Molecular biology as applied to the prevention of chronic-degenerative diseases. From preventive to predictive medicine

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Dear Sir,

Chronic-degenerative diseases (CDD), including atherosclerosis, cancer and chronic-obstructive pulmonary diseases, are the main causes of death in developed countries, accounting for approximately 90% of mortality. These pathologies arise from a complex network of risk-factors both exogenous and endogenous in origin. The role of preventive medicine in counteracting this major public-health problem is pivotal, especially since treatment of these diseases is difficult. The goal of preventive medicine is to shed light on the network of CDD-related risk factors in healthy subjects, thus allowing for primary preventive interventions aimed at removing harmful exposures, increasing host-defence mechanisms and activating targeted early-screening programs in susceptible individuals. CDD risk-factors are identified by anamnestic evaluation and questionnaires as well as chemical-laboratory analyses and the clinical examination of patients. For example, the risk assessment for atherosclerosis is performed by analysing diet and smoke-exposure habits, measuring blood pressure, lipid profile, etc., however, a similar approach may only in part be pursued for cancer because the majority of the related pathogenic phenomena occurs at molecular level.

Molecular biology has been developing a variety of tools useful to identify at molecular level the risk for developing CDD in relation to the occurrence of exogenous and endogenous risk factors. In the case of exposure to environmental genotoxic factors, it is possible to verify whether or not this situation triggered the host's defence mechanisms, thus affecting the biological target represented by DNA. The binding between genotoxic metabolites and DNA (DNA-adducts) can be analysed *in vivo* by non-invasive and sensitive procedures, such as ^{32}P-postlabelling,[1] thus quantifying the molecular effective dose of environmental risk factors.[2]

As far as endogenous risk factors are concerned, molecular biology employs an array of PCR-methods for analysing genetic susceptibility, thus disclosing the possibility of identifying in healthy subjects the genetic assets which increase the risk of developing CDD.[3] Interestingly in primary prevention, these methods are non-invasive because any cell of the organism can be used to perform the analyses. Examples of molecular risk-analysis for CDD are already available for cancer, atherosclerosis and glaucoma, the main cause of irreversible blindness in the world.

It has been demonstrated that cancer risk is increased where are a high levels of bulky DNA adducts,[4] cytogenetic alterations,[5] adverse polymorphisms of genes involved in activation and detoxification of carcinogens [6] and mutations of proto-oncogenes.[3]

DNA-adduct formation is known to be a common event in atherosclerosis.[7] The occurrence of endogenous risk-factors for atherosclerosis can be examined by testing the genetic asset of genes involved in thrombosis,[8] antioxiant activites [9] and endogenous production of atherogenic metabolites.[10]

The risk for glaucoma can be evaluated by exploring the genetic asset of genes encoding proteins [11] and antioxidant activities [12] maintaining the integrity of the trabecular meshwork, the structure regulating the outflow of the aqueous humour from the anterior chamber of the eye.

Recently, the decoding of the human genome disclosed the possibility of analysing the expression of thousands of genes in a single test using microarray technology.[13] It is therefore possible nowadays to move from single to multi-gene analysis of CDD risk-factors, thus considerably increasing the chance of having predictive results. This goal has already been achieved in animal experimental models. Microarray analysis successfully identified the gene-expression profile conferring the high risk of developing skin cancer in healthy rats after 28 days of exposure to a sunlight-mimicking source,[14] after which the predicted cancer occurred almost 1 year later.[15] The same method predicted, after 28 days, the selective occurrence of lung cancer in healthy smoke-exposed mice. The cancer occurred only in the strain bearing a mutation of the p53 gene while not appearing in their wild-type litter-mates,[16] the related cancers
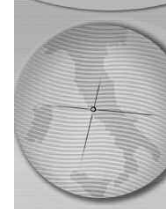
developed 9 months after exposure.[17] The main problem in transferring these methods to humans is that in preventative medicine materials need to by collected by non-invasive procedures. Great technological effort is ongoing to satisfy this prerequisite, the main target is to improve the sensitivity of these methods by using mRNA reverse-transcription to complementary-DNA and its amplification by PCR.[18] In such a way it is possible to use negligible amounts of tissue thus allowing for the possibility of applying non-invasive sampling procedures, such as mucosal scraping.

Therefore, it can be concluded that preventive medicine already has available an array of molecular methods to analyse the risk of developing CDD in individuals who are still healthy. It is likely that new effectual tools will become available in the next year. Hygienists should be involved both in the development and the application of these powerful methodologies to guarantee their availability to the preventive medicine of CDD.

References

1) Izzotti A. Detection of modified DNA nucleotides by post labelling procedures. Toxicol Meth 1998;8:175-205.

2) De Flora S, Izzotti A, Randerath K. DNA adducts and chronic degenerative disease. Pathogenetic relevance and implications in preventive medicine Mutat Res 1996;366(3):197-238.

3) Izzotti A, Cartiglia C, De Flora S. L'epidemiologia molecolare nell'identificazione dei soggetti suscettibili agli effetti dei cancerogeni ambientali. Annali di Igiene 1999;11(6):439-49.

4) Tang D, Phillips DH, Stampfer M. Association between carcinogen-DNA adducts in white blood cells and lung cancer risk in the physicians health study. Cancer Res 2001;61(18):6708-12.

5) Hagmar L, Brogger A, Hansteen IL, et al. Cancer risk in humans predicted by increased levels of chromosomal aberrations in lymphocytes: Nordic study group on the health risk of chromosome damage. Cancer Res 1994;54(11):2919-22.

6) Tanningher M, Malacarne D, Izzotti A, Ugolini D, Parodi, Drug metabolism polymorphisms as modulators of cancer susceptibility. Mutat Res 1999;436(3):227-61.

7) De Flora S, Izzotti A, Walsh D, Degan P, Petrilli GL, Lewtas J. Molecular epidemiology of atherosclerosis. FASEB J 1997;11(12):1021-31.

8) Doggen CJ, Cats VM, Bertina RM, Rosendaal FR. Interaction of coagulation defects and cardiovascular risk factors: increased risk of myocardial infarction associated with factor V Leiden or prothrombin 20210A. Circulation 1998;97(11):1037-41.

9) Izzotti A, Cartiglia C, Lewtas J, De Flora S. Increased DNA alterations in atherosclerotic lesions of individuals lacking the GSTM1 genotype. FASEB J 2001;15(3):752-7.

10) Jee SH, Song KS, Shim WH. Major gene evidence after MTHFR-segregation analysis of serum homocysteine in families of patients undergoing coronary arteriography. Hum Genet 2002;111(2):128-35.

11) Alward WL, Fingert JH, Coote MA, et al. Clinical features associated with mutations in the chromosome 1 open-angle glaucoma gene (GLC1A). New Engl J Med 1998;338(15):1022-7.

12) Izzotti A, Sacca SC, Cartiglia C, De Flora S. Oxidative deoxyribonucleic acid damage in the eyes of glaucoma patients. Am J Med 2003;114(8):638-46.

13) Izzotti A, Cartiglia C, Longobardi M, Tampa E, De Flora S. Valutazione del rischio di insorgenza di malattie cronico-degenerative mediante tecniche di genomica e postgenomica. In: Gammanco G, De Flora S, editors. Metodi Molecolari in Sanità Pubblica. Torino: Centro Scientifico Editore, 2004:195-201.

14) Izzotti A, Cartiglia C, Longobardi M, et al. Alterations of gene expression in skin and lung of mice exposed to light and cigarette smoke. FASEB J 2004;18(13):1559-61.

15) D'Agostini F, De Flora S. Potent carcinogenicity of uncovered halogen lamps in hairless mice. Cancer Res 1994;54(19):5081-5.

16) Izzotti A, Cartiglia C, Longobardi M, et al. Gene expression in the lung of p53 mutant mice exposed to cigarette smoke. Cancer Res 2004;64(23):8566-72.

17) De Flora S, Balansky RM, D'Agostini F. Molecular alterations and lung tumors in *p53* mutant mice exposed to cigarette smoke. Cancer Res 2003;63(4):793-800.

18) Eberwine J. Single-cell molecular biology. Nat Neurosci 2001;4 suppl:1155-6.