Reviewer's report

Title: Tobacco, Alcohol, and p53 Overexpression in Early Colorectal Neoplasia

Version: 1 Date: 26 September 2003

Reviewer: Anna Maria M Valentini

Reviewer's report:

General

Discretionary Revisions (which the author can choose to ignore)

Minor Compulsory Revisions (such as missing labels on figures, or the wrong use of a term, which the author can be trusted to correct)

1. Page 6, line 3 from the top. Change “Chromagin” to Chromogen

Major Compulsory Revisions (that the author must respond to before a decision on publication can be reached)

This is a very interesting paper of molecular epidemiology, well written by the Authors, and timely. However, there are some points that need clarification:

1. In a cited paper of Therry et al (reference 35), Authors correctly suggest to use two categories, such as high grade (severe dysplasia, in situ carcinoma, intramucosal carcinoma) vs low grade dysplasia (mild and moderate dysplasia) to improve the reliability in the classification for degree of dysplasia in the adenomas. On the contrary in this manuscript remains the division in adenomatous polyps, in situ carcinoma and intramucosal carcinoma. The definition like “carcinoma in situ” or intramucosal carcinoma “should be abandoned, since they lead to confusion.

2. In the INTRODUCTION it is correctly stated that in literature the p53 overexpression is over 60% in adenomas with severe dysplasia and invasive cancer. The finding of 21% and 34.9% of p53 overexpression in CIS and in IM carcinoma respectively is very lower. This decreases of nuclear staining can be due to the use of stored paraffin slides. Did you use for immunohistochemical technique stored slides?

3. In the immunohistochemical methods, procedure controls are necessary for the validation of staining results. Procedure controls also involve tissue controls. Please mention the positive control used.

4. In the background section of the Abstract the aim of the paper seems to know how epidemiological risk factors relate to p53 status, instead in the method section the aim of the analysis seems to know weather p53 status was different for risk factors for colorectal neoplasia relative to polyp free control group. The aim of the analysis does not overlap clearly to the the aim of the paper, or viceversa.

5. The analysis of the data seems quite confusing, and its epidemiological objectives not clear (to us,
of course). The baseline category in the polytomous logistic regression is not indifferent, and should be linked to the hypothesis tested or explored in the model. The more interesting comparisons (to us are: no disease vs disease with p53 negative (that explores the association of risk factors with disease only), and disease with p53 positive vs disease with p53 negative (that explores the association of risk factors with p53 status only). The comparison no disease vs disease with p53 positive does not disentangle the effect of risk factors on the variation of disease from their effect on the variation of p53 status. So, we think that could be interesting to use disease with p53 status negative as a reference category of the dependent variable in the polytomous logistic regression model.

6. If the aim of the paper is to know how epidemiological risk factors relate to p53 status, and the p53 relative frequency increases gradually from normal tissue to colorectal adenoma and then to cancer, why was not ordinal logistic regression used in the analysis to regress the variation of p53 status on epidemiological risk factors, instead of polytomous or dichotomous logistic regression?

Advice on publication: Unable to decide on acceptance or rejection until the authors have responded to the major compulsory revisions

Level of interest: A paper whose findings are important to those with closely related research interests

Quality of written English: Acceptable

Declaration of competing interests: None