Basics of clinical enzymology

12. week

Factors affecting the amount of the plasma enzymes

Specific causes:

- different rate of enzyme diffusion from cells to serum
- different localization of individual enzymes in cells
- changes in the activity of enzymes after their release from the cells
- different rate of inactivation and elimination of enzymes from serum

Non-specific causes:

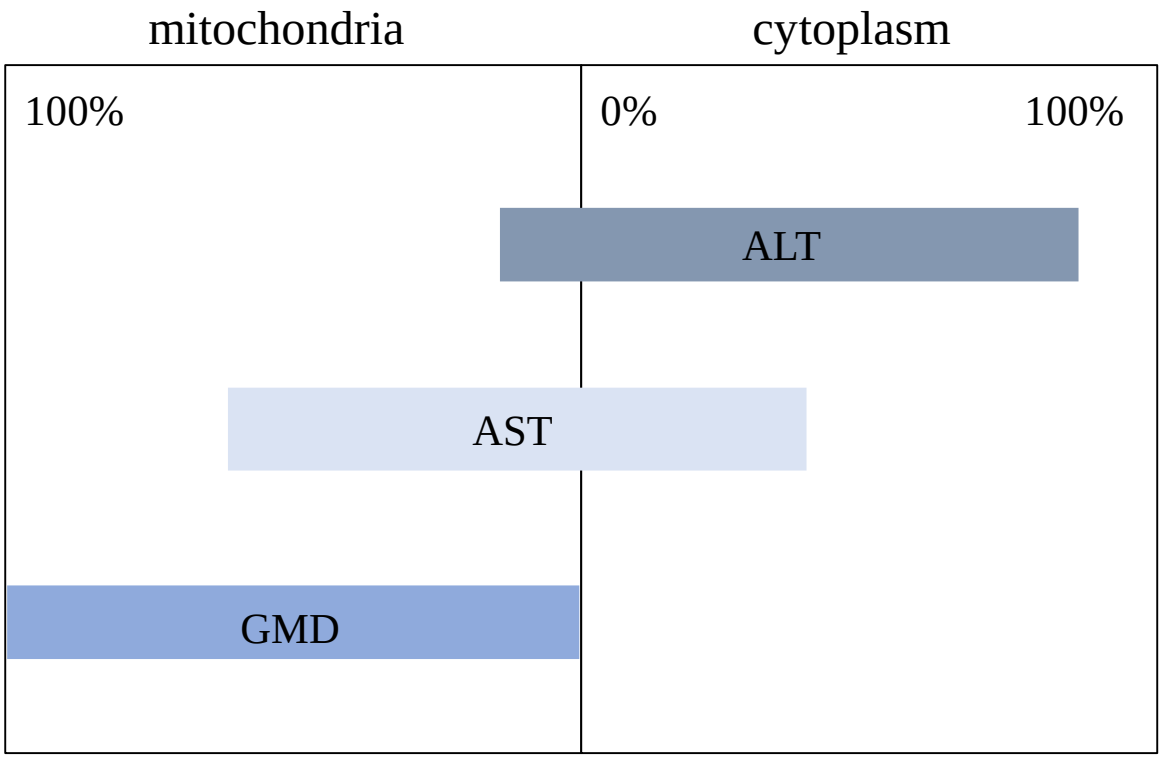
- changes in the content of enzymes in the organ as a result of the death of functional cells - especially with chronic damage
- changes in the spectrum of cellular enzymes during organ damage
- interference of enzymes from various organs in the bloodstream

Origin of enzymes in plasma

	Formation in	Changes in plasma during cell damage
Secretory enzymes: a) functional plasma enzymes • blood clotting enzymes • pseudocholinesterase	Liver	**decrease**
b) functional GIT enzymes • amylase, lipase	Pancreas	**increase**
Cellular enzymes: • transaminases, creatinkinase • aldolase, LDH • glutamatedehydrogenase • γ-glutamyltransferase	Various tissues	**increase**

Cellular enzymes

- different rate of enzyme diffusion from cells to serum
  - physical and chemical properties
  - binding to compartments



Elimination half-lives of enzymes in serum

Enzyme	Elimination half time
CK (CPK)	**15 hours**
AST (GOT)	**17 hours**
ALT (GPT)	**47 hours**
GMD (GLDH)	**18 hours**
LD_1 (LDH_1)	**113 hours**
LD_5 (LDH_5)	**10 hours**
HBD (HBDH)	**113 hours**
ALP (AF)	**3-7 days**
GMT (γ-GT)	**3-4 days**
CHS (CHE)	**10 days**
Amylase	**3-6 hours**
Lipase	**3-6 hours**

Enzyme profile of the organ

Crucial enzymes examined in organ damage:

Liver	ALT, AST, GMT, ALP, CHE
Myocardium	CK, CK-MB, LD, LD_1, AST
Skeletal muscle	CK, CK-MM, AST, aldolase
Skeleton	ALP, ACP
Pancreas	α-amylase, lipase
Prostate	ACP

Enzyme profile of the organ

Activity of enzymes in organs

Enzyme	Liver	Myocardium	Skeletal muscle	Erytrocytes
AST	980	870	600	13,3
ALT	580	48	57	1,7
LD	2 420	2 068	2 450	600
CK	12	5 830	330 840	under 0,1
aldolase	95	82	800	16

enzyme activity in nkat/g of fresh tissue at 25°C

Clinical forms of ischemic heart disease (IHD)

Chronic forms:

- asymptomatic IHD
- stable angina pectoris (exertional)
- Post-myocardial infarction condition

Acute forms:

- unstable angina pectoris
- myocardial infarction

Sudden cardiac death

Acute myocardial infarction (AMI)

5 - 20 minutes after the onset of ischemia: necrosis of the first cardiomyocytes

Current diagnostics:

- Clinical presentation:

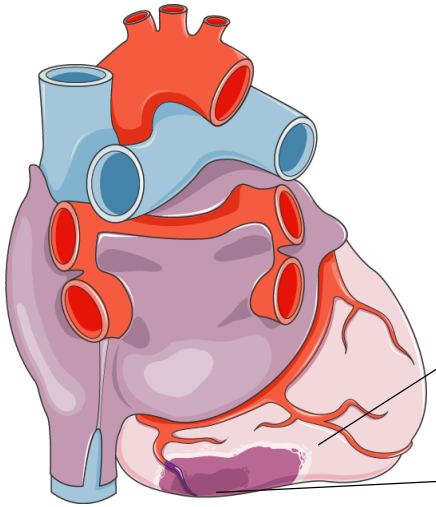
– chest pain, radiating pain to the left arm, lower jaw, nausea, shortness of breath ...

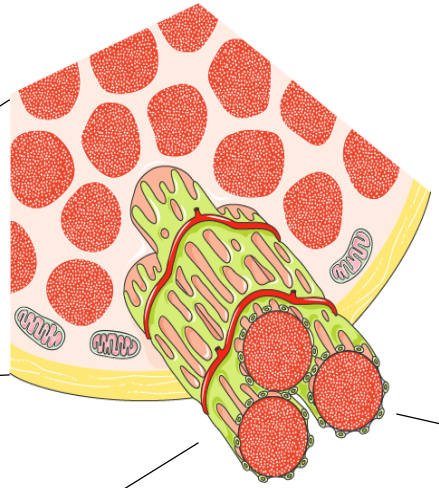
- ECG and imaging methods - echocardiography, MRI
- Cardiac troponins:

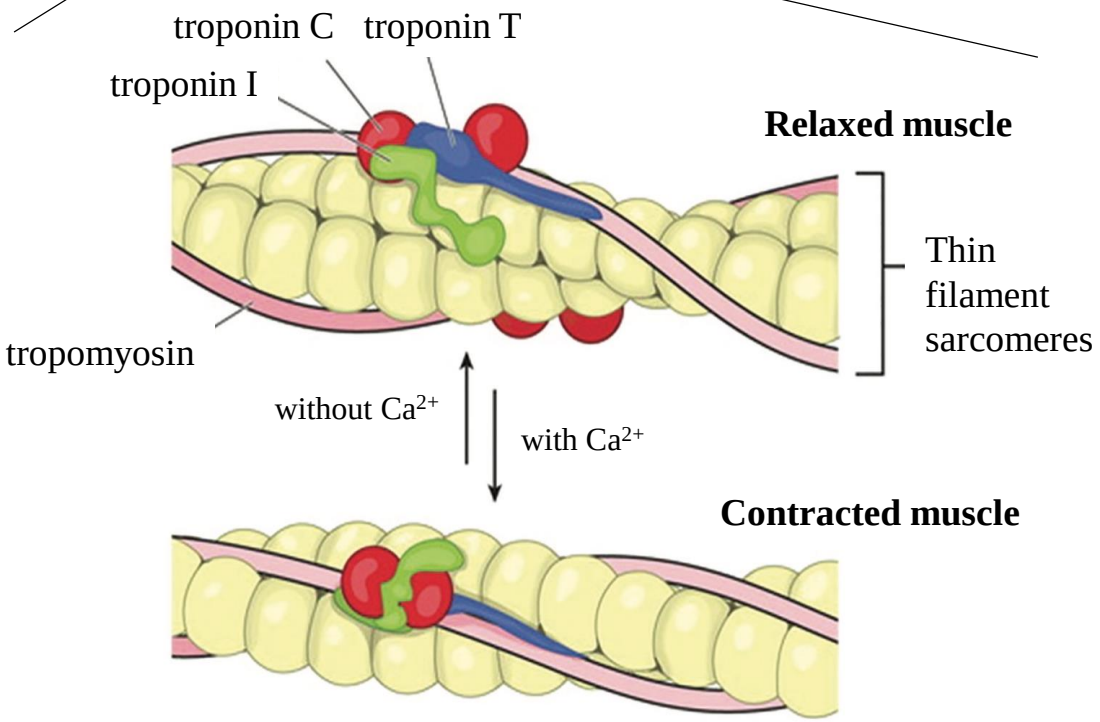
– types: cTnI, cTnT, TnC (same in the heart and skeletal muscle)

- acute damage causes increase of > 20%
- rise within 3 hours of onset of infarction
- peak up to 300 times the upper reference limit

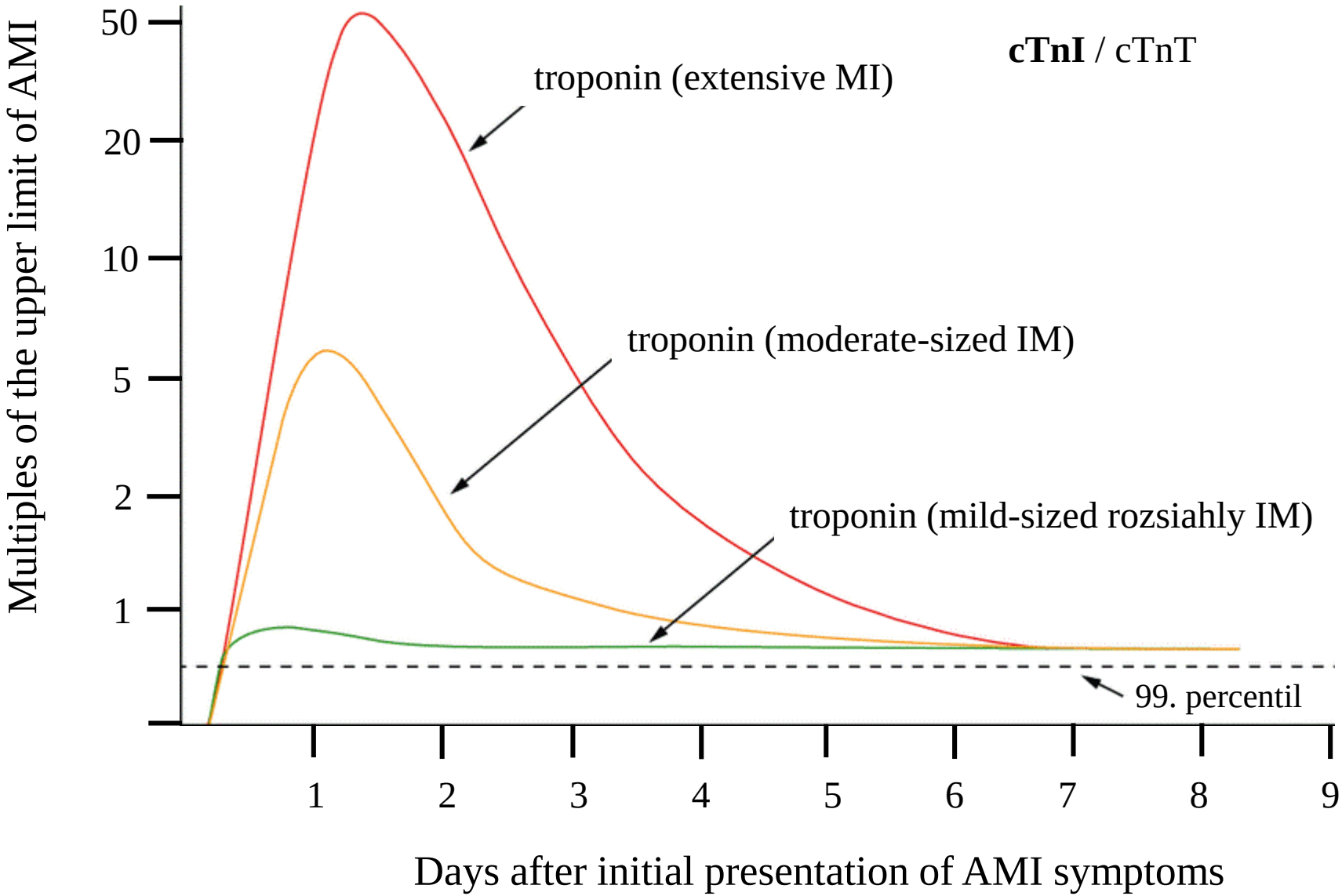
Troponins






troponin C
troponin T
troponin I
Relaxed muscle
Thin filament sarcomeres
tropomyosin
without Ca^{2+}
with Ca^{2+}
Contracted muscle

Myocardial infarction - troponins



Additional parameters in AMI

Myoglobin

- early marker of AMI (rise 2-6 hours)
- rapidly decreases, returns to normal within 24 hours (reinfarction)
- nonspecific (also present in skeletal muscle)
- strong negative predictive value

CK-MB mass

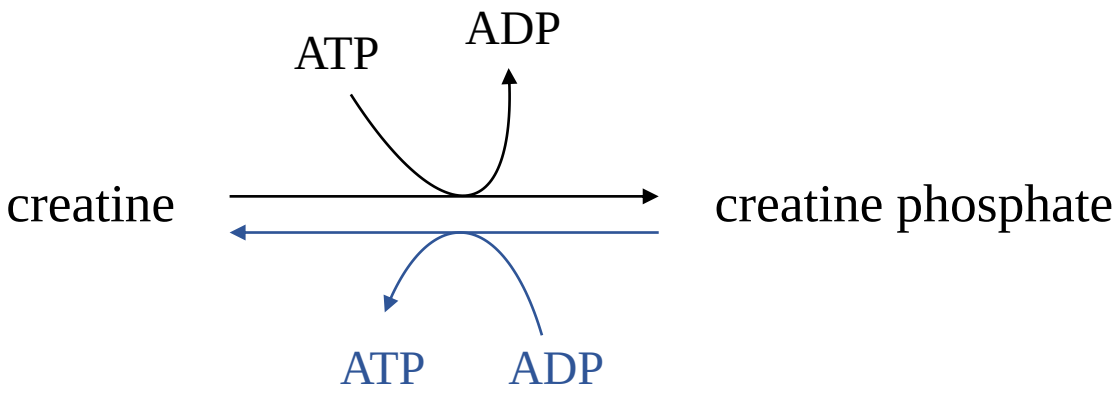
- mass concentration of cardiac isoform of creatine kinase (μg/l)
- alternative for diagnosing AMI if troponins cannot be measured (rarely needed)
- increases 3-10 hours after symptom onset
- nonspecific, can also originate from skeletal muscle

Additional parameters in AMI

Creatine kinase (CK)

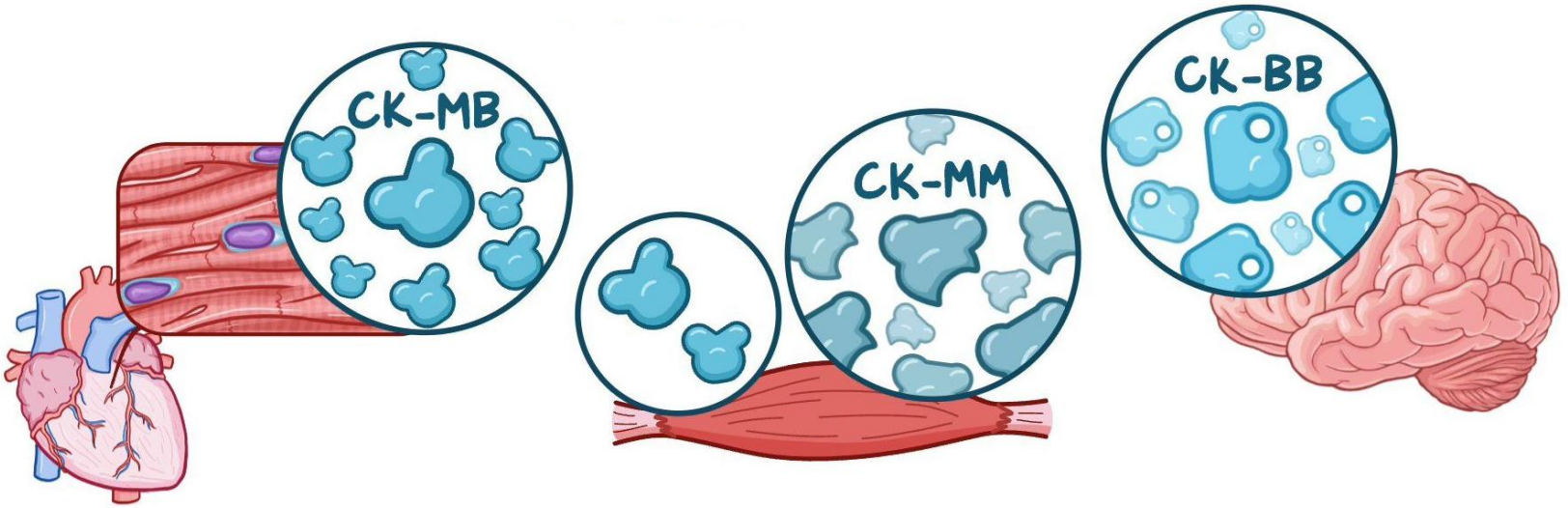
- primarily a cytoplasmic enzyme
- formed as a dimer of two subunits

  M (muscle) and B (brain) – 3 isoenzymes
- best marker for muscle damage
- elevated CK levels can occur with increased muscle exertion, skeletal muscle trauma, myopathies, as well as injections



Additional parameters in AMI

Creatine kinase (CK)

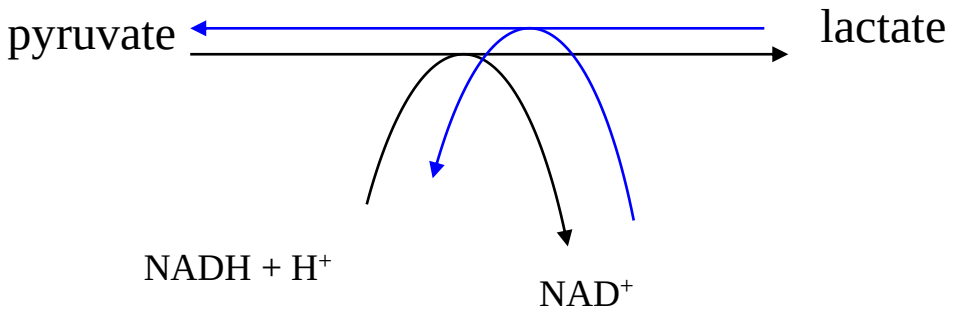


	CK-MM (CK_3)	**CK-MB (CK_2)**	**CK-BB (CK_1)**
origin	muscle	heart	brain
%	94 - 96	< 6	trace amount
increase	muscle exertion, surgeries injections	after 1 hour post-infarction	severe brain damage

Additional parameters in AMI

Lactate dehydrogenase (LD)

- cytoplasmic ubiquitous enzyme
- highest activities – skeletal muscle, myocardium, liver, ery
- interferences – hemolysis
- non-specific – isoenzymes (4 subunits of 2 types)



Additional parameters in AMI

Lactate dehydrogenase

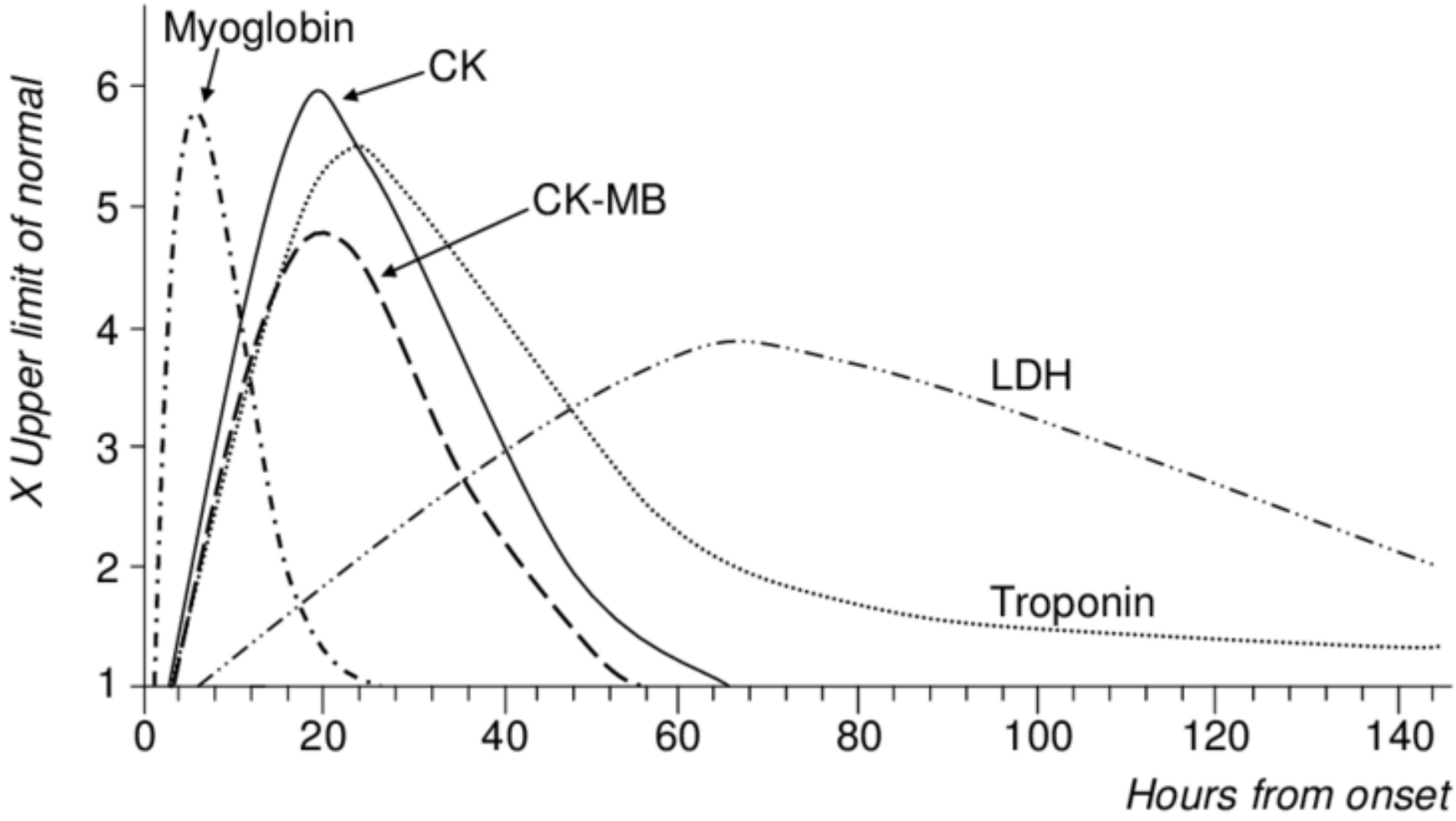
subunits

muscle

heart

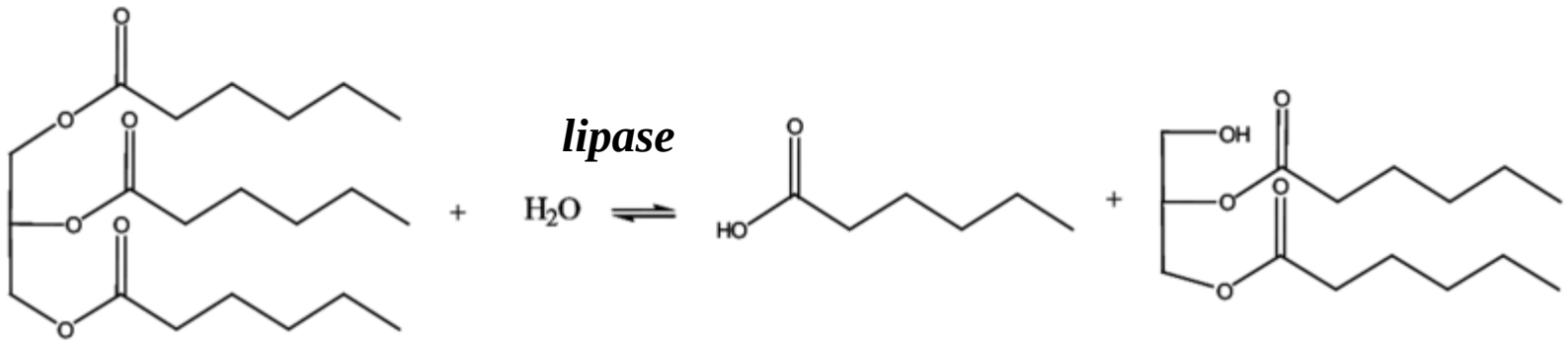
	LD_1	LD_2	LD_3	LD_4	LD_5
%	31	49	11	5	3,1
Origin	heart, ery, brain	mononuclear phagocytic system	lungs	kidney, placenta, pancreas	liver, skeletal muscle
Increase	myocardial infarction pernicious anemia hemolysis hemoblastosis carcinomas			liver metastases chronic/acute liver diseases	

AIM cardiac markers



Acute pancreatitis

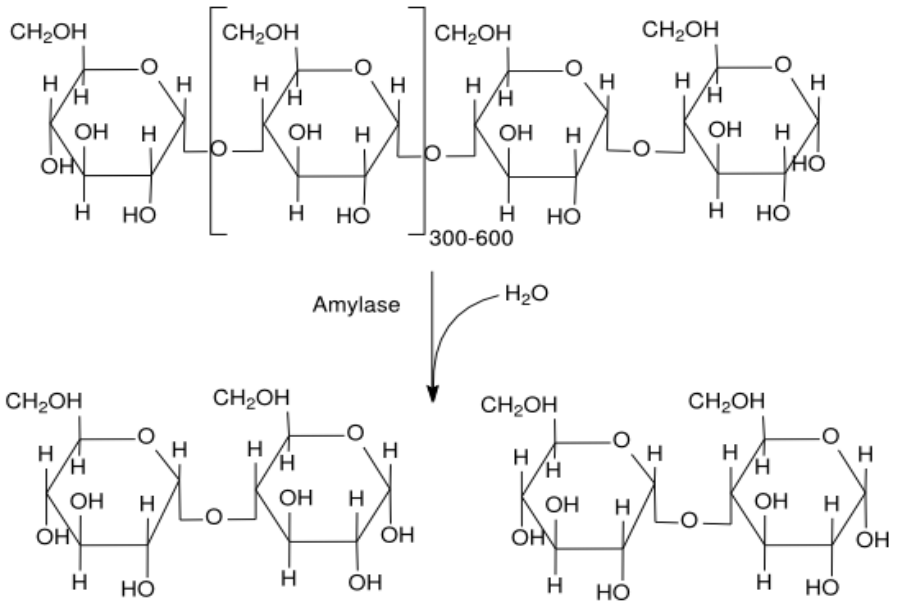
- basic test: serum ***lipase***
- transient increase in acute abdominal events
- produced 100 times more in the pancreas than in the liver, endothelium, and lipoproteins
- significant is a threefold increase from the upper reference value 3 to 6 hours after the onset of pain
- peaks at 24 hours - elevated for up to 2 weeks



triacylglycerol fatty acid diacylglycerol

Acute pancreatitis

- ***α-amylase***
- produced mainly in saliva and the pancreas
- increase similar to lipase, but decline is faster
  - half-life of 6-12 hours, returns to normal within 3 to 5 days
- it is also increased in cases of peptic ulcer rupture, ectopic pregnancy, ischemia of the bowel, appendicitis …



Chronic pancreatitis

Lipase

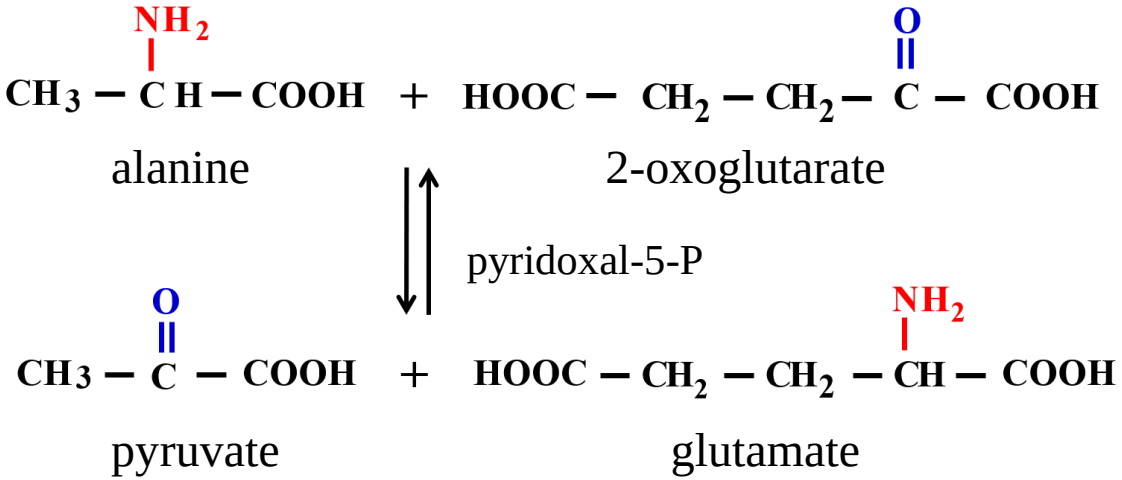
Elastase-1

- enzyme produced exclusively in the pancreas
- measured in stool
- falsely low values in about 10% of healthy individuals
- used for:
  - determining the etiology of diarrhea
  - marker in the therapy of chronic pancreatitis and cystic fibrosis

Liver - hepatocellular damage

Alanine aminotransferase (ALT) *(mainly a cytoplasmic enzyme)*

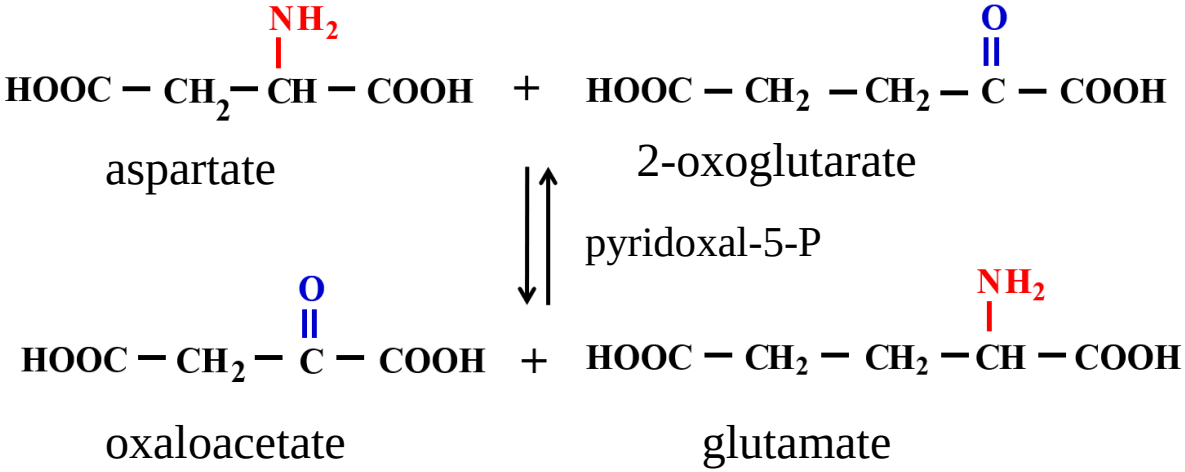
- increased even with minor damage
- most commonly elevated in both alcoholic and non-alcoholic liver steatosis
- most significant increase observed in acute viral hepatitis and toxic hepatitis



Liver - hepatocellular damage

Aspartate aminotransferase (AST) *(mainly a mitochondrial enzyme)*

- increased with severe liver damage (necrosis)
- highest activities found in the heart, skeletal muscle, and liver
- most significant increase observed in acute myocardial infarction, liver diseases, and skeletal muscle disorders
- interference: hemolysis



Hepatic protein synthesis function

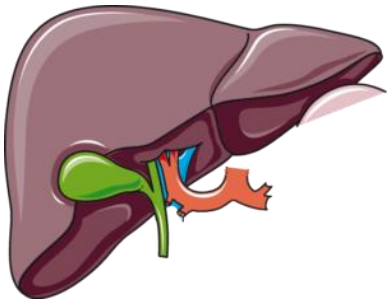
Cholinesterase (CHE) Secretory enzyme of the liver

- monitoring chronic liver diseases
- decreased activity - chronic liver diseases, prolonged fasting, malignant tumors, severe anemia, organophosphate poisoning
- increased activity - obesity, hyperlipoproteinemia, liver steatosis, nephrotic syndrome

Liver - cholestasis

Alkaline phosphatase (ALP):

- high activities in the bile ducts of the liver, bones, placenta and intestines
- physiological isolated rise: children, adolescents, pregnant, rises slightly even after eating



Gama-glutamyl transferase (GMT):

- highest activities: liver, pancreas, kidneys
- induced by drugs, alcohol
- used in hepatobiliary system diseases
  - in cholestasis, alcoholic hepatopathies

Enzymes in saliva

- ***α-amylase***
- ***salivary lipase***
- ***kallikrein***
- antibacterial enzymes:
  - *lysozyme*
  - *lactoperoxidase*
  - *lactoferin*

Gingivitis

- initial, mild stage of gum disease
- measurement of enzymes in gingival crevicular fluid (GCF)
- an increase in ALT, AST, ALP, ACP, GMT enzymes indicates cell damage and breakdown
- not common in practice:
  - complex sampling technique
  - reflects gum inflammation only at the specific sampling site
  - not suitable for mass screening

Bone

Alkaline phosphatase (ALP)

- high activities in bile ducts of the liver, bones, placenta, and intestines
- most significant increase - skeletal disorders (osteomalacia, bone tumors)
- decreased activity - bone growth disorders

Acid phosphatase (ACP)

- highest activities - prostate, liver, bones, erythrocytes, platelets
- most significant increase - prostate carcinoma, bone disorders, hemolytic conditions, thrombocythemia