

High Sensitivity C-Reactive Protein In Cardiovascular Disease

C-Reactive Protein (CRP)

CRP is an acute phase reactant of molecular weight 125 kD. It is produced in the liver in response to tissue injury or infection. Production is regulated by interleukin-6 and is normally found in the plasma at concentrations of <2.0 mg/L (60% percentile). While its role in diagnosis of acute inflammation is well known, clinical studies have demonstrated that chronic, low-grade elevation of CRP indicates inflammatory conditions that are predictive of cardiovascular diseases (CVD).¹⁻³

CRP is associated with initiation of the fatty streak, development of atheromatous lesions, thinning of the fibrous cap and thrombosis. To accurately determine CRP results and assign risk of cardiovascular events, a high sensitivity method capable of accurately determining plasma concentrations of 0.2 mg/L is required.

Clinical Significance of High Sensitivity CRP (hsCRP)

Studies have clearly illustrated the role of hsCRP as an independent risk factor in the pathophysiology of atherosclerosis and acute coronary syndromes.¹⁻³ Measurement of hsCRP may be recommended in the following situations:³

- Evaluation and support of primary prevention therapies in patients with a 10-year risk of CVD of 10-20%.
- Prediction of initial and recurrent cardiovascular events in stable coronary disease or acute coronary syndromes as far as 5-10 years in advance.
- As a motivating factor to support patient lifestyle changes.

Measurement of hsCRP as a population screen is not recommended at this time.³

Increased blood levels of hsCRP demonstrate a synergistic effect on the relative risk of CVD when combined with an individual's lipid data. Several studies have shown that measurement of hsCRP is able to predict risk even in the absence of hyperlipidemia related risk.^{3,4}

Various factors affect the concentration of hsCRP in serum. Table 1 summarizes preanalytical factors that influence the concentration of hsCRP. It appears that there is little diurnal or seasonal variation, but wide within-subject and between-subject variances have been reported.³ Although population statistics are positively skewed, these studies illustrate that greater than 95% of subjects evaluated had an hsCRP less than 10 mg/L.



Specimen Collection

Fasting is not required, although preferred, to avoid assay interference due to lipemia. The patient should be metabolically stable.

Due to the increased within-individual variation for this analyte, it is recommended that two measurements be used to determine the patient's average hsCRP and risk level. Ideally, the specimens should be collected two weeks apart.

Result Interpretation

Assay values and the associated risks are shown as quintiles in tables 2A and 2B.⁵

Patients with values in quintile 1, with a hsCRP value of ≤ 0.7 mg/L, have the lowest cardiovascular relative risk. Similarly, patients in quintile 5 have a very high cardiovascular relative risk. Quintiles 2, 3, 4 represent low, moderate and high risks, respectively. In general, values of hsCRP from 0.5 – 1.9 mg/L imply moderate risk, while concentrations of 2.0 mg/L are associated with a two-fold relative risk compared to values in the first quintile. Risk of CVD associated with each quintile of hsCRP and cholesterol ratio is provided in Table 2B. Current data suggest that relative risks are not significantly different for males and females.⁵

To utilize the tables, determine the patient's hsCRP quintile and their lipid quintile (TC/HDL-C ratio) from Table 2A. The relative risk for future cardiovascular events based on the combination of the quintile of hsCRP and the quintile of TC/HDL-C may be determined using the data in Table 2B.

For example, a female patient with a hsCRP of 1.2 mg/L (quintile 3) and TC/HDL-C ratio of 5.0 (quintile 4) has a 4.2 fold increased risk of experiencing a future cardiovascular event compared to a low-risk patient with a hsCRP of 0.1 mg/L (quintile 1) and TC/HDL-C ratio <3.4 (quintile 1).

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If the blood level of hsCRP is greater than 10 mg/L, other causes should be considered and the test repeated after ruling out acute inflammation. Patients with persistent and unexplained elevation of hsCRP (>10 mg/L) should be evaluated for non-cardiovascular pathologies.

Reference intervals and classification of hsCRP results are still

subject to much discussion in the literature. Use of quintiles continues to be debated by medical and scientific leaders in this field.

Sheila Boss, PhD, FCACB, is a Clinical Biochemist and the Laboratory Director for the MDS International Reference Laboratory in Toronto.

**TABLE 1
PREANALYTICAL FACTORS WHICH INFLUENCE THE CONCENTRATION OF hsCRP**

INCREASE hsCRP

- Elevated blood pressure
- Increased body mass index
- Smoking
- Diabetes mellitus
- Estrogen/Progesterone hormone use
- Chronic infections
- Chronic inflammation
- Abnormal lipid profile:
 - Decreased HDL-cholesterol
 - Elevated triglycerides

DECREASE hsCRP

- Moderate alcohol consumption
- Exercise
- Weight loss
- Medications including:
 - Fibrates
 - Niacin
 - Salicylates
 - Statins



**TABLE 2A
QUINTILES OF hsCRP AND TOTAL CHOLESTEROL/HDL-CHOLESTEROL RATIO IN MEN AND WOMEN**

| Quintile | hsCRP mg/L | Cholesterol/HDL Ratio Women | Cholesterol/HDL Ratio Men |
|----------|------------|-----------------------------|---------------------------|
| 1 | 0.1 - 0.7 | <3.4 | <3.4 |
| 2 | 0.8 - 1.1 | 3.4 - 4.1 | 3.4 - 4.0 |
| 3 | 1.2 - 1.9 | 4.2 - 4.7 | 4.1 - 4.7 |
| 4 | 2.0 - 3.8 | 4.8 - 5.8 | 4.8 - 5.5 |
| 5 | 3.9 - 15.0 | >5.8 | >5.5 |

**TABLE 2B
RELATIVE RISK RATIOS FOR FUTURE CARDIOVASCULAR EVENTS USING hsCRP AND LIPID QUINTILES**

| | | hsCRP Quintile | | | | | |
|----------------------------|-----------|----------------|-----|-----|-----|-----|-----|
| | | 1 | 2 | 3 | 4 | 5 | |
| Cholesterol/HDL-C Quintile | Low Risk | 1.0 | 1.2 | 1.4 | 1.7 | 2.2 | |
| | High Risk | 2 | 1.4 | 1.7 | 2.1 | 2.5 | 3.0 |
| | | 3 | 2.0 | 2.5 | 2.9 | 3.5 | 4.2 |
| | | 4 | 2.9 | 3.5 | 4.2 | 5.1 | 6.0 |
| | | 5 | 4.2 | 5.0 | 6.0 | 7.2 | 8.7 |

References

1. Fritsma GMS. High sensitivity C-reactive protein. Clinical Laboratory Science 2001; 14 (4): 276 – 228.
2. Legrys, VA. The use of high sensitivity C-reactive protein in assessing the risk for coronary heart disease. Clinical Laboratory Science 2001; 14 (4): 243 – 246.
3. Pearson, TA et al. Markers in inflammation and cardiovascular disease. Application to clinical and public health practice. Circulation 2003; 107: 499-511.
4. Ridker, PM et al. Measurement of C-Reactive protein for the targeting of statin therapy in the primary prevention of acute coronary events. N Engl J Med 2001; 344 (26): 1959 – 1965.
5. Rifai, N and Ridker, PM. Proposed cardiovascular risk assessment algorithm using high sensitivity C-reactive protein and lipid screening. Clin. Chem. 2001; 47: 28-30.

Cervical Cytology Bethesda 2001 Reporting System

SPECIMEN ADEQUACY STATEMENT

Bethesda 2001 recommends that cervical cytology specimen adequacy be reported as either "UNSATISFACTORY" or "SATISFACTORY FOR EVALUATION". Unsatisfactory samples will be reported with explanatory text. Satisfactory samples will have comments on the presence or absence of the transformational zone components.

The absence of the transformational zone components does not imply that the sample is unsatisfactory. This should be emphasized, for during formulation of Bethesda 2001, the significance of transformation zone identification was discussed. The observation was recorded "longitudinal studies fail to show that women lacking transformation zone elements are at increased risk for epithelial lesions".

There are, in fact, several reasons why transformation zone components may not be detected, including some related to monolayer cytology and to screening instruments.

The medical literature, however, convincingly documents that monolayer cytology is superior to conventional smears with a decreased false negative rate and therefore increased ability to detect significant epithelial abnormalities.

It is our impression that many women are recalled for immediate repeat sampling when they have smears reported on which transformation zone components are not identified but the epithelial cells are normal.

As a routine, this early repeat sampling may result in unnecessary anxiety, is wasteful of resources and does not comply with current follow-up recommendations which indicate that in most situations women do not need to be recalled and the routine screening interval is appropriate.

Situations for which early recall may be indicated include smears obtained in follow-up of a previous epithelial abnormality and on which transformation zone elements are not observed, possibly when the sample represents the first Pap smear or when there is a defined clinical indication.

The elimination of the emotionally charged terminology "satisfactory but limited by" in the new Bethesda System is an important step and also presents an opportunity to review and emphasize follow-up recommendations.

ENDOMETRIAL CELLS AND WOMEN AGED 40 YEARS AND OLDER

Bethesda 2001 requires the reporting of exfoliated endometrial cells in cytology samples from women 40 years or older. Because cytologically normal exfoliated glandular endometrial cells are not associated with significant pathology in women less than 40, they need not be reported. This represents a change in reporting pattern, which may be confusing to clinicians. The following is intended to assist by providing clarification.

The significance of exfoliated endometrial cells in women 40 years or older cannot be determined by the laboratory, particularly in the absence of menstrual or hormonal data.

The following message will be appended as a reminder of the need to relate the finding of exfoliated endometrial cells to the clinical situation:

"Endometrial cells after age 40, particularly out of phase or after menopause, may be associated with benign endometrial, hormonal alterations and less commonly endometrial/uterine abnormalities."

Abraded endometrial cells obtained by vigorous sampling techniques are normal and do not have the same significance as exfoliated cells. These generally are present in sheets in contrast to exfoliated cells. Cytopathologists will differentiate between the two types when possible. The Cervex broom as well as the Cytobrush may obtain abraded cells. You should always note the types of devices used for collection on the cytology requisition.



An introductory comment to Bethesda 2001 states, "Cervico-vaginal cytology is a screening tool for squamous carcinoma and its precursor lesions. It is not an accurate test for the detection of endometrial lesions and should not be used for investigation of suspected endometrial abnormalities."

In other words, as a bonus, cervical vaginal cytology may result in the detection of significant endometrial abnormalities but without interpretation in clinical context may result in inappropriate endometrial sampling.

Conversely, when endometrial abnormalities are suspected on clinical grounds, direct endometrial sampling is indicated.

References

1. http://bethesda2001.cancer.gov/postwrkshp_recs.html
2. Selvaggi SM, Guidos BJ. Endocervical Component. Is it a determinant of specimen adequacy? *Diagnostic Cytopathology* 2002; 26 (1); 53-55.
3. Ontario Cervical Screening Collaborative Group. Interim Recommendations for Follow-up of Pap Test Results, Ontario Medical Review December 1997
4. <http://www.cancercare.on.ca/cervical/three>
5. Karim BO, Burroughs FH, Rosenthal DO and Ali SZ. Endometrial-type cells in Cervico-vaginal Smears. Clinical significance and cytopathologic correlates. *Diagnostic Cytopathology* 2002; 26; 123-127.

Frank E. Thompson, MD, FRCPC, is Medical Director MDS Ontario.

Ontario Update

LIPID REPORTING REQUIREMENTS

On February 10, 2003, MDS implemented use of target values for assessment of cholesterol, triglycerides, HDL-C and LDL-C concentrations. Accreditation requirements state that we must provide reference intervals for all analytes and as you know, the new lipid guidelines recommend target values for treatment, based on the individual's 10-year risk assessment for cardiovascular disease (CVD), rather than use of reference intervals. Since we do not have all of the criteria necessary to provide a valid risk interpretation of the lipid data, the patient report now includes all risk levels and their associated lipid goals. The target values provided on the MDS report were derived from the Canadian cholesterol working group

publication in CMAJ¹.

We have received a number of telephone calls from physicians who are concerned that the new report is very long and does not provide helpful information. We are presently reviewing all of your comments and are investigating alternative means to provide the required interpretive guidelines in a clear, concise format. Your suggestions for report changes are most welcome.

References

1. Fodor JG, et al. Recommendation for the management and treatment of dyslipidemia. Report of the Working group on Hypercholesterolemia and other dyslipidemias. CMAJ 2000; 162 (10); 1441-1447.

VITAMIN B12 AND FOLATE: ONTARIO REFERENCE INTERVAL CHANGES

In May 2003, MDS will implement the use of new automated technologies for the determination of serum vitamin B12 and folate as well as RBC folate. Internal validation studies have shown that the new methods are accurate and precise across the analytical and clinical ranges of interest; however, a change to the reference intervals for both analytes is required. Manufacturer recommended ranges will be implemented as follows:

| | Vitamin B12 pmol/L | Folate nmol/L | RBC Folate nmol/L |
|--------------|-----------------------|------------------|----------------------|
| Normal | > 133 | > 6.8 | > 164 |
| Inconclusive | 107 - 133 | 5.7 - 6.8 | N/A |
| Deficient | < 107 | < 5.7 | < 164 |

WHITE BLOOD CELL COUNTS: DISCONTINUATION OF THE % DIFFERENTIAL

Good laboratory and clinical practice requires that the white blood cell differential be reported in absolute numbers. The relative quantity (%) of the cell population has been retained due to its long time use in clinical practice whilst physicians became acquainted

with the more appropriate value. MDS Laboratories in Ontario will discontinue this practice as of May 1, 2003 bringing our reports in line with those of most Academic Centres in Canada.

MICROBIOLOGY SPECIMEN COLLECTION

For microbiology specimens collected in the clinician's office, please indicate the date and time of collection on the requisition form or on the label of the specimen. This information is now part

of the patient report and is an essential component of the MDS quality system.

