINS** 6 HOURS APART ARE GOOD (BUT NOT ABSOLUTE) EVIDENCE OF NO RECENT AMI.
- **POSITVE TROPONIN** IN PATIENTS **WITHOUT ECG** CHANGES & WITH **NORMAL CK-MB** LEVELS MAY IDENTIFY PATIENTS **AT INCREASED RISK OF CARDIAC EVENTS**

HEART-TYPE FATTY ACID-BINDING PROTEIN (H-FABP)

- H-FABP IS A SMALL CYTOSOLIC PROTEIN FOUND IN THE CARDIAC TISSUES.
- IT IS CHIEFLY PRESENT IN THE **MYOCARDIUM** AND, TO A LESSER EXTENT, IN THE BRAIN, KIDNEY AND SKELETAL MUSCLE.
- RESPONSIBLE FOR THE **TRANSPORT OF FATTY ACIDS** FROM THE PLASMA MEMBRANE TO:

➢ SITES OF B-OXIDATION IN **MITOCHONDRIA** AND **PEROXISOMES**.

➢ **ENDOPLASMIC RETICULUM** FOR LIPID SYNTHESIS.

- H-FABP IS RELEASED **EXTREMELY EARLY** INTO THE SERUM FOLLOWING MYOCYTE RUPTURE.

➢ ⇇⇇⇇ AS EARLY AS **30 MIN** AFTER MYOCARDIAL INJURY

➢ **PEAKS** AT **6–8 H** AND

➢ **RETURNS** TO BASELINE LEVELS AT **~24 H.**

- IT COULD BE USED TO QUICKLY **RULE OUT** AMI.

ISOENZYME BB GLYCOGEN PHOSPHORYLASE (GPBB)

- IT IS ONE OF THE 3 ISOFORMS OF GLYCOGEN PHOSPHORYLASE.
- GPBB EXISTS IN **CARDIAC** AND **BRAIN TISSUE**. BECAUSE OF THE BLOOD–BRAIN BARRIER, GP-BB CAN BE SEEN AS BEING **SPECIFIC TO HEART MUSCLE.**
- ⇇⇇⇇ BLOOD LEVELS CAN BE SEEN IN **ISCHEMIA, MI** AND **UNSTABLE ANGINA**.
- **EARLY BIOCHEMICAL MARKER** OF MYOCARDIAL NECROSIS.
- ⇇⇇⇇ WITHIN THE **FIRST HOUR** OF **MI**.
- ⇇⇇⇇ **1–3 HOURS** AFTER **ISCHEMIA**.
- **VERY SENSITIVE** INDICATOR OF **MI** WITH A SENSITIVITY SUPERIOR TO THAT OF MYOGLOBIN, CK-MB MASS, AND CTNT.

COPEPTIN

- COPEPTIN, THE C-TERMINAL PORTION OF PROVASOPRESSIN IS COSECRETED WITH VASOPRESSIN.

- ⇇⇇⇇ **WITHIN MINUTES** IN PATIENTS WITH **AMI**.

- ADDING **COPEPTIN** + **CTNI** CAN **RULE OUT OF AMI.**

ISCHEMIA-MODIFIED ALBUMIN (IMA)

- IT IS RAISED IN THE PRESENCE OF ***MYOCARDIAL ISCHEMIA.***
- NORMAL ALBUMIN CAN BIND METALS AT ITS N TERMINUS.
- DURING ISCHEMIA, ***FREE RADICALS, ALTER THE BINDING SITE, DECREASING BINDING ABILITY*** MAKE IT MORE RESISTANT TO BIND METALS.
- ***POSITIVE TEST*** – ISCHEMIA
- ***NEGATIVE TEST*** (TOGETHER WITH NEGATIVE TROPONIN AND NEGATIVE ECG) HAS A 99% **NEGATIVE PREDICTIVE VALUE** FOR MI.
- ***RAPIDLY CLEARED***
- ***NOT SPECIFIC*** FOR CARDIAC ISCHEMIA.
- IT IS A MARKER ***SENSITIVE FOR ISCHEMIA*** RATHER THAN NECROSIS.

➢ IT IS DETECTED WITHIN A ***FEW MINUTES.***

➢ PEAKS AT ***2-4 HOURS***.

➢ DISAPPEARS WITHIN ***6 HOURS.***

MICRO-RNAS

MI, AS A **DAMAGE** AND **CELL DEATH** PROCESS

↓

AFFECTS A NUMBER OF **GENETIC PROCESSES** THAT AIM **REPAIR** AND **SURVIVAL** OF THE CARDIOMYOCYTE.

↓

CHANGES IN CIRCULATING LEVELS OF **MIRNA**

(IN THE FIRST FEW HOURS AFTER MI)

MICRO RNA (MIRNA)

- MICRORNAS (MIRNAS) CIRCULATE IN THE BLOODSTREAM IN A REMARKABLY STABLE FORM.
- BECAUSE OF THEIR ***STABILITY*** AND OFTEN ***TISSUE- AND DISEASE-SPECIFIC*** EXPRESSION AND THE POSSIBILITY TO MEASURE THEM WITH ***HIGH SENSITIVITY*** AND ***SPECIFICITY***, MIRNAS ARE EMERGING AS **NEW DIAGNOSTIC & PROGNOSTIC BIOMARKERS.**

DIAGNOSTIC ABILITIES OF CIRCULATING MIRNAS FOR MI.

- IT HAS BEEN FOUND THAT **MIR-1, MIR-133**, AND **MIR-499** WERE **ELEVATED** IN PATIENTS WITH MI.
- THE SLOW TIME COURSE OF **MIR-499** MIGHT LEAD TO INCREASED DIAGNOSTIC PERFORMANCE AT **LATE TIME POINTS** AFTER MI WHEN **CTNI** HAS ALREADY RETURNED BACK TO NORMAL LEVELS.

MICRO RNA (MIRNA)

DIAGNOSTIC ABILITIES OF CIRCULATING MIRNAS FOR MI.

- THE **CARDIAC-SPECIFIC MIR-208** WAS NOT DETECTABLE IN PLASMA OF HEALTHY CONTROLS OR IN PATIENTS WITH STABLE CAD.
- **WITHIN 4 H** AFTER THE ONSET OF SYMPTOMS, MIR-208 WAS DETECTED IN **ALL** PATIENTS, WHEREAS CTNI WAS ONLY DETECTED IN 85% OF THE PATIENTS, CONFIRMING THE ***SUPERIOR SENSITIVITY*** OF **MIR-208** ***AT EARLY TIME POINTS***.

DIAGNOSTIC ABILITIES OF CIRCULATING MIRNAS FOR MI.

- **MIR-122** AND **MIR-375** EXPERIENCED A **DROP** IN THEIR PLASMA LEVELS FOLLOWING MI.

……..

- IT MAY BE EXPECTED THAT **IN THE FUTURE**, A PANEL OF **MIRNAS**, PROBABLY IN COMBINATION WITH **CTNI**, HAS A BETTER POTENTIAL TO OFFER SENSITIVE AND SPECIFIC DIAGNOSTIC TESTS FOR **AMI.**

SALIVARY BIOMARKERS ASSOCIATED WITH MI

- **SALIVA** OFFERS AN EASY, SIMPLE AND NON-INVASIVE PROCEDURE.
- WHOLE SALIVA **CONTAINS** CONSTITUENTS FROM **SERUM**, **GINGIVAL FLUID** AND **ORAL MUCOSAL TRANSUDATE.**

SALIVARY MARKERS OF ACUTE MYOCARDIAL INFARCTION:

- **MYELOPEROXIDASE (MPO), C-REACTIVE PROTEIN (CRP), MYOGLOBIN, CK-MB AND CTN.**
- SALIVA CAN BE USED AS AN **ALTERNATIVE** TO SERUM IN THE DIAGNOSIS OF MI.

RECENTLY: USING **NANOCHIPS** AND A **SWAB OF THE CHEEK**,

CARDIAC BIOMARKER READINGS FROM SALIVA WITH **ECG READINGS**

DETERMINE **WITHIN MINUTES** WHETHER SOMEONE HAD A HEART ATTACK.

MY BEST WISHES