HUMAN WELFARE

BIOMONITORING in Terra dei Fuochi

NEW MANHATTAN PROJECT - SCIENCE FOR PEACE THE WORLD OVER
International Seminars
49° Session

PROJECT- 17

COORDINATOR: Prof. F. Buonaguro
I dati di incidenza e mortalità dell’Associazione italiana registri tumori (AIRT), 1998-2002


Figure 1. AIRT, 1998-2002, age-specific incidence rates, all cancers, males and females.
I dati di incidenza e mortalità dell'Associazione italiana registri tumori (AIRT), 1998-2002


Figura 2. Tassi di incidenza standardizzati (popolazione mondiale) per tutte le sedi escluse la cute, maschi. Fonte: Globocan 2002.

Tumore del polmone
(ICD-10 = C33-34)
Lung cancer
Tumore del colon-retto
(ICD-10 = C18-21)
Colorectal cancer
Tumore dello stomaco
(ICD-10 = C16)
Stomach cancer
Tumore della prostata (ICD-10 = C61)
Prostate cancer

Tumore della cervice uterina (ICD-10 = C53)
Cervical cancer
Tumore della mammella femminile (ICD-10 = C50)
Female breast cancer

Tumore del testicolo (ICD-10 = C62)
Testis cancer
Ecological studies of cancer incidence in an area interested by dumping waste sites in Campania (Italy)

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Summary. Cancer incidence was investigated in an area which has been affected by the illegal practices of dumping hazardous waste and setting fire to mismanaged waste. For the 35 municipalities of this area that are served by a Cancer Registry, municipal standardized incidence ratios (SIR) and hierarchical Bayesian estimators (BIR) were computed. Moreover, municipal spatial clustering and a Poisson regression by municipality index of waste-related exposure were performed for 10 cancer types. Increased municipality SIRs were found for some cancer types. The BIRs confirmed the increases for liver cancer in two municipalities. Statistically significant clusters were detected for liver, lung, leukaemia and soft tissue sarcomas. In the regression analysis, testis cancer showed significant trend with the index of waste-related exposure (RR = 1.18).

Key words: cancer incidence, waste dumping sites, testicular neoplasms, liver neoplasms.
The Map of the study area of Naples Province in Campania Region (Italy)
Biomedical Environmental Studies

1. Environmental analysis and monitoring;
2. Incidence of diseases in specific areas;
3. Correlation between doses and biomedical data
Biomedical Environmental Studies

1. Environmental analysis and monitoring;
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Several studies on “Environmental analysis and monitoring”
Tabella 2.14: Piano sorveglianza PCDD/PCDF e DL-PCB - matrice suolo: numero campioni previsti nel triennio 2008-2010

<table>
<thead>
<tr>
<th></th>
<th>Zona A Contaminazione medio-alta</th>
<th>Zona B Contaminazione medio-bassa</th>
<th>Zona C Contaminazione bassa</th>
<th>Ricontrolli da incrocio dati ARPAC/IZSM</th>
<th>Totali anno</th>
</tr>
</thead>
<tbody>
<tr>
<td>Campioni suolo I anno</td>
<td>160</td>
<td>120</td>
<td>10</td>
<td>50</td>
<td>340</td>
</tr>
<tr>
<td>Campioni suolo II anno</td>
<td>50</td>
<td>60</td>
<td>20</td>
<td>40</td>
<td>240</td>
</tr>
<tr>
<td>Campioni suolo III anno</td>
<td>70</td>
<td>80</td>
<td>20</td>
<td>10</td>
<td>180</td>
</tr>
<tr>
<td>TOTALI</td>
<td>320</td>
<td>270</td>
<td>50</td>
<td>100</td>
<td>740</td>
</tr>
</tbody>
</table>

Legenda: Le zone A, B, C sono quelle della studio APAT del 2004-2005 che, per una maggiore aderenza alla situazione di uso dei suoli, sono state ridotte con informazioni legate al territorio come di seguito specificato:


** Zona B: Zona B1-APAT rimodulata con zone urbanizzate, escludendo zone boschive e montane (ex Classe A1 APAT).

*** Zona C: Zona C1-APAT rimodulata con zone collinari e montane (quota>600 mt) e zone ad uso forestale.
Geographical areas of soil contamination

- Il SIN Litorale Domizio Flegreo conta 77 comuni;
- La c.d. Terra dei Fuochi conta 55 comuni;
- 38 comuni appartengono sia al SIN sia alla c.d. TDF
Biomedical Environmental Studies

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Mortality frequency for bladder cancer

Figure 4 – Mortalità per tumore della vescica (A: popolazione totale; B: uomini)

Comba et al., 2014
Biomedical Environmental Studies

1. Environmental analysis and monitoring;
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3. Correlation between doses and biomedical data
Bladder Cancer and role of Heavy Metals (i.e. Arsenic)
Correlation between doses and biomedical data: environmental Arsenic concentration and bladder cancer incidence
New methods, in particular “omics”, currently allow specific approaches able to correlate exposure to a potential carcinogen to specific cancers.

Introducing the concept of

Meet in the middle - MITM
Causality

Von Wright stresses the importance of manipulation:

something (p) is a necessary cause of q if we can eliminate p and show that also q disappears

something (p) is a sufficient cause of q if we can introduce p in the absence of q and show that q appears

However this model is rarely relevant to epidemiology
Multicausality

• More than one cause of same disease
• Cancer contributory factors
  – Previous infections
  – Immunodeficiency
  – Genetic predisposition
  – Exposure to environmental carcinogens
  – Body weight
  – Life style (i.e. alcohol drinking and smoking)

So illness rarely has one cause and the causes are rarely necessary and sufficient
The “exposome”

Measurements of biomarkers can provide information about both exogenous and endogenous exposures

Overview of the process of exposure and biotransformation and possibilities of biomonitoring
http://www.plosmedicine.org/article/info:doi/10.1371/journal.pmed.1001117
Methods available

- Long-term epigenetic markers
- Metabolomic markers of dietary habits and hypertension
- Envirogenomarkers: chemical exposures and omics, omics and disease
Epigenetics

Determinants of methylation as a cancer short-term as well as long-term risk marker

• **Short-term marker:** DNA methylation is dynamic and unstable and can change rapidly in response to immediate exposures.

  • **Evidence:** Cyclical methylation at ER target genes

• **Long-term marker:** DNA methylation is a stable epigenetic mark, through cell division, and may represent a long term history of exposures, and accumulated cancer risk.

  • **Evidence:** DNA methylation associated with former smoking
    Shenker et al, Epidemiology, 2013
Methylation Index to detect former smokers (or to quantify former exposure)

The study we propose: Biomonitoring in the Campania region

- a specific cancer: bladder cancer;
- a specific population to study and the related controls;
- "omics" methodology

within the context of MITM approach
The study we propose:
Training set followed by validation stage
The study we propose:
Aims to be achieved

Specific Aims

1. To verify different exposure levels to heavy metals (in particular As) in geographic areas at high and low cancer incidence of the Campania region;

2. To evaluate accumulation of aberrant methylation in cases and controls, with correlation to environmental exposure length (age and years of residence) as well as to other co-factors including cigarette-smoking and working activities.

3. To identify individual susceptibility to chemical carcinogenesis for abnormal accumulation of genetic and epigenetic changes, associated to inefficient cellular response to oxidatively-induced DNA damage and polymorphism of DNA-repair genes (Savina et al. 2016).
The study we propose:
Methods to be implemented

Biomolecular methods:
TP53 gene mutations by ultra-deep-next generation sequencing- NGS

Methylation of TP53 gene
Evaluation of TP53-promoter methylation in the 638-978 nt region and compared to control regions (i.e. repetitive LINE sequences) (Jensen et al., 2009; Intarasunanont et al., 2012).

Analysis of polymorphisms associated to As genotoxic damages
Several genes gave been involved in such pathogenic mechanisms, including those of FA/BRCA pathway and their related SNPs, (Levran et al., 2005); and BER pathway (Xing et al 2012).
The study we propose:
Further methods to be implemented

**Citogenetics:** COMET assay.
- Il Comet assay (Single Cell Gel Electrophoresis o SCGE)

**Citogenetics:** DEB-MMC Test

**Genotoxicity:** reporter-genes p53 Calux

**Biochemical:** metabolomic analysis by NMR

**Bioinformatics and Statistical Analysis** will be implemented as needed.
In particular, it will be implemented the non-parametric Mann-Whitney U test, T test for independent data and the Fisher exact test (test di Fisher-Yates), and differences will be considered significant for p <0.05.
Correlation between protein levels and clinical/biochemical data will be evaluated using Pearson correlation coefficient and correlation matrix.
The odds ratio (OR with confidence limit of 95%) of different factors will be calculated by multiple regression logistic
The current biomonitoring project focused on a specific area of the Campania Region, will be used as pilot model for the establishment of a biomonitoring center to be used in other geographical regions with complex exposure to multiple environmental carcinogenic agents.