nascent field. *Journal of Nutrition*, 131:2787-2810.
GILLUM, R. F.; JACOBS Jr., D. R.; LUEPKER, R. V.; PRINEAS, R. J.; HANNAN, P.; BAXTER, J.; GOMEZ-MARIN, O.; KOTTKE, T. E. & BLACKBURN, H., 1984. Cardiovascular mortality trends in Minnesota, 1960-1978. The Minnesota Heart Survey. *Journal of Chronic Diseases*, 37:301-309.
O'CONNOR, S.; TAYLOR, C.; CAMPBELL, L. A.; EPSTEIN, S. & LIBBY, P., 2001. Potential infectious etiologies of atherosclerosis: A multifactorial perspective. *Emerging Infectious Disease*, 7:780-788.

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Association between influenza and coronary heart disease: how convincing is available evidence?

The first report of a temporal association between influenza epidemics and mortality from coronary heart disease (CHD) was published by British epidemiologists in 1978. However, the authors state that they "*could not hope to provide clear-cut evidence of a causal relationship, nor the sequence of the two conditions*" from their data (Bainton et al., 1978:238-239).

The old hypothesis that chronic inflammation and infection are involved in the pathogenesis of atherosclerosis and CHD has been revived in the 1980s and 1990s (Javier Nieto, 1998). Much of the current research has been stimulated by the finding that CHD patients more frequently present antibodies to pathogens such as *Chlamydia pneumoniae*, cytomegalovirus, and *Helicobacter pylori*, as well as serum markers of inflammation, than population controls (Muhlestein, 2001). Moreover, *Chlamydia* is frequently detected in atherosclerotic plaques of coronary arteries in CHD patients and induces atherosclerotic lesions in experimental models (Grayston, 2000). Evidence suggesting that this pathogen is involved in all stages of CHD is available. Chronic *Chlamydia* infections are associated with lymphoproliferative inflammatory responses characterizing the early stages of atherosclerosis, with changes in the lipid metabolism that may accelerate plaque formation, and with intra-plaque inflammation leading to plaque rupture and artery occlusion.

If a strong and plausible association between *Chlamydia* infection and CHD has been found, why should we look for other infections putatively associated with CHD? For at least two reasons: (a) when a careful adjustment for potential confounders (including socioeconomic factors) is made, the statistical association between the presence of anti-*Chlamydia* antibodies and CHD becomes rather weak (Danesh et al., 2000b), and (b) the positive association between low-grade chronic inflammation and CHD is unrelated to the presence of antibodies to *Chlamydia* or *H. pylori* (Danesh et al., 2000a). These findings suggest that other pathogens to which patients have been exposed, or their "pathogen burden", may be associated with chronic inflammation and CHD (Javier Nieto, 1998; Zhu et al., 2001). Here Azambuja & Duncan report an association between prior exposure to influenza and CHD mortality in the United States.

The ecological design represents a major limitation of the study by Azambuja & Duncan, since we cannot compare the exposure to several known risk factors for CHD and respiratory infections (including socioeconomic variables) in affected and non-affected subjects. The specificity (in the sense used by Bradford Hill) of a putative causal association between H1N1 influenza virus infection and CHD is disputable. We can hypothesize, for example, that a proportion of subjects infected with influenza virus during major epidemics are also more susceptible to other respiratory tract infections, including *Chlamydia*, perhaps because many of them are current or past smokers. In this example, therefore, *Chlamydia* infection and smoking represent major potential confounding factors that should be controlled for. Furthermore, the decline of CHD mortality over the last third of the twentieth century has alternatively been interpreted as a late consequence of the introduction of antibiotics, some of them active against *Chlamydia*, two to three decades earlier (Javier Nieto, 1998).

The biological plausibility of the etiologic association between influenza virus infection and CHD deserves further discussion. CHD pathogenesis has been associated with chronic inflammation caused by persistent *Chlamydia*, *H. pylori*, and cytomegalovirus infections (Muhlestein, 2001), but not by acute infections. Furthermore, in contrast with *Chlamydia* and cytomegalovirus, influenza A virus infection rarely involves the myocardium, the pericardium, or the vascular endothelium. The alternative hypothesis that a systemic, rather than local, inflammatory or autoimmune mechanism leads to plaque formation in influenza infection, proposed by Azambuja & Duncan, is insightful, but lacks further experimental support. On the other hand, the finding that influenza virus induces platelet aggregation, cit-

ed by Bainton et al. (1978) as a possible link between influenza and myocardial infarction, could be explored using modern epidemiological and experimental methods.

After an absence of 21 years, H1N1 viruses reappeared in the United States in 1977, infecting mostly college-aged individuals. It would be interesting to follow the incidence of CHD-associated events in this cohort of exposed subjects to confirm (or rule out) the association found by Azambuja & Duncan. Moreover, the availability of vaccines and amantadine or rimantadine prophylaxis provides the basis for clinical trials to address this topic.

BAINTON, D.; JONES, G. R. & HOLE, D., 1978. Influenza and ischaemic heart disease – A possible trigger for acute myocardial infarction? *International Journal of Epidemiology*, 7:231-239.

DANESH, J.; WHINCUP, P.; WALKER, M.; LENNON, L.; THOMPSON, A.; APPLEBY, P.; GALLIMORE, J. R. & PEPYS, M. B., 2000a. Low grade inflammation and coronary heart disease: Prospective study and updated meta-analyses. *BMJ*, 321:199-204.

DANESH, J.; WHINCUP, P.; WALKER, M.; LENNON, L.; THOMPSON, A.; APPLEBY, P.; WONG, Y. K.; BERNARDES-SILVA, M. & WARD, M., 2000b. *Chlamydia pneumoniae* IgG titres and coronary heart disease: Prospective study and meta-analysis. *BMJ*, 321:208-213.

GRAYSTON, J. T., 2000. Background and current knowledge of *Chlamydia pneumoniae* and atherosclerosis. *Journal of Infectious Diseases*, 181(Sup. 3):S402-S410.

JAVIER NIETO, F., 1998. Infections and atherosclerosis: New clues from an old hypothesis? *American Journal of Epidemiology*, 148:937-948.

MUHLESTEIN, J. B., 2001. Chronic infection and coronary heart disease. *Medical Clinics of North America*, 84:123-148.

ZHU, J.; JAVIER NIETO, F.; HORNE, B. D.; ANDERSON, J. L.; MUHLESTEIN, J. B. & EPSTEIN, S. E., 2001. Prospective study of pathogen burden and risk of myocardial infarction or death. *Circulation*, 103:45-51.

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In their insightful paper, Maria Inês Azambuja & Bruce Duncan consider the possibility that, at least in part, the pathogen burden of H1N1 influenza infection might have been an important determinant in the rise and fall of coronary heart disease (CHD) mortality observed in the 20th century. Although I consider the paper well-founded, with plausible arguments, the empirical evidence is not very convincing to me. My comments are general thoughts that may hopefully contribute to the design of new studies on the topic, which I agree is important and deserves further consideration.

The authors drawn on two major pieces of empirical evidence to supporting their hypothesis: (1) the similarity between the relative mortality associated with the 1918-1919 influenza pandemic and the distribution of CHD deaths in the period 1920-1985, across successive birth cohorts; and (2) the ecological association between an indirect measure of longer persistence of H1N1 influenza virus and delayed onset of decline in CHD death rates.

Concerning the first piece of empirical evidence, at least two questions might be raised:

- Part of the "drop" in CHD mortality that we see in the graph is actually derived from the fact that more recent cohorts have not actually finished evolving over time. Therefore, there are successive missing bars of mortality for the older ages in the more recent cohorts, which, if included, would make the decline less sharp.
- It does not seem to me that the increase in CHD mortality among younger individuals, which really appears to "follow" the influenza pandemic mortality, tends to return to the levels expected for a population not exposed to the burden of influenza.

Regarding the second piece of evidence, the ecological correlation is based only on nine areas, at least one of which may be an extreme observation. To get a feeling of the uncertainty underlying the data, we approximated the values by inspecting the graph, and performed a simple exercise of estimating a series of Spearman correlation coefficients for random samples of the data (with replacement, using bootstrap): in more than 40% of the samples, the Spearman correlation coefficient was not statistically significant at the 0.05 level.

I wish to congratulate the authors for sharing their provocative thoughts with us. If more robust evidence could be gathered, they might provide important clues regarding CHD epidemiology and be useful for public health purposes.