europass Curriculum vitae

INFORMAZIONI PERSONALI

Chiara Mazziotta

(Italia)

OCCUPAZIONE PER LA QUALE SI CONCORRE

Borsa di studio per attività di ricerca post lauream

ISTRUZIONE E FORMAZIONE

11/2018–alla data attuale

Dottorato di ricerca in Medicina Molecolare

Livello 8 QEQ

Università degli studi di Ferrara, Ferrara (Italia)

09/2016–27/09/2018

Laurea magistrale in Biotecnologie per l'ambiente e la salute (LM-8)

Livello 7 QEQ

Università degli studi di Ferrara, Ferrara (Italia)

Conseguimento titolo il 27/09/2018. Valutazione: 110/110 e lode.

- Conoscenze in ambito farmacologico (progettazione preparazione di farmaci e principi attivi biotecnologici, farmacologia in biotecnologie, cenni di cosmesi, fitochimica applicata e biotecnologie delle piante officinali).
- Competenze in ambito biotecnologico e bioinformatico (bioniformatica e analisi dei genomi, consultazione di banche dati, biologia applicata alle biotecnologie, astrobiologia, biocatalisi applicata).
- Conoscenze e competenze in ambito ambientale (biochimica ambientale, impatto ambientale, ecologia, controlli e certificazioni di qualità ambientale, valutazione di impatto ambientale e conoscenza delle normative inerenti).
- Conoscenze in ambito biomedico e molecolare (microbiologia, virologia molecolare, allergologia e immunopatologia, biomateriali e tecnologia dei dispositivi medici, basi molecolari delle patologie, OMICA e diagnostica molecolare).

Tesi : **Cellule staminali mesenchimali dell'adulto (hMSCs) per lo studio di un biomateriale innovativo: identificazione di geni e microRNA osteogenici.**

07/10/2013

Laurea in scienze Biologiche (L-13)

Livello 6 QEQ

Università degli Studi di Ferrara, Ferrara (Italia)

09/2008–07/2013

Maturità scientifica

Livello 4 QEQ

Liceo Scientifico G. Peano, Marsico Nuovo (Italia)

ESPERIENZA PROFESSIONALE

2017–2018

Tirocinio formativo

Università di Ferrara, Dipartimento di Morfologia, Chirurgia e Medicina Sperimentale, Sezione di Patologia, Oncologia e Biologia Sperimentale, Ferrara (Italia)

Attività di ricerca sull'associazione di virus oncogeni a DNA (Polyomavirus e Papillomavirus) e cancro umano, e sulla biocompatibilità di nuovi biomateriali per la rigenerazione ossea.

Svolgimento tesi sperimentale dal titolo "Cellule staminali mesenchimali dell'adulto (hMSCs) per lo studio di un biomateriale innovativo: identificazione di geni e microRNA osteogenici", relativo articolo in

preparazione.

Altri Progetti di ricerca: 1) Associazione tra Leucemia Linfatica Cronica (LLC) e virus polyoma delle cellule di Merkel (MCPyV); 2) Studio di biocompatibilità, osteoinduttività e osteoconduttività di nuovi biomateriali per la rigenerazione dell'osso; 3) Analisi di miRNA coinvolti nel differenziamento osteogenico; 4) NIBSC (International collaborative study to assess the suitability of candidate 1 st WHO International Standards for HPV types 6, 11, 31, 33, 45, 52 and 58 DNA.)

Le tecniche di laboratorio apprese sono le seguenti:

- Test ELISA indiretto
- Allestimento colture cellulari: linee cellulari umane immortalizzate; cellule staminali mesenchimali umane (hMSC), allestimento di colture cellulari su differenti biomateriali
- Campioni di tessuto: estrazione e colture di cheratinociti, fibroblasti e cellule mesenchimali umane
- Estrazione e quantificazione di acidi nucleici e proteine da campioni di tessuto congelati ed FFPE
- Retrotrascrizione
- PCR: end point; qRT-PCR; ddPCR; RCA, PCR Array
- Gel di agarosio ed elettroforesi
- Estrazione e quantificazione di DNA, RNA e proteine da colture cellulari e campioni di tessuto
- Western Blot
- Immunoistochimica ed immunofluorescenza
- Trasfezione cellulare
- Colorazioni cellulari

03/2016–07/2016 **Tirocinio formativo**

Università di Ferrara, Dipartimento di Medicina Sperimentale e Diagnostica, Sezione di Genetica medica, Ferrara (Italia)

Diagnosi di malattie genetiche (Corea di Huntington, atassie spinocerebellari autosomiche dominanti, atassia di Friedrich) e analisi di mutazioni nel gene BRCA2 per rilevare la predisposizione genetica allo sviluppo del carcinoma della mammella ed ovarico.

Le tecniche di laboratorio apprese sono le seguenti:

- Sequenziamento mediante metodo di Sanger
- Purificazione DNA
- Estrazione di DNA da gel di agarosio
- Elettroforesi capillare

COMPETENZE PERSONALI

Lingua madre: Italiano

Lingue straniere

	COMPRENSIONE		PARLATO		PRODUZIONE SCRITTA
	Ascolto	Lettura	Interazione	Produzione orale	
inglese	B2	B2	B2	B2	B2

Livelli: A1 e A2: Utente base - B1 e B2: Utente autonomo - C1 e C2: Utente avanzato
Quadro Comune Europeo di Riferimento delle Lingue

Competenze comunicative

- Buone competenze comunicative acquisite durante i tirocini formativi presso l'Università di Ferrara.
- Buone competenze collaborative con i colleghi e capacità di assumere diversi compiti in un gruppo di ricerca.

- Ottime competenze relazionali con adulti e bambini acquisite durante la mia esperienza come volontaria presso l'associazione AVIS (associazione volontari italiani sangue).

Competenze organizzative e gestionali

- Ottime capacità organizzative e collaborative acquisite durante gli anni di studio universitari e i tirocini formativi in laboratori di ricerca.
- Buone capacità di gestire situazioni difficili ed impreviste in ambito lavorativo.
- Disponibilità, se richiesto, ad effettuare trasferte in zone diverse da quella dove si svolge la normale attività lavorativa, ed eventualmente a trasferirsi per lavoro.

Competenze professionali

- Buona padronanza dell'uso di apparecchiature e strumenti da laboratorio biomolecolare.
- Buona capacità di interpretazione di cromatogrammi per la diagnosi di malattie genetiche.
- Buone capacità di allestimento e interpretazione di test per la diagnosi di malattie genetiche quali Corea di Huntington, Atassie Spinocerebellari dominanti, Atassia di Friedrich.
- Capacità di allestimento di test di predisposizione genetica allo sviluppo del carcinoma alla mammella ed ovarico: analisi di oncologia molecolare del gene BRCA2.
- Buone capacità di allestimento di studi di associazione tra virus oncogeni a DNA e tumore.
- Padronanza di tecniche per lo studio della biocompatibilità di nuovi materiali per la rigenerazione dell'osso.
- Buone capacità di stesura di un progetto di ricerca.

Competenze digitali

AUTOVALUTAZIONE				
Elaborazione delle informazioni	Comunicazione	Creazione di Contenuti	Sicurezza	Risoluzione di problemi
Utente avanzato	Utente avanzato	Utente autonomo	Utente autonomo	Utente autonomo

Competenze digitali - Scheda per l'autovalutazione

- Buona padronanza degli strumenti Microsoft Office.
- Buona capacità di utilizzare supporti informatici per effettuare ricerche in banche dati come NCBI.
- Buona conoscenza del software GeneMapper e del software Chromas lite per l'analisi dei dati di sequenziamento.
- Buona capacità di utilizzo del software CFX Manager per l'analisi dei dati qRT-PCR e del software GraphPad Prism 7.
- Buona conoscenza del software QuantaSoft per l'analisi dei dati ddPCR.

Altre competenze

- Buone conoscenze delle tecniche di primo soccorso ottenute frequentando un seminario di BLSD.
- Buone competenze didattiche, ottenute aiutando giovani studenti nello svolgere i compiti a casa una volta a settimana.

Patente di guida B

ULTERIORI INFORMAZIONI

Seminari e corsi di formazione

- Aggiornamenti nell'analisi degli acidi nucleici con "droplet digital PCR" (ddPCR) e del trascrittoma di popolazioni cellulari specifiche tramite "Cell Sorting" e "digital sequencing" (ddSEQ), Università di Ferrara, 18/10/2017
- Seminario storico "Dalla botanica medica alla moderna farmacologia", Università di Ferrara, 13/12/2017
- "Novartis commitment to onco-hematology: the Translational Clinical Oncology Pipeline (TCO)",

Univeristà di Ferrara, 02/03/2018

- "Droplet Digital PCR Scientific Conference 2018", Università di Ferrara, 21/06/2018
- Corso di formazione in materia di impiego degli animali a fini scientifici, Università di Ferrara, 18/09/2018

Certificazioni: FORMAZIONE SICUREZZA NEI LUOGHI DI LAVORO AI SENSI DEL D.LGS.81/2008 E S.M.I., 21/02/2017

Documenti collegati Error: Reference source not found

Presentazioni
- "Valutazione dell'infezione del virus polyoma MCPyV in pazienti affetti da Leucemia Linfatica Cronica", presentazione orale, Scientific retreat, San Vito di Ostellato (Fe), 20/11/2017

Appartenenza a gruppi / associazioni: Membro dell'Associazione Italiana Biologia e Genetica (AIBG), a partire da Giugno 2018

Congressi: XVIII Congresso Nazionale AIBG, Ferrara, 21-22/09/2018

Abstract a congresso

Documenti collegati ABSTRACT 13.pdf, ABSTRACT AIBG 1.pdf, ABSTRACT AIBG 2.pdf, ABSTRACT AIBG 3.pdf, ABSTRACT AIBG 4.pdf, ABSTRACT AIBG 5.pdf, ABSTRACT AIBG 6.pdf, ABSTRACT AIBG 7.pdf, ABSTRACT AIBG 8.pdf, ABSTRACT AIBG 9.pdf, ABSTRACT AIBG 10.pdf, ABSTRACT AIBG 11.pdf, ABSTRACT AIBG 12.pdf

Trattamento dei dati personali: Autorizzo il trattamento dei dati personali contenuti nel mio curriculum vitae in base all'art. 13 del D. Lgs. 196/2003 e all'art. 13 del Regolamento UE 2016/679 relativo alla protezione delle persone fisiche con riguardo al trattamento dei dati personali.

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ABSTRACT AIBG 1.pdf

18° Congresso Associazione Italiana di Biologia e Genetica Generale e Molecolare, 21 Settembre 2018. In stampa.

Circulating microRNAs found to be dysregulated in pleural mesothelioma patients as potential new biomarkers.

Ilaria Bononi, Francesca Frontini, Antonella Santoro, Elena Torreggiani, Lucia Oton-Gonzalez, John Charles Rotondo, Carmen Lanzillotti, Chiara Mazziotta, Maria Rosa Iaquinta, Fernanda Martini, Mauro Tognon.

Malignant pleural mesothelioma (MPM) is a very aggressive malignancy of the pleural surface due to the asbestos exposure. The MPM incidence increased over the past decade, with an estimated peak in 2025. MPM will continue to represent a significant health concern even after the peak incidence mentioned above (1). As MPM is largely unresponsive to chemo- and radio-therapies and considering the long latency period of MPM onset, the identification of new and specific markers is of a paramount importance for an early diagnosis and treatment of MPM. In recent years, together with protein markers, microRNAs (miRNAs) from MPM cells or sera, have been proposed as new biomarkers (2). Indeed, miRNAs expression was found dysregulated, both in cancer cells and sera, in patients affected by tumors of different histotypes, including MPM. Moreover, it has been reported that circulating miRNAs are stable in biological fluids and, consequently, they could be employed as potential MPM biomarkers. In this investigation, circulating miRNAs from serum samples of MPM patients and healthy subjects (HS) were comparatively analyzed by microarray and RT-qPCR technologies. Our results allowed (i) to select miR-3665, an endogenous stable miRNA, as the internal control to quantify circulating miRNAs in our analyses; (ii) to detect miR-197-3p, miR-1281 and miR-32-3p up-regulated in MPM compared to HS (3). In conclusion, the three circulating up-regulated miRNAs, mentioned above, are proposed as potential new MPM biomarkers and targets for innovative therapeutic approaches.

References

1. Park E.K. et al. Environ. Health Perspect.119:514-8, 2011.

2. Balatti V. et al. J. Thorac. Oncol. 6:844-51, 2011.

3. Bononi I. et al.Oncotarget.7:82700-11, 2016.

ABSTRACT AIBG 2.pdf

18° Congresso Associazione Italiana di Biologia e Genetica Generale e Molecolare, 21 Settembre 2018. In stampa.

Workers ex-exposed to asbestos fibers carry circulating dysregulated microRNAs.

Francesca Frontini, Ilaria Bononi, Antonella Santoro, Paola Rizzo, John Charles Rotondo, Maria Rosa Iaquinta, Carmen Lanzillotti, Chiara Mazziotta, Fernanda Martini, Mauro Tognon.

The WHO estimates that 125 million workers are at present exposed to asbestos fibers. The asbestos is a cancerogenic mineral responsible of a fatal cancer, the malignant pleural mesothelioma, (MPM), which causes more than 100,000 deaths/year (1). MPM could arise in a long latency period (up to 50 years) after the asbestos exposure. Workers/subjects potentially at risk may benefit of an early diagnosis based on specific biomarkers. Cellular and circulating microRNAs (miRNAs) have been proposed as new biomarkers (2). Indeed, miRNAs expression was found dysregulated in patients affected by MPM, suggesting their potential role as oncogenes or tumor suppressor genes. The use of circulating miRNAs as MPM biomarkers may simplify the surveillance procedure of subjects exposed to asbestos and facilitate the early detection of MPM, with a simple blood test. In this investigation, circulating miRNAs from serum samples of workers ex-exposed to asbestos (WEA) and healthy subjects (HS) were comparatively analyzed by microarray and RT-qPCR technologies. Our results allowed to select miR-3665, an endogenous stable miRNA, as the internal control to quantify in our analyses circulating miRNAs, and to detect miR-1281 up-regulated in WEA compared to HS (3). As miR-1281 was found up-regulated also in MPM patients, this miRNA could be proposed as potential new predictive MPM biomarker in WEA, and in screening/preventive programs.

References

1. Foddis R. et al. J. Thorac. Dis. 10 (Suppl 2):S360-8, 2018.

2. Balatti V. et al. J Thorac. Oncol. 6:844-51.2011.

3. Bononi I. et al. Oncotarget 7:82700-11, 2016.

ABSTRACT AIBG 3.pdf

18° Congresso Associazione Italiana di Biologia e Genetica Generale e Molecolare, 21 Settembre 2018. In stampa.

Human adipose stem cells induced to osteogenic differentiation by an innovative hydroxyapatite hybrid scaffold.

Maria Rosa Iaquinta, Elisa Mazzoni, Chiara Mazziotta, Carmen Lanzillotti, Elena Torreggiani, Mauro Tognon and Fernanda Martini.

In this study we investigated human adipose stem cells (hASCs) for biocompatibility, osteoconductivity and osteoinductivity effects of an innovative hydroxylapatite hybrid scaffold composed of granular hydroxyapatite and collagen (Coll/HA) materials (1-2). In hASCs cultures, grown on this scaffold, showed normal cellular cytoskeleton organization, cellular morphology and cell viability when investigated by immunohistological staining (IHS), metabolic assay (Alamar blue) and scanning electron microscopy (SEM) (3). Expression of the extra-cellular matrix (ECM), adhesion molecule, and osteogenic genes were evaluated by quantitative PCR (Q-PCR) array technologies. Osteocalcin, osteopontin, Alkaline phosphatase (ALP) and phosphorylated focal adhesion kinase p-FAK (Tyr397) proteins were detected in hASC by HIS and E.L.I.S.A.assays. It turned out that the cytoskeleton architecture of hASC seeded on biomaterial was well organized. hASC expression of CLEC3B, LAMB3, ITGAM, ITGA3, LAMA2, ITGB5, COL6A2, SELE, COL6A1, and SPP1 genes was up-regulated. In hASC culture mRNAs of 24 genes of the ossification pathway, i.e. CSF 2/3, SP7, SPP1, TNFSF11, BMPR1B, BMP1/2, BGLAP, IGF1, NOG, RUNX2, TGFB1, EGFR, FGFR1/2, VDR, TWIST1, SOX9, ALPL, IGF1R, COL1A1, EGF, ITGA2 were upregulated compared to the control. The hASC culture expressed the osteogenic proteins such as ALP and osteocalcin. In addition, the mineralized matrix was present in hASC culture. These results suggest that the hASC cultures are a reliable tool to evaluate the biocompatibility, osteoconductivity of the biomaterial. Our data demonstrate that the innovative scaffold provides the good microenvironment in which hASCs adhesion and proliferation are enhanced, while inducing the up-regulation of osteogenic genes with improvement in matrix mineralization.

References

1. Mazzoni E. et al. FASEB J. 31:4555-65, 2017.

2. D'Agostino A. et al. J. Oral Maxillofac. Surg 74 1238.e1-e15, 2016.

3. Manfrini M. et al. J. Cell. Physiol. 228:1229-37, 2013.

ABSTRACT AIBG 4.pdf

18° Congresso Associazione Italiana di Biologia e Genetica Generale e Molecolare, 21 Settembre 2018. In stampa.

Gene expression changes during cancer progression in cervical neoplastic keratinocyte.

Carmen Lanzillotti, Ilaria Bononi, Elena Torreggiani, John Charles Rotondo, Marika Rossini, Maria Rosa Iaquinta, Chiara Mazziotta, Mauro Tognon, Fernanda Martini.

Gene expression changes occurring in cervical intraepithelial neoplasia (CIN) progression are poorly understood. Using microarray analyses, large-scale gene expression profile was carried out in cervical neoplastic keratinocytes naturally infected by HPV-16, CIN2 and CIN3 keratinocytes, and normal cervical keratinocytes derived from normal cervical tissues belonging to the same CIN affected patients (1). Comparative analyses of differentially expressed genes identified 37 candidate genes progressively up- or down-expressed from CIN2 to CIN3 keratinocytes. One of these genes, the phosphoglycerate dehydrogenase (PHGDH), was selected for further characterization. Quantitative reverse transcription-polymerase chain reaction and immunohistochemical analysis confirmed that expression of PHGDH consistently increases during progression of CIN toward cancer (2,3). In conclusion, this study revealed 37 downexpressed or overexpressed genes which may contribute to CIN progression. In addition, protein expression of PHGDH increased from CIN1 to cancer according to the degree of malignant transformation. Thus, PHGDH likely plays an important role in the initiation and progression of cervical tumorigenesis and may be a prognostic marker for progression of CIN to invasive cancer.

References

1. Bononi et al. J. Cell. Physiol. 227:3787-95, 2012

2. Rotondo et al., J.Cell. Physiol. 230:806-12, 2015

3. Rotondo et al. Clin. Cancer Res. 23:3929-34, 2017.

ABSTRACT AIBG 5.pdf

18° Congresso Associazione Italiana di Biologia e Genetica Generale e Molecolare, 21 Settembre 2018. In stampa.

Hypermethylation-induced inactivation of IRF6 and RARβ genes in vulvar squamous cell carcinoma associated with lichen sclerosus.

Fernanda Martini, John Charles Rotondo, Lucia Oton-Gonzalez, Antonella Santoro, Maria Rosa Iaquinta, Carmen Lanzillotti, Chiara Mazziotta, Elisa Mazzoni, Ilaria Bononi, Mauro Tognon.

Vulvar squamous cell carcinoma (VSCC) represents 5% of gynecological malignancies. About 80% of VSCCs arises from an inflammatory dermatosis, lichen sclerosus (LS) (1). The molecular alterations involved in onset/progression of LS -associated VSCC are unknown. IRF6 and RARβ tumor-suppressor genes are downregulated by promoter methylation in cancer. In our investigation we aimed to evaluate the possible involvement of the IRF6 and RARβ in the development of VSCC from LS (2). We analyzed the IRF6 and RARβ mRNA expressions by quantitative PCR and the promoter methylations by sequencing of PCR-amplified bisulfite-treated DNA (3), in VSCC (n=20) and the corresponding adjacent LS (n=20), cancer-free LS (cfLS, n=20) and normal skin (n=20). mRNA expression of p63 and c-jun, IRF6 and RARβ pathway-related genes, respectively, was also investigated. IRF6 was down-regulated in progression from cfLS, LS, VSCC, and p63 was over-expressed in progression from cfLS, LS, VSCC. IRF6 promoter was hypermethylated in 10% cfLS, 45% LS, and 80% VSCC. In VSCC,

RARβ was downregulated and c-jun overexpressed. RARβ promoter was hypermethylated in 90% VSCC, 55% cfLS, 50% LS and 25% normal skin. Our data indicate hypermethylation-induced IRF6 down-expression is involved in development of VSCC from LS, whereas hypermethylation-induced RARβ down-expression is a late event in LS-associated VSCC. IRF6 and RARβ promoter methylation may be used as prognostic biomarker in clinical management of LS and LS-associated VSCC patients.

References

1. Rotondo J.C. et al. JAMA Dermatol. 152:928-33, 2016.

2. Rotondo J.C. et al. JAMA Dermatol. 154:1-5, 2018.

3. Rotondo J.C. et al. Epigenetics 8:990-7. 2013.

ABSTRACT AIBG 6.pdf

18° Congresso Associazione Italiana di Biologia e Genetica Generale e Molecolare, 21 Settembre 2018. In stampa.

Antibodies against Simian virus 40 large T antigen, the viral oncoprotein, in sera from osteosarcoma patients.

Chiara Mazziotta, Elisa Mazzoni, Maria Rosa Iaquinta, Ilaria Bononi, Francesca Frontini, Carmen Lanzillotti, Mauro Tognon and Fernanda Martini.

Human osteosarcoma (OS) is a rare human cancer, mostly occurring in children and adolescents (1). Simian virus 40 (SV40) sequences have been detected in different human cancers, including osteosarcoma. SV40 is an oncogenic virus in vivo, whereas it transforms different kinds of mammalian cells, as well as distinct human cell types. SV40 injected in rodents induces tumors of different histotypes, such as brain and bone tumors. Herein, the association between OS and SV40 large T antigen (Tag) was studied by employing indirect ELISAs using synthetic peptides that mimic different epitopes of the SV40 Tag, the viral oncoprotein. Indirect ELISAs were used to detect serum IgG antibodies against this oncogenic virus in samples from OS patients (2-3). Controls were sera from healthy subjects (HS) and oncological patients affect by breast cancer, a tumor which is not associated with SV40. It turned out that sera of OS patients had a higher prevalence of SV40 Tag antibodies, 35%, compared to HS, 20% and BC, 19%, respectively. The different prevalence of SV40 Tag antibodies revealed in OS vs HS and vs BC is statistically significant with $P<0.05$ and $P<0.01$, respectively (3). Our immunological data suggest a significantly higher prevalence of IgG antibodies against SV40 Tag epitopes in serum samples from OS patients compared to HS and BC, the controls. These results suggest an association between OS and SV40 Tag, indicating that this oncogenic virus may be a cofactor in the OS onset/progression.

References

1. Hattinger C.M. et al. Expert Opin. Emerg. Drugs 20:495-514, 2015.

2. Mazzoni E. et al. Cancer 121:708-15, 2015.

3. Mazzoni E.et al. Front. Cell Develop. Biology, June 2018.

ABSTRACT AIBG 7.pdf

18° Congresso Associazione Italiana di Biologia e Genetica Generale e Molecolare, 21 Settembre 2018. In stampa.

Chronic lymphocytic leukemia tested positive for the oncogenic Merkel cell polyomavirus, MCC-350 strain.

Elisa Mazzoni, Maria Rosa Iaquinta, Chiara Mazziotta, Carmen Lanzillotti, John Charles Rotondo, Ilaria Bononi, Fernanda Martini and Mauro Tognon.

Merkel cell carcinoma (MCC) have been found to be associated with the oncogenic Merkel cell polyomavirus (MCPyV) (1). However, MCPyV sequences have been detected at low prevalence in buffy coats (2) and sera of blood donors (3). Chronic lymphocytic leukemia (CLL) were also found associated with MCPyV by some investigators, whereas other studies did not confirm this result. In our investigation, DNA sequences belonging to MCPyV were identified with a different prevalence in sera from CLL patients and blood donors. MCPyV sequences in sera of CLL patients and blood donors had a prevalence of 7% (16/224) and 5% (14/284), respectively. Specifically, MCPyV DNA sequences, coding for the viral oncoprotein large T antigen (LT), analyzed by the droplet digital polymerase chain reaction (ddPCR) method and DNA sequencing, showed the circulation of two different MCPyV strains, MCC350 and MKL-1. Indeed, DNA sequencing performed in MCPyV-positive sera indicated that MCPyV LT sequences belong to the ubiquitous MKL-1 (1-2) and oncogenic MCC350 strains. Interestingly, the more oncogenic MCC-350 strain was present at higher prevalence, 81% (13/16), in CLL samples, while the prevalence of MCC-350 was only 21% (3/14) in sera of blood donors ($P<0.05$). It is worth recalling that MCPyV MCC350 was the strain originally identified in MCC, a rare skin cancer. In our study, this oncogenic strain is found to be more prevalent in CLL patients, whereas the ubiquitous MKL-1 is more prevalent in blood donors. These data suggest that the MCC 350 strain could be responsible of the CLL onset in a fraction of these patients, whereas the MKL-1 strain seems to be the MCPyV circulating in normal subjects.

References

1. Rotondo J.C. et al. Clin. Cancer Res. 23:3929-34, 2017.

2. Pancaldi C. et al. Blood 117:7099-101, 2011.

3. Mazzoni E. et al. Front. Oncol. 7 294, 1-7, 2017.

ABSTRACT AIBG 8.pdf

18° Congresso Associazione Italiana di Biologia e Genetica Generale e Molecolare, 21 Settembre 2018. In stampa.

Metformin inhibits malignant pleural mesothelioma cell proliferation and induces apoptosis by targeting NOTCH-1.

Marika Rossini, Elena Torreggiani, Lucia Oton-Gonzalez, Francesca Frontini, Maria Rosa Iaquinta, John Charles Rotondo, Chiara Mazziotta, Carmen Lanzillotti, Fernanda Martini, Paola Rizzo, Mauro Tognon.

Malignant pleural mesothelioma (MPM) is an aggressive malignancy arising from the mesothelial cells lining the pleural cavity exposed to asbestos fibers (1-2). Notch signaling is an evolutionarily conserved cell pathway involved in many cellular biology processes, such as cell proliferation and apoptosis (3). Notch dysregulation has been reported in vitro, in primary MPM cells, indicating a role for Notch in mesothelial cell transformation (1). In recent reports metformin (1,1-dimethylbiguanide hydrochloride), which is a common drug used to treat patients affected by type II diabetes, has been proposed as an adjuvant to treat solid tumors. Its indirect effects allow reducing the glucose and insulin blood levels, which results in the growth regression of insulin-dependent tumors (1). In our study, the Notch1 signalling pathway, the anti-proliferative and pro-apoptotic effect of metformin in MPM cells were investigated. Specifically, protein levels of Notch-1 full length and its active form, represented by the intracellular domain (N1ICD), were investigated by western blot. Cell proliferation assay of MPM cells after metformin treatment it has been performed. Programmed cell death is part of a natural mechanism that regulates cell populations. Several therapeutic agents capable to modulate the apoptosis process are without effect in MPM. Here, the apoptosis was investigated after treatment with metformin in MPM cells. Taken together our preliminary data contribute to elucidate relevant issue of MPM, such as the identification of new therapeutic targets and biomarkers. In the mesothelioma model of study, metformin could act as an anti-proliferative and pro-apoptotic drug against MPM cells targeting Notch-1.

References

1. Rossini M. et al. Front. Oncol. 8:91, 1-15, 2018.

2. Mazzoni E. et al. Proc. Natl. Acad. Sci. U S A 109:18066-71, 2012

3. Rizzo P. et al. Oncogene 27:5124-31, 2008.

ABSTRACT AIBG 9.pdf

18° Congresso Associazione Italiana di Biologia e Genetica Generale e Molecolare, 21 Settembre 2018. In stampa.

Merkel cell carcinoma development in patients affected by autoimmune diseases treated with biological drugs.

John Charles Rotondo, Elisa Mazzoni, Lucia Oton-Gonzalez, Chiara Mazziotta, Carmen Lanzillotti, Maria Rosa Iaquinta, Fernanda Martini, Mauro Tognon.

Merkel cell carcinoma (MCC) is a rare but aggressive tumor with an incidence of 1/~3 million/yr in Europe and USA. The oncogenic Merkel cell polyomavirus (MCPyV) is its causative agent. In immunocompromised patients the anti-viral/cancer response is an adverse event, which may occur due to the therapies with biologics. In our investigation, three MCC arisen in patients (n=750) affected by autoimmune diseases and treated with biologics were characterized (1). MCPyV DNA sequences were studied using PCR methods in MCCs and in peripheral blood mononuclear cells (PBMCs) (2) Sera from patients were analyzed for the presence and titer of antibodies against the oncogenic Merkel cell polyomavirus (MCPyV) antigens. IgG antibodies against the viral oncoproteins large-T (LT) and small-T (ST) antigens and the viral capsid protein-1 were analyzed by ELISA. Viral antigens were recombinant LT/ST and virus-like particles, respectively. Immunohistochemical (IHC) analyses were used in MCC tissues to reveal MCPyV-LT. MCPyV DNA sequences identified showed 100% homology with the European MKL-1 strain (3). PBMCs tested MCPyV-negative. Viral DNA loads in the MCCs were in 0.1-30 copy/cell range. IgG antibodies against MCPyV LT/ST were detected in patients 1 and 3, whereas patient 2 was negative. Sera from the three patients contained IgG antibodies against MCPyV VP1. MCCs tested MCPyV LT-antigen-positive in IHC assays, with strong/nuclear LT expression. Normal tissues tested MCPyV LT-negative. We investigated three new MCCs in rheumatologic diseases-affected patients treated with biologics, including anti-TNF. A possible cause–effect relationship between pharmacologic immunosuppressive treatment and MCC onset is suggested.

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ABSTRACT AIBG 10.pdf

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Association between uveal melanoma and the oncogenic polyomavirus BK.

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The uveal melanoma (UM) is the most common human intraocular tumor. BK polyomavirus (BKPyV) is a small DNA tumor virus, which footprints have been detected in different human cancers. In vitro BKPyV is (i) clastogenic (ii) mutagenic and (iii) transforms different animal and human cells of different histotypes, whereas in vivo induces tumors in animal models, such as mouse and hamster (1). In this investigation, the association between UM and BKPyV was analysed investigating the presence and the titer of serum IgG antibodies against this DNA tumour virus. Serum samples were from UM affected patients and controls, represented by healthy subjects (HS) with the same mean age. Sera were analysed by indirect ELISAs employing two synthetic peptides as mimotopes/antigens belonging to the BKPyV viral capsid protein 1 (VP1) (2). It turned out that serum samples from UM patients had a higher prevalence of BKPyV antibodies, 85% (51/60), compared to controls represented by two different groups of HS1, 62% (54/87) and HS2, 57% (68/120). The different prevalence of BKPyV antibodies detected in UM vs two control groups, HS1 and HS2, is statistically significant ($P<0.005$). Our immunologic data suggest a significant higher prevalence of antibodies against BKPyV VP1 epitopes in serum samples from UM patients compared to HS. These results indicate an association between UM and BKPyV, suggesting that this small DNA tumor virus may be a cofactor in the UM onset/progression (3).

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Immunological evidence of a strong association between non-Hodgkin lymphoma and Simian virus 40 large T antigen, the viral oncoprotein.

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Association between non-Hodgkin lymphoma and Simian virus 40 (SV40) has been reported (1). Herein, a new indirect ELISA was employed with two mimotopes from SV40 large T antigen (Tag), the viral oncoprotein, to analyse for specific reactions to antibodies in sera from non-Hodgkin lymphoma patients and controls, represented by healthy subjects (HS) and breast carcinoma (BC) patients (2). This study allowed us to assay a new sera collection from non-Hodgkin lymphoma patients (NHL, n = 254). To verify the association between NHL and SV40 Tag, two totally independent cohorts were analysed: NHL1 n = 150 and NHL2 n = 104. The epidemiological survey included sera from HS1, n = 150; HS2, n = 104 and BC, n = 78. This new indirect ELISA revealed that antibodies against SV40 Tag mimotopes are detectable in NHL1 and NHL2 sera with a prevalence of 37 and 36%, respectively. The prevalence of SV40-antibodies detected in both NHL1 and NHL2 cohorts differs statistically from controls, at 19% for HS1 ($p < 0.01$), HS2 ($p < 0.05$) and BC patients ($p < 0.05$). This study, carried out with an immunological assay with specific Tag oncoprotein mimotopes of Simian virus 40 (2), reports the presence of IgG antibodies against the large Tumour antigen in non-Hodgkin lymphomas for the first time. Our immunological data with two independent NHL cohorts show a statistically significant association between Simian virus 40 Tag and non-Hodgkin lymphoma (3). These results suggest that SV40-positive non-Hodgkin lymphomas could be treated differently from those tested SV40-negative.

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Innovative protocol for long-term culture of human primary keratinocytes from the normal colorectal mucosa.

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Until now, procedures for in vitro culturing of human primary keratinocytes from normal colon mucosa specimens were not fully feasible. Currently, the main methods exploit to set up primary human colon epithelial cell cultures, foresee the use of replication-inactivated murine 3T3 embryonic fibroblasts as feeder cells, as well as the employment of digestive enzymes, e.g. collagenase, dispase, trypsin-EDTA or thermolysin, to dissociate colonic mucosal cells (1). However, these methods are tricky and long-lasting. Moreover, some treatments are not effective in yielding a sufficient amount of cells and give rise to only short term cell cultures, which are often contaminated with fibroblasts (2). Our approach, described herein, allows primary keratinocytes from small tissue fragments of colorectal mucosa biopsies to grow in vitro. The procedure was set up and developed in three steps: (i) the enzymatic digestion of the tissue biopsy; (ii) the use of cloning rings to purify primary keratinocyte colonies, (iii) a defined keratinocyte medium to grow these cells in long-term culture. Our cultural method enables normal primary keratinocytes to be obtained by simple and rapid techniques (3). In our culture condition, primary keratinocytes express specific epithelial markers. Colorectal mucosa keratinocyte colonies require approximately two weeks to grow. Compared to previous approaches, our protocol provides a valuable model of study for human primary keratinocytes from colorectal mucosa both at the cellular and molecular levels. In addition, normal keratinocytes grown employing our protocol will be very useful to investigate comparatively different colorectal pathologies, including the colorectal carcinoma.

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Human papillomavirus DNA status together with the downregulation of IRF6 and RARβ genes correlate with prognosis in head and neck cancer patients.

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Human papillomaviruses (HPVs) are detected in approximatively 40% of the head and neck squamous cell carcinomas (HNSCCs). The incidence of HPV-positive HNSCCs is rapidly increasing worldwide. The presence of HPV usually confers an improved overall- and progression-free survival, yet 30% of the HPV-positive HNSCC patients will have poor prognosis. In this study the viral DNA status, episomal, integrated or a mixture of both, was investigated in correlation to the patient's outcome, along with expression of IRF6 and RARβ, two tumor suppressor genes previously found hypermethylated in vulvar carcinomas (1-2). DNA and RNA were extracted from frozen tumor biopsies. DNA was studied for the viral presence using the GP5+/GP6+ universal primers in real-time PCR (RT-QPCR) (3); the melting temperature was used to determine the HPV genotype and the cycle threshold was used to calculate the viral load. Specific primers amplifying the E2 and E6 viral regions in RT-QPCR were used to determine the integration status. IRF6 and RARβ expression was assessed by RT-QPCR. Our preliminary data show a correlation between the viral status and the prognosis of the patients. Interestingly, HPV-positive HNSCCs tumors showed a greater IRF6 and RARβ downregulation compared to HPV-negative HNSCC and to other tumor tissues. Therefore, IRF6 and RARβ, along with the viral integration status may be used as potential prognostic factors in HNSCCs.

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