

# HIGH-SENSITIVITY C-REACTIVE PROTEIN

## A NOVEL AND PROMISING MARKER OF CARDIOVASCULAR DISEASE

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### The need for additional cardiovascular risk factors

E Braunwald, N Engl J Med, 1997

Approximately 50% of patients who present with  
acute myocardial infarction or unstable angina pectoris  
do not have classical cardiovascular risk factors, such as:

- lipid levels (LDLC, TC:HDLC)
- hypertension
- diabetes
- smoking

## Emerging cardiovascular risk factors



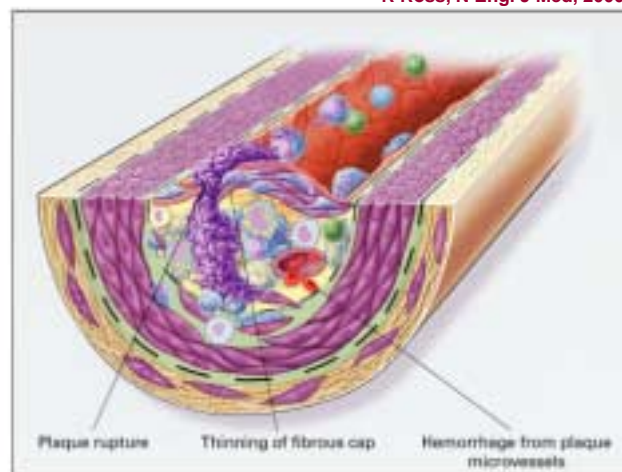
E Braunwald, N Engl J Med, 1997

- estrogen deficiency
- homocysteine
- plasma fibrinogen
- factor VII
- endogenous tissue plasminogen activator
- plasminogen-activator inhibitor type I
- D-Dimer
- lipoprotein(a)
- C-reactive protein
- *chlamydia pneumoniae*

## Atherosclerosis is an inflammatory disease



R Ross, N Engl J Med, 2000



Unstable fibrous plaques in atherosclerosis

## C-reactive protein as a marker of inflammation

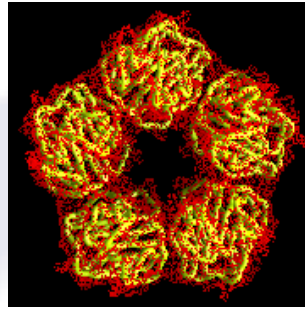


### acute inflammatory events:

increases by up to 1000-fold with infection, trauma & surgery

### chronic inflammatory disorders:

persistent increases with autoimmune diseases & malignancy



## Involvement of CRP in various processes of atherogenesis



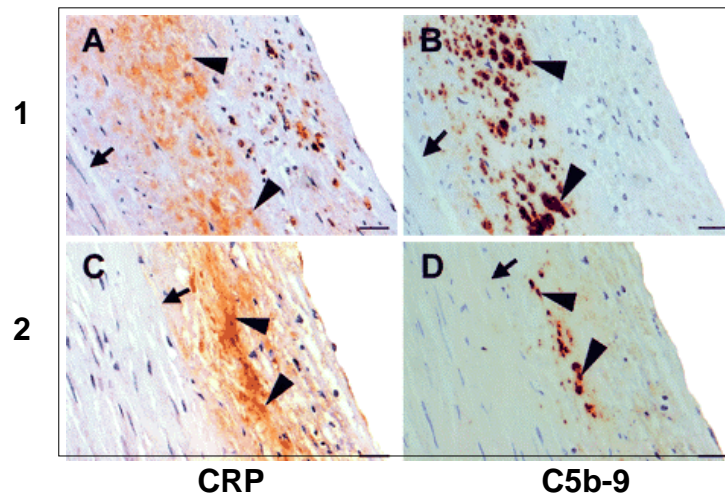
- **CRP is found in the vessel wall**  
(even in the very early stages of plaque formation)



## CRP localization in atherosclerotic lesions



J Torzewski, Arterioscler Thromb Vasc Biol, 1998



## Involvement of CRP in various processes of atherogenesis



- **CRP is found in the vessel wall**  
(even in the very early stages of plaque formation)
- **CRP may act as a procoagulant**  
(it induces the expression of tissue factor in monocytes)
- **CRP is chemotactic for monocytes and avidly binds to human neutrophils**
- **CRP induces complement activation, and may enhance tissue injury via this mechanism**
- **CRP is associated with impaired endothelial function**

■ **primary prevention**

( = prevention of a first event)

■ **secondary prevention**

(= prevention of recurrent events)

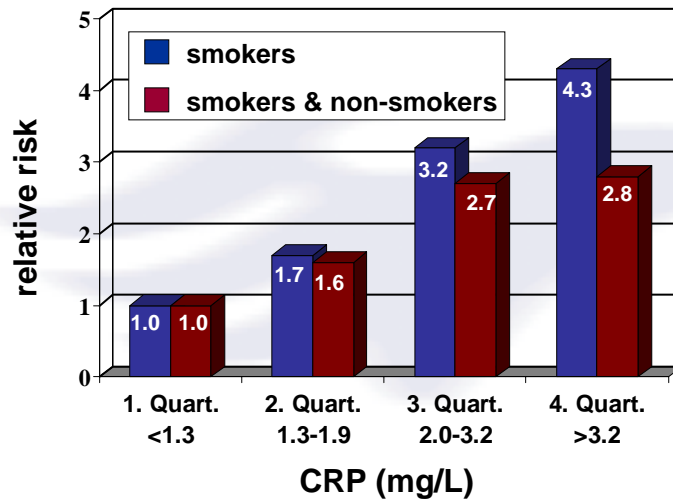
■ **primary prevention**

( = prevention of a first event)

### Relation of CRP & coronary heart disease (MRFIT study)



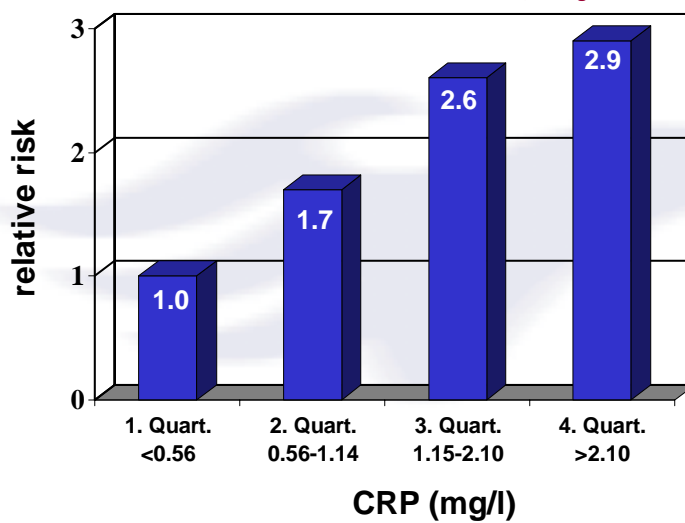
LH Kuller, Am J Epidemiol, 1996



### Physician's Health Study (PHS)



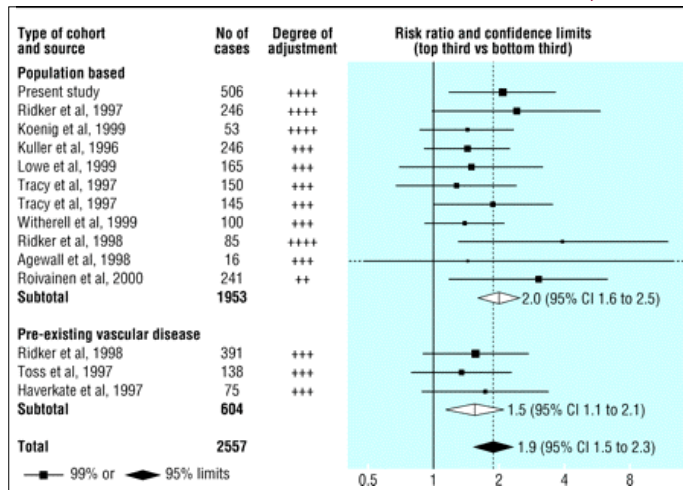
PM Ridker, N Engl J Med 1997



## Prospective studies of CRP & coronary heart disease



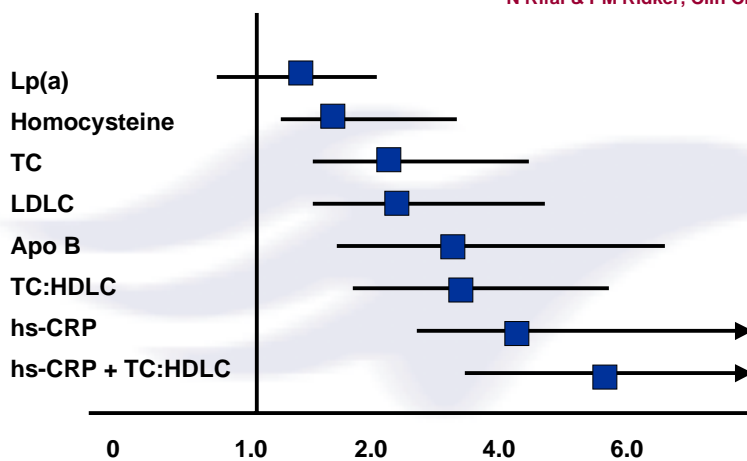
Danesh, BMJ 2000



## Relative risk of future cardiovascular events (Women Health Study-WHS)



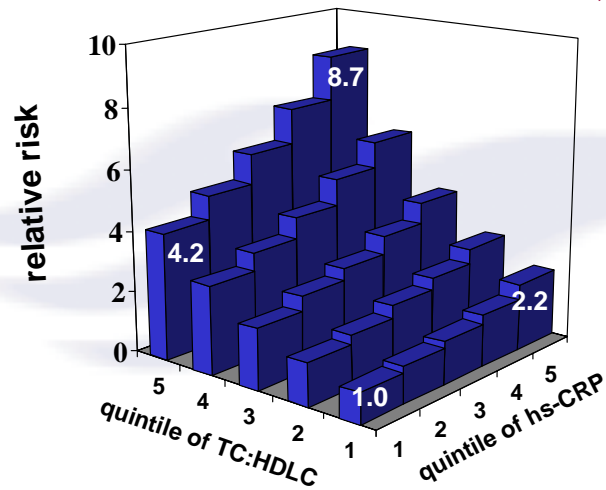
N Rifai & PM Ridker, Clin Chem 2001



## CRP is additive in risk prediction to lipid parameters



N Rifai & PM Ridker, Clin Chem 2001



## CRP and risk of coronary heart disease



### ■ secondary prevention

(= prevention of recurrent events)

## CRP & the risk of cardiovascular events in unstable angina



G Liuzzo, N Engl J Med, 1994

CRP (mg/L)	N	CABG-angioplasty	MI	death
< 3	11	2	-	-
≥ 3	20	12	5	2

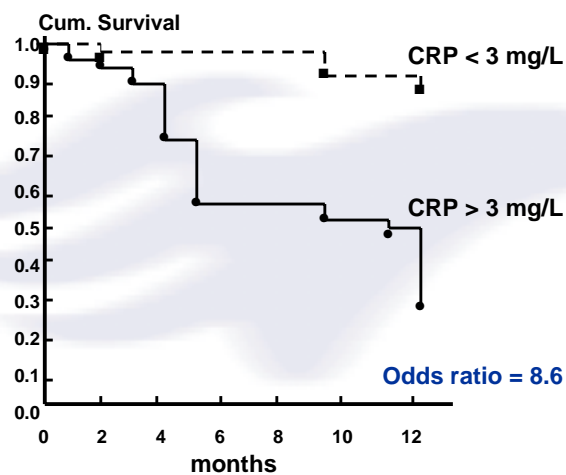
CRP ≥ 3 mg/L : 90% sensitivity and 82% specificity for in-hospital cardiovascular events

CRP >10 mg/L: 100% sensitivity

## CRP levels at discharge predicts event free survival in unstable angina



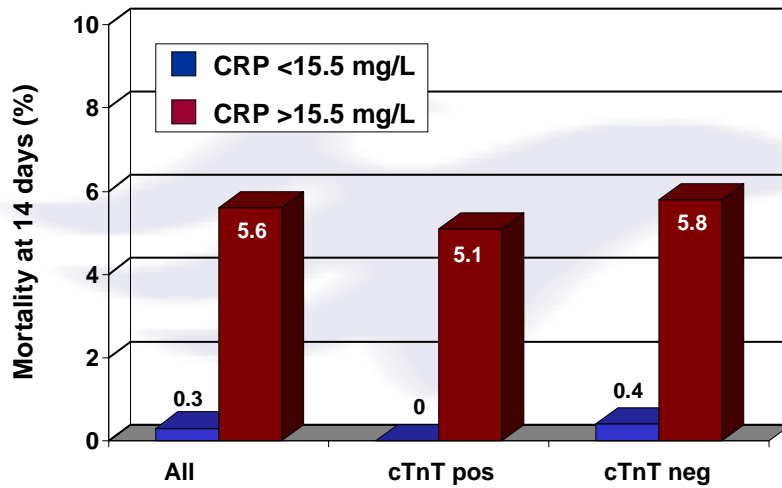
LM Biasucci, Circulation 1999



**Incremental prognostic value of CRP & troponin on admission in patients with UA or non-Q wave MI**



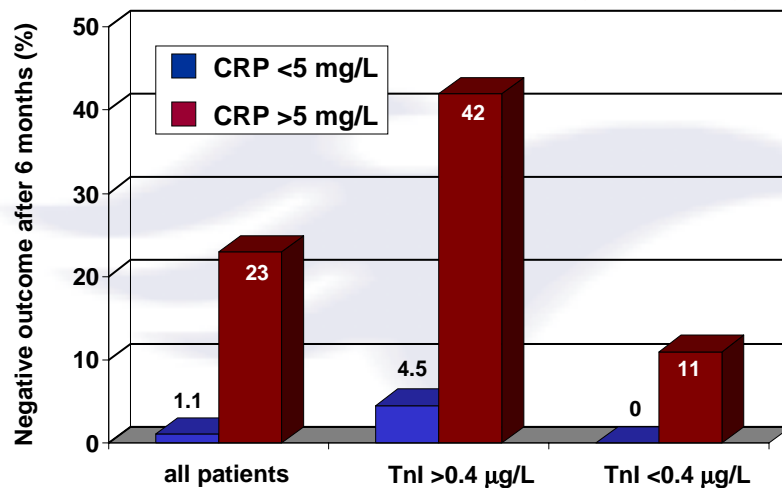
DA Morrow, J Am Coll Cardiol, 1998



**Incremental prognostic value of CRP & troponin on admission in patients with UA or non-Q wave MI**



RJ de Winter, Cardiovasc Res, 1999



## CRP & troponin in risk prediction



- troponin is a marker of myocardial necrosis (short term prognosis)
- CRP reflects the underlying inflammatory process of coronary heart disease (long term prognosis)
- CRP & troponin are independent and additive risk predictors

DA Morrow, J Am Coll Cardiol, 1998

AG Rebuzzi, Am J Cardiol, 1998

RJ de Winter, Cardiovasc Res, 1999

## Measurements of human CRP



- Traditional CRP methods in the clinical laboratory have limits of detection of 3-5 mg/L and are unsuitable for cardiovascular risk prediction
- Recently, several automated high-sensitivity methods have been developed for routine measurement of CRP concentrations <5 mg/L

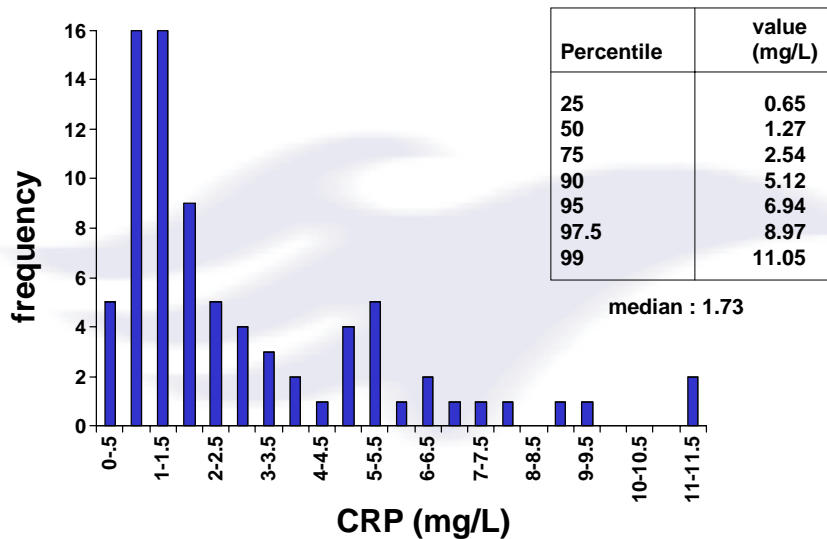
## Measurements of human CRP



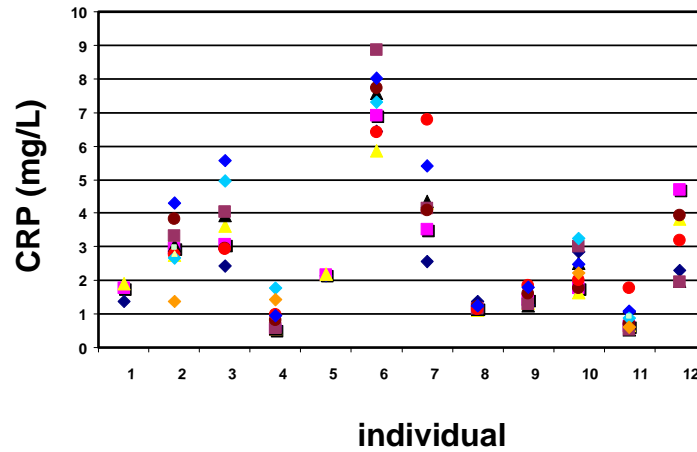
### Reference values (mg/L)

- range : 0.05 - 5
- median : 0.8
- P90 : < 3
- P95 : < 5
- P99 : < 10

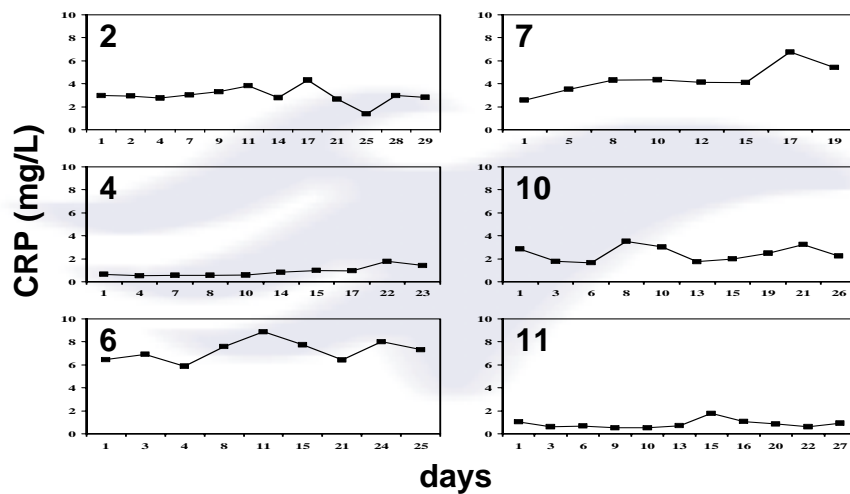
## CRP distribution in 80 apparently healthy individuals



**Biovariability of CRP for 12 individuals enrolled in a 1-month longitudinal study**



**Biovariability of CRP for 6 individuals enrolled in a 1-month longitudinal study**



**Variability analysis for CRP (mg/L) & cholesterol (mmol/L)**



EM Macy, Clin Chem, 1997

	<b>cholesterol</b>	<b>CRP</b>
mean conc.	5.1	1.7
CV <sub>A</sub> , %	1.0	5.2
CV <sub>I</sub> , %	9.3	42.2
CV <sub>G</sub> , %	19.4	92.5
index of individuality	0.48	0.46
index of heterogeneity	2.59	2.92
critical difference, %	26	118

**Age-adjusted correlation coefficients for log-normalized CRP levels and for lipid parameters**



PM Ridker, Circulation, 1999

**At baseline and after 5-year follow-up among study participants (CARE trial) randomly allocated to placebo**

<b>parameter</b>	<b><i>r</i></b>	<b><i>P</i></b>
CRP	0.6	0.001
Total cholesterol	0.37	0.001
LDL cholesterol	0.32	0.001
HDL cholesterol	0.74	0.001
triglycerides	0.49	0.001

## Factors associated with CRP concentration in blood



modifiable		non-modifiable	
factor	effect	factor	effect
obesity (BMI)	positive*	age	positive
smoking	positive	sex	higher in women
alcohol	negative		
coffee	positive	blood pressure	positive
strenuous exercise	positive	type II diabetes	positive
chronic bronchitis	positive		
periodontal disease	positive		
statins	negative		
aspirin	negative		
oral contraceptives	positive		

\* explains  $\pm 30\%$  of variation of CRP

## Cutt-off levels for appropriate differentiation of low and high risk patients

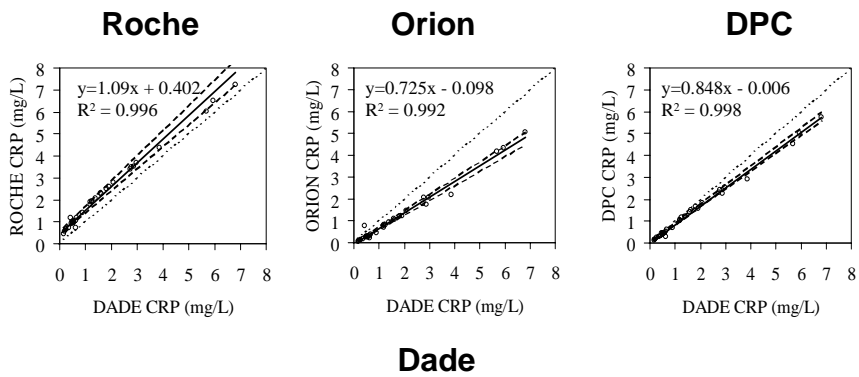


■ categorization into quintiles (quartiles or tertiles) are sample and population-dependent and therefore not universally applicable

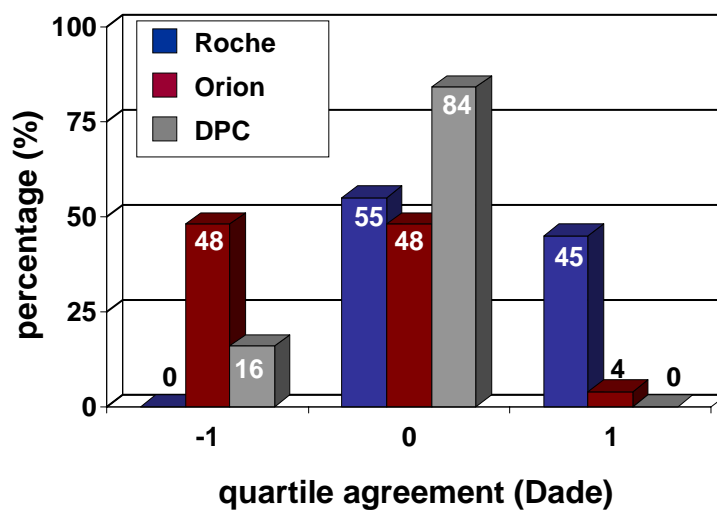
■ categorization using a single decision limit

In most studies a cutt-off point of 3 mg/L has been used (smaller category above 3 mg/L)

## Method comparison of four automated hs-CRP assays



## Quartile agreement of CRP methods using the Dade comparison method



**Agreement of CRP methods according to cut-off level of 3 mg/L**



n=31

method	low risk (n)	high risk (n)
DADE	27	4
ROCHE	23	8
ORION	28	3
DPC	28	3

**Blood sampling approach and decisions after analysis**



- when **below** decision limit (eg 3 mg/L): accept = low risk
- when **above** decision limit: take a second blood sample after two weeks or later
- when second sample is **again above** decision limit (with both samples <10 mg/L and CV <100%): accept = high risk
- when any sample >10 mg/L discard and start all over after two weeks or later

## Who should be assayed for CRP?



### For primary risk assesment:

- no screening policy
- individuals checked for CVD for any reason
- when cholesterol is assessed

**Combine CRP with TC:HDL**

N Rifai & PM Ridker, Clin Chem 2001

## Who should be assayed for CRP?



### For secondary risk assesment:

- **stable angina:** any time
- **unstable angina:** at admission or discharge
- **AMI:** at admission or outside the acute situation (three weeks after the event)

**Combine CRP with troponin**

DA Morrow, J Am Coll Cardiol, 1998

AG Rebuzzi, Am J Cardiol, 1998

RJ de Winter, Cardiovasc Res, 1999

## Factors associated with CRP concentration in blood



modifiable		non-modifiable	
factor	effect	factor	effect
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<b>statins</b>	<b>negative</b>		
<b>aspirin</b>	<b>negative</b>		
oral contraceptives	positive		

\* explains  $\pm 30\%$  of variation of CRP

## Modifying the inflammatory response by drug treatment



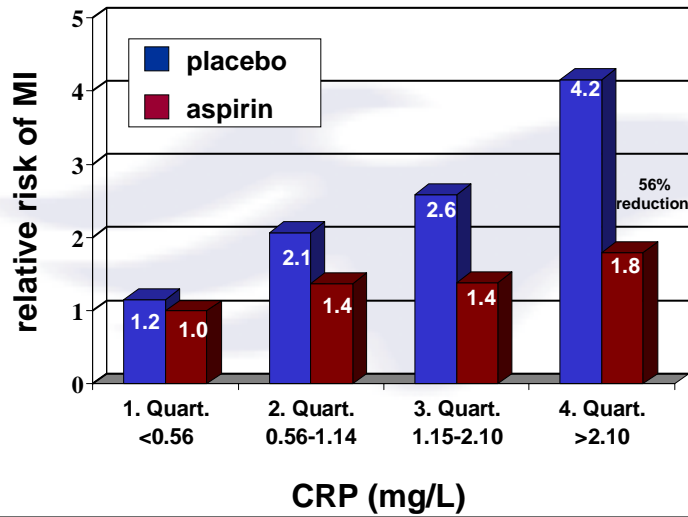
■ **Aspirin**

■ **Statins**

## Effect of aspirin on first myocardial infarction (PHS)



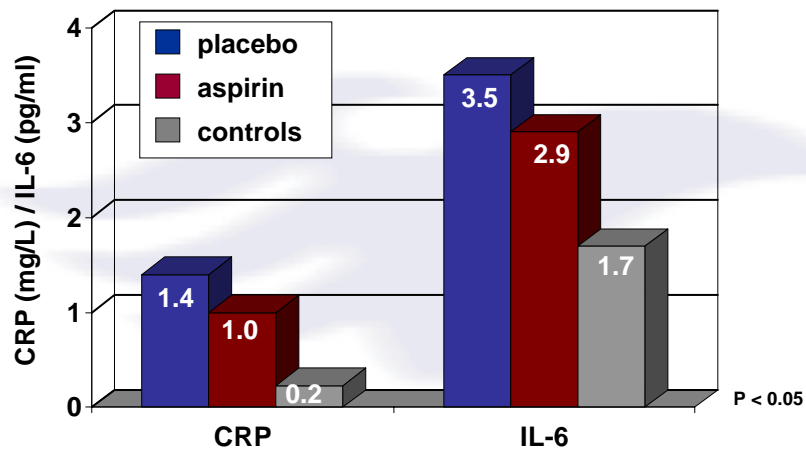
PM Ridker, N Engl J Med 1997



## CRP & cytokine reduction by aspirin in chronic stable angina



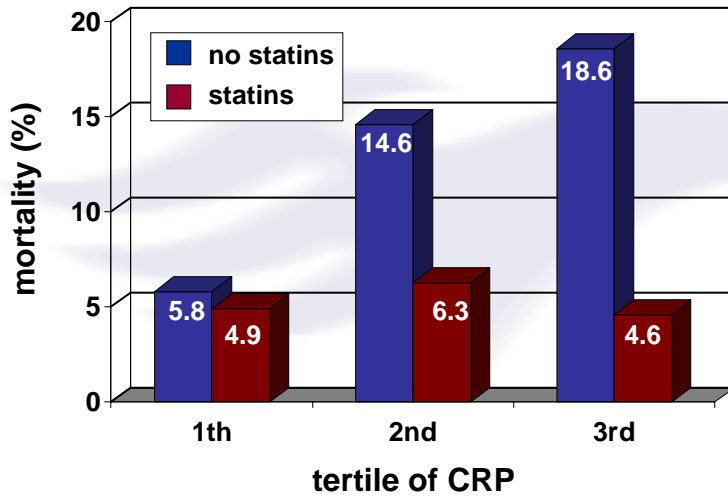
I Ikonmidis, Circulation 1999



### CRP & statin therapy in severe coronary artery disease



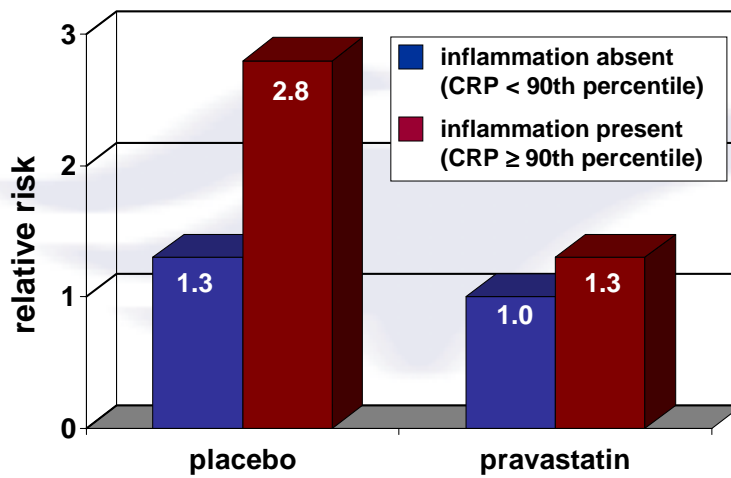
BD Horne, J Am Coll Cardiol 2000



### Inflammation, pravastatin and the risk of coronary events in patients with medium cholesterol (CARE study)



PM Ridker, Circulation 1998



**Inflammation, pravastatin and the risk of coronary events in patients with medium cholesterol (CARE study)**



PM Ridker, Circulation 1999

- the relationship between baseline CRP and risk of recurrent coronary events in CARE was attenuated and not statistically significant after treatment with pravastatin
- risk reduction was greater in individuals with increased CRP (54%) compared to those with low CRP (25%).
- treatment with pravastatin was associated with a 17.4% decrease in median CRP compared with a 4.2% increase with placebo use after 5 year.

**CRP fulfils most of the requirements needed to serve as a new risk factor for cardiovascular disease I**



- consistent results from 11 prospective population-based studies in initially healthy subjects
- the combined risk ratio for CHD is ~2 if subjects with baseline CRP in the upper tertile of the population distribution are compared with those in the lower tertile
- powerful: even stronger risk factor than lipids

**CRP fulfils most of the requirements needed to serve as a new risk factor for cardiovascular disease II**



- **independent of a wide variety of other risk factors**  
(remains strong predictor even after adjustment for contributing variables like lipid levels, age, smoking..)
- **comparable index of individuality to cholesterol**
- **stable analyte, automated and precise method**  
(standardization?)
- **additive to lipid screening in primary risk assessment**
- **additive to troponin in secondary risk assessment**

**CRP fulfils most of the requirements needed to serve as a new risk factor for cardiovascular disease III**



- **CRP levels can be lowered by aspirin, statins &**

**ALCOHOL**

