POSTERS

062
A PATIENT WITH WILLIAMS SYNDROME, JUVENILE POLYPS, AND PULMONARY AVMS

C.J. Gallione,[1] C.L. Clericuzio,[2] J.C. Fahl,[3] D.A. Marchuk[1]

[1]Department of Molecular Genetics and Microbiology, Duke University Medical Center, Durham, NC; [2]Division of Genetics/Dysmorphology; [3]Division of Gastroenterology, Department of Pediatrics, University of New Mexico, Albuquerque, NM, USA

This patient presented at age 3 with typical facial, cardiac, developmental, and behavioral features of Williams syndrome. Cytogenetic analysis showed normal 46, XY chromosomes and Fluorescence *In Situ* Hybridization (FISH) identified a deletion of the Elastin gene, confirming the clinical diagnosis. Family history was negative for Williams syndrome. At 4 years old, this patient was first hospitalized for bloody diarrhea. He underwent colonoscopy at 13 years for recurrent bloody stools. Multiple juvenile polyps were found in the ascending colon and rectum. Tissue testing for HNPCC by immunohistochemistry was negative. Persisting mild hypoxemia prompted CT angiography of the chest at 13 years which identified two pulmonary arteriovenous malformations (PAVMs). Head MRI with and without contrast at 13.5 years of age showed no evidence of cerebral AVMs. This patient has no history of epistaxis, no mucocutaneous telangiectasias, and no family history of HHT. The combination of multiple juvenile polyps and PAVMs is a common feature of JP-HHT syndrome. Sequence analysis of *SMAD4* revealed a 302G>A, W101X mutation in exon 2. This is the most proximal mutation observed to date, as well as the first reported mutation, in the MH1 domain of *SMAD4* in JP-HHT. The phenotype in this patient is due to the combination of the elastin deletion and the apparent loss of function mutation in *SMAD4*. At this point, it is unclear how this unique mutation in *SMAD4* and the deletion of *Elastin* lead to this complicated phenotype.

063
BEYOND *ENDOGLIN*, *ALK1*, AND *SMAD4*: SEARCHING FOR NEW GENES FOR HEREDITARY HEMORRHAGIC TELANGIECTASIA

C.J. Gallione,[1] D. Perera,[1] A. Burroughs,[1] J.A. Richards,[2] T.G.W. Letteboer,[3] D. Rushlow,[4] N.L. Prigoda,[4] T.P. Leedom,[1] A. Ganguly,[2] J.K. Ploos van Amstel,[3] C.J.J. Westermann,[5] R.E. Pyeritz,[6] D.A. Marchuk[1]

[1]Department of Molecular Genetics and Microbiology, Duke University Medical Center, Durham, NC, USA; [2]Genetic Diagnostic Laboratory, 3DBG-Department of Medical Genetics, University Medical Center, Utrecht, The Netherlands; [4]HHT Solutions, Toronto Western Hospital, Toronto, Canada; [5]St. Antonius Hospital, Nieuwegein, The Netherlands; [6]Departments of Medicine & Genetics, Institute for Translational Medicine and Therapeutics, University of Pennsylvania School of Medicine, Philadelphia, PA, USA

Mutations in *Endoglin*, *ALK1*, or *SMAD4* account for up to 90% of all cases of HHT or related phenotypes (JP-HHT). Mutations in poorly understood regulatory regions of any of these three genes will account for some of these remaining cases. However, linkage of two unrelated families to a region on Chromosome 5 and another unrelated family to Chromosome 7 indicate that at least two other genes are involved in the pathogenesis of HHT. We are employing both a positional cloning and candidate gene approach to searching for additional HHT genes. We have assembled a cohort of 36 HHT patients for this mutation screen. All are *ENG*/*ALK1*/*SMAD4* mutation negative by sequence analysis and *ENG*/*ALK1* mutation negative by deletion screening. In one approach, we are systematically sequencing patients in our cohort for each of the genes mapping within the HHT3 and HHT4 intervals. Our second approach assumes that there may be more HHT loci than those for which we currently have provisional genome locations. Since each of the three identified genes are members of the TGF-β superfamily signaling pathway, and recent biochemical data implicates the BMP subfamily as the specific pathway directly involved in HHT pathogenesis, we have chosen to also sequence a series of genes encoding important members of these signaling cascades. This work is currently underway, and we will report the results of our screen. The identification of additional HHT genes will aid in the elucidation of the complexities of HHT pathogenesis.

064
ROLE OF PROMOTER REGION SEQUENCE VARIATIONS IN HEREDITARY HEMORRHAGIC TELANGIECTASIA

K. Damjanovich,[1] J. McDonald,[1,2] F. Gedge,[1] P. Bayrak-Toydemir[1,3]

[1]ARUP Institute of Clinical and Experimental Pathology; [2]Radiology Department, University of Utah; [3]Pathology Department, University of Utah, Salt Lake City, UT, USA

Mutations in the activin A receptor type II-like 1 (*ACVRL1*) and endoglin (*ENG*) genes are known to cause Hereditary Hemorrhagic Telangiectasia (HHT). However, in about 15–20% of patients, mutation analyses of the coding regions of these genes failed to show mutations, which suggest either mutations in non-coding regions of these genes, or mutations in yet-to-be identified genes are responsible for the disease in these patients. In this study, we sequenced the regulatory regions of both *ACVRL1* and *ENG* genes. Our study group consisted of patients diagnosed with HHT according to the Curacao Criteria, in whom no causative mutation was found in the coding regions of *ACVRL1* and *ENG* genes by sequencing or deletion/duplication testing. When available we performed linkage studies for the *ACVRL1* or *ENG* regions and determined the affected gene. We sequenced approximately 9kb upstream of the *ACVRL1* which includes exon 1 and the first two introns. We sequenced two segments, 600bp each, within the 8kb upstream of the *ENG*. We also sequenced the 3' UTRs of both genes. We found previously reported and unreported variants. We will discuss the significance of each variant by comparisons to SNP databases, to our study group and by family segregation studies.

065
AN OPTIMIZED PROBABILISTIC METHOD TO IDENTIFY MUTATIONS THAT PREDISPOSE TO HEREDITARY HEMORRHAGIC TELANGIECTASIA

K.E. Vandezande

HHT Solutions Molecular Diagnostic Laboratory, Toronto Western Hospital, Canada

Mutations to (at least) three genes lead to HHT (*ENG* or *ALK-1*) and JPHT (*MADH-4*). For several reasons, to detect mutations linked to HHT (JPHT) is a complex and resource-intensive undertaking: multiple genes, diverse mutation spectrum, and variable phenotypic presentation across individuals, even within the same family. Despite its cost and complexity, routine high-sensitivity identification of mutations offers the potential of both higher quality care and lower overall costs.[1] Widespread clinical implementation of HHT molecular diagnosis requires rapid, reliable and cost-effective methods to identify mutations. Here I present a stochastic dynamic programming model meant to help a reference lab assemble, perform and analyze multiplex PCR reactions in a dynamically determined order that achieves the highest possible mutation discovery rate per unit cost (costs may be measured either by time or money). The model's utility is shown to increase with sample volume, the number of distinct molecular assays required, and the degree of correlation between assay results. Computer simulations to order the 36 different assays used in our lab ranked more than 3.7×10^{41} candidate strategies in less than two hours.

Reference

1. Cohen, JH et al. Am J Med Genet A, 2005;137:153-60

066
GENETIC TEST IN HHT: EXPERIENCE OF THE ITALIAN GROUP ON A LARGE COHORT

C. Olivieri,[1] F. Pagella,[2] E. Matti,[2] L. Lanzarini,[3] S. Ceccomancini,[3] R. Dore,[4] V. Vespro,[4] C. Reduzzi,[4] M. Grosso,[5] G. Pongiglione,[6] E. Boccardi,[7] G. Manfredi,[8] the HHT-net Italia, E. Buscarini,[8] C. Danesino[1,9]

[1]Medical Genetics, University of Pavia, Pavia; [2]ENT Unit, Fondazione IRCCS Policlinico S Matteo, Pavia; [3]Cardiology Institute, Fondazione IRCCS Policlinico S Matteo, Pavia; [4]Radiology Institute, Fondazione IRCCS Policlinico S Matteo, Pavia; [5]Radiology Unit, Santa Croce Hospital, Cuneo; [6]Cardiovascular Department, Istituto G. Gaslini Children's Hospital, Genova; [7]Interventional Neuroradiology Unit, Niguarda Hospital, Milan; [8]Gastroenterology Unit Ospedale Maggiore, Crema; [9]Genetic Counselling Service, Fondazione IRCCS Policlinico S Matteo, Pavia, Italy

763 DNA samples from HHT families (including patients and their unaffected relatives) were collected by the goups colaborating in the HHT-net. Index cases (assuming as Index case a subject completely fullfilling diagnostic criteria from each family) are 213 and a disease causing mutation has been already found in 86/110 cases in whom the screening has been completed. ACVRL1 was the gene mutated in 64/86 patients (74%), ENG in 21/86 (24%). We found 46 different mutations in ACVRL1, 9 of them were previously unreported and 18 different mutations in ENG, 8 of them not already present in the literature or in the HHT mutation database. In one of our patients, a history of colon cancer at age 34 suggested *MADH4* involvement and a new mutation in this gene was identified (Gallione). When a mutation was found in an index case all other subjects belonging to his/her (the) family were analysed using direct sequencing or endonuclease restriction digestion, and genetic counselling was provided. As a total, 177 indivuduals harboured a mutation in ACVRL1 gene, 59 in ENG, 2 in MADH4. Moreover, 179 subjects were reassured not to be carriers of their familiar mutation, avoiding unnecessary clinical investigtions. Testing in children was performed only after accurate genetic counselling of the parents. We also accepted to perform a prenatal diagnosis in a family with three members with severe clinical complications of cerebral AV malfromations.

067
FOUNDER EFFECT FOR THE ACVRL1 IN FRAME DEL H97N98 MUTATION IN ITALY. HAPLOTYPE ANALYSIS AND EPIDEMIOLOGICAL DATA

C. Olivieri,[1] F. Pagella,[2] E. Matti,[2] L. Lanzarini,[3] S. Ceccomancini,[3] R. Dore,[4] V. Vespro,[4] C. Reduzzi,[4] M. Grosso,[5] G. Pongiglione,[6] E. Boccardi,[7] G. Manfredi,[8] the HHT-net Italia, E. Buscarini,[8] C. Danesino[1,9]

[1]Medical Genetics, University of Pavia, Pavia; [2]ENT Unit, Fondazione IRCCS Policlinico S Matteo, Pavia; [3]Cardiology Institute, Fondazione IRCCS Policlinico S Matteo, Pavia; [4]Radiology Institute, Fondazione IRCCS Policlinico S Matteo, Pavia; [5]Radiology Unit, Santa Croce Hospital, Cuneo; [6]Cardiovascular Department, Istituto G. Gaslini Children's Hospital, Genova; [7]Interventional Neuroradiology Unit, Niguarda Hospital, Milan; [8]Gastroenterology Unit Ospedale Maggiore, Crema; [9]Medical Genetics, Fondazione IRCCS Policlinico S Matteo, Pavia, Italy

A *Founder Effect* for specific mutations observed with high frequency was suggested in the Netherlands Antilles (*ENG*: c.1238G>T; g.IVS1+1g>a), France (*ACVRL1*: c.1112_1113 dupG) and Denmark (*ENG*: c.360C>A). We observed in the North Italian county of Bergamo the same mutation (*ACVRL1*: c.289_294 del CACAAC) in 9 families with 37 affected subjects, from a population for the county of 1,021,700 in 2004. All the families are unrelated for at least 4 generations and come from a limited region east of the town of Bergamo. Other HHT families not carrying this mutation are known in the same area. To the best of our knowledge, this mutation has been observed only in these families, at variance with the previously quoted mutations, which have been found in different populations. The mutation we studied causes the *in frame* deletion of His 97 and Asn 98, which are conserved residues in orthologous genes. In our series of more than 120 index cases we were able to found, till now, 86 mutations. *ACVRL1* harboured 62/86 mutations and 9/62 (14.5%) were represented by the c.289_294 del CACAAC. Preliminary results suggests a founder effect for these families. After completing the study of the families by determining the ancestral haplotype carrying the mutation by microsatellite analysis, we intend to apply the formula:

$$\frac{\log (1-\delta)/(1-p_n)}{\log (1-\theta)}$$

to calculate how long ago this mutation occurred (Brusgaard *et al* (2004)). Identification of this haplotype will be relevant for epidemiological studies on HHT in the region of Bergamo.

068
CLINICAL AND MOLECULAR DIAGNOSIS OF 20 NEW HHT FAMILIES IN SIERRALLANA HOSPITAL, CANTABRIA, SPAIN

A. Fontalba, J.L. Fernández-Luna, E. García-Alegría, A. Pérez del Molino, C. Morales, M. Salcedo, A. Bustamante, J. Zarauza, A. Fernandez-L, E.M. Garrido-Martin, F. Sanz-Rodriguez, M.E. Bernabeu, C. Bernabeu, L. Botella, R. Zarrabeitia

Unidad HHT. Hospital Sierrallana & Hospital Universitario Marqués de Valdecilla; Cantabria, Spain; Centro de Investigaciones Biológicas; CSIC. Madrid; Center for Biomedical Research on Rare Diseases (CIBERER), Instituto de Salud Carlos III, Madrid, Spain

From June 2006 to January 2007, 20 new HHT different families and a total of 32 patients belonging to them, were clinically and molecularly diagnosed in the Spanish HHT unit. This unit includes the Spanish HHT reference Hospital of Sierrallana, the Medical Genetics unit of Valdccilla Hospital, placed both in northern Spain, Cantabria and the center of Investigaciones Biologicas, CSIC, Madrid (Spain). The results of the medical screening, concerning clinical features, epistaxis, telangiectasias in skin and gastro-intestinal tract, and arterio-venous malformations, in lung, brain and liver are shown. Internal arterio-venous malformations are about the same type in HHT1 and HHT2, especially common is the liver involvement in both HHT type 1 and 2. The presence of PAVM is also found as a common trait in both HHT types although especially in HHT1. Molecular diagnosis by DNA sequencing found a total of 13 mutations in *ALK1*, [R411Q, A49fs (c.144_145insG), c.1048+5G>T (intron 7), W221X, C344R, R479X, L310fs (c.921_927dup), E379K, c.1378-1G>T, s225fs/c.673_674delAG (intron 9) and R374Q], and 7 mutations in *endoglin* [Y455X, c.1686+5G>C(intrón11), Q472X, Y258fs (c.771dupC), P131L, A425G, and W390X]. Most of these mutations had not been previously described. The trend for an increased rate of HHT2 versus HHT1 is maintained in Spain, as in other mediterranean countries.

069
GENETIC MODIFICATION OF THE HEREDITARY HAEMORRHAGIC TELANGIECTASIA PHENOTYPE

T.G.W. Letteboer,[1] K.A. Harradine,[2] D. Quigley,[2] C.J.J. Westermann,[5] J.J. Mager,[5] J.K. Ploos van Amstel,[1] R.J. Snijder,[5] D. Lindhout,[1] B.E. Aouizerat,[4] R.J. Akhurst[2]

[1]DBG-Department of Medical Genetics, University Medical Center Utrecht, the Netherlands; [2]Cancer Research Institute [3]Department of Anatomy; [3,4]Institute for Human Genetics; [4]Department of Nursing, University of California at San Francisco, USA; [5]St. Antonius Hospital, Nieuwegein, The Netherlands

It is well established that considerable phenotypic variability exists between HHT1 and HHT2. However, there is also considerable intra- and interfamilial variability in both diseases. The presence of arteriovenous malformations (PAVM, CAVM, HAVM) is partly dependent on the HHT-causative mutation (*ENG versus ACVRL1/ALK1*), but also highly likely influenced by environmental factors

and/or unlinked genetic modifiers. *ENG* and *ACVRL1* both encode proteins of the TGFβ pathway, and the influence of polymorphic variation in TGFβ signalling components and TGFβ genetic modifiers (*TGFBMs*) has previously been documented for cardiovascular development, disease and cancer, in mice and humans. We examined whether variations in genes encoding components of the TGFβ signaling pathway and/or TGFβ modifier genes influence the phenotype of HHT. SNP analysis was performed using the Illumina Bead array in 736 subjects (486 HHT mutation carriers and 250 related controls). 768 SNPs were selected representing haplotypes for 74 known TGFβ signalling pathway genes and 6 genes within *TGFBM2*, 29 genes within *TGFBM3*. Patient samples were stratified according to the presence or absence of different manifestations of HHT (PAVM, CAVM, HAVM), and the data are being analyzed using a variety of analytical tools, including genetic association, parametric and nonparametric linkage analysis and sib pair analysis. Preliminary results will be available for presentation. Identification of genetic modifier genes that influence HHT severity will lead to a better understanding of disease mechanisms that could lead to novel therapies, and may result in better risk assessment for the onset of life-threatening symptoms.

070
GENOTYPE AND EPISTAXIS (ONSET AND FREQUENCY)

T. Letteboer, F. Disch, R. Kuit, J. Mager, R. Snijder, P. Zanen, K. Westermann

St. Antonius Hospital, Nieuwegein, The Netherlands

Introduction. HHT-1 and HHT-2 have a different phenotype regarding visceral AVM's and (muco-) cutaneous telangiectasia. Three small studies (Berg, Kjeldsen, Bayrak) yield conflicting results about epistaxis. This study concerns the onset and frequency of nosebleeds in a large number of patients and addresses the relation to genotype, gender and race. *Methods*. A standard questionnaire has been used in all patients at their first visit to the hospital. These were studied retrospectively. *Results*. There were 294 patients with HHT-1, 105 Afro-Caribbeans and 189 Caucasians, 123 men (42%). 173 Patients had HHT-2, all Caucasians, 72 men(42%). Patients with HHT-1 were younger (37.7 vs 44.8 years; $p<.005$). The onset of nosebleeds was significantly earlier in HHT-1($p<.001$), as well in men as in women. At age 20, 80% of patients with HHT-1 had epistaxis and only 54% of patients with HHT-2. The point-prevalence in the entire population was respectively 95% and 91%. The frequency of epistaxis increased significantly with age in men with HHT-1 ($p<.001$) and HHT-2($p<.01$), but not in women. The age-related frequency did not differ between HHT-1 and HHT-2. The onset of nosebleeds was earlier in Caucasians than in Afro-Caribbeans with HHT-1($p<.05$), but the frequency did not differ. *Comment*. Clinical diagnosis is more difficult in HHT-2 than in HHT-1 at young ages. HHT-1 has an earlier onset of epistaxis, but age-related frequency is the same as in HHT-2. Frequency of nosebleeds increases significantly with age only in men. Genotype-phenotype studies should be stratified for gender.

071
POTENTIAL EFFECTS OF *SILENT* VARIANTS IN HEREDITARY HEMORRHAGIC TELANGIECTASIA

J. McDonald,[1,2] R. Mao,[1,3] P. Bayrak-Toydemir[1,3]

[1]ARUP Institute for Clinical and Experimental Pathology; [2] Department of Radiology, University of Utah; [3]Department of Pathology, University of Utah; Salt Lake City, UT, USA

The genetic heterogeneity of HHT, with two known genes, does not explain the variable clinical symptoms and manifestations routinely seen within families. Neither *ENG* nor *ACVRL1* mutations seem to correlate with the severity of the phenotype. This wide range of clinical presentation and the lack of an obvious genotype-phenotype correlation suggest effects of genetic modifiers on the HHT phenotype. However, it is unknown if the clinical variability is due to *silent* variants in known HHT genes or due to other modifier genes. In this study we analyzed the potential effects of polymorphisms in the HHT genes by sequencing the coding regions and exon-intron boundries of *ENG* and *ACVRL1* genes. Our study group consisted of 200 HHT patients in whom mutations were found in *ENG* or *ACVRL1* and 96 healthy individuals. As expected, we found some sequence variants in both groups with similar frequencies. However, since some sequence changes were observed only in the patient group, we speculate that these variants might be associated with the variability of the clinical phenotype. In addition, via segregation analyses, we show that some previously reported mutations are not disease causing changes. We will present and discuss the significance of some variants seen in HHT patients with known mutations.

072
THE ACTIVIN RECEPTOR-LIKE KINASE 1, ALK1, AND BMPRII COOPERATE TO PHOSPHORYLATE ENDOGLIN IN RESPONSE TO BMP9

B.A. Conley,[1] M.L. Mancini,[1] T. Nicola,[2] D. Romero,[1] R.I. Koleva,[3] C.P.H. Vary[1]

[1]Center for Molecular Medicine, Maine Medical Center Research Institute, Scarborough, Maine; [2]Department of Cell Biology, University of Alabama, Birmingham, AL; [3]Department of Cancer Biology, Dana-Farber Cancer Institute, Harvard Medical School, Boston, MA, USA

In humans, mutations in TGFβ-family signaling proteins are responsible for inherited vascular diseases. Mutations in endoglin and ALK1 result in the autosomal dominant vascular dysplasia, hereditary hemorrhagic telangiectasia. In addition, genetic data suggest a functional relationship between ALK1, BMPRII, and endoglin, because mutations in BMPRII, ALK1 and less frequently, endoglin, are associated with primary pulmonary hypertension. However, the molecular mechanisms underlying these vascular diseases remain poorly characterized. We provide evidence that ALK1 and BMPRII physically associated, and that a functional consequence of this interaction was threonine phosphorylation of endoglin by ALK1. Endoglin phosphorylation by ALK1 was specifically enhanced in the presence of BMPRII. This enhancement was independent of the BMPRII kinase activity.

Additionally, BMPRII was able to rescue Smad activity by a Smad binding-defective ALK1 receptor, suggesting a novel kinase-independent role for BMPRII in ALK1 signaling. Endoglin was phosphorylated in response to ligand stimulation of particular type I and type II receptor pairs. BMP9 specifically induced endoglin phosphorylation through the ALK1/BMPRII complex, while TGFβ1 preferentially utilized the ALK5/T_RII complex. BMP9, but not BMP10 inhibited endothelial cell tube formation in collagen gels. This effect was partially rescued by endoglin, but not by a threonine phosphorylation-defective endoglin mutant. Previous studies suggest that threonine phosphorylation of endoglin plays a role in the endothelial cell response to ALK1 activation. The current study supports the view that BMP9 is a physiologic ligand for the ALK1/BMPRII complex that regulates endothelial cell organization via the phosphorylation of endoglin.

073
CHARACTERIZATION OF THE PROMOTER REGION OF ALK1, THE TARGET GENE FOR HEREDITARY HAEMORRHAGIC TELANGIECTASIA TYPE 2

E.M. Garrido-Martin,[1] A. Fernandez-L,[1] C.P. Vary,[2] L.M. Botella,[1] C. Bernabeu[1]

[1]Centro de Investigaciones Biologicas, CSIC, and Center for Biomedical Research on Rare Diseases (CIBERER), Instituto de Salud Carlos III, Madrid, Spain; [2]Center for Molecular Medicine, Maine Medical Center Research Institute, Scarborough, ME, USA

ALK1 is a type I TGF-β receptor predominantly expressed in endothelial cells. Heterozygous mutations in the gene coding for ALK1 (*ACVRL1*), give rise to HHT type 2. Since the pathogenic mechanism of HHT2 is due to ALK-1 haploinsufficiency, the study of the regulation of ALK1 gene expression is critical to correct the disease. As a first step to ascertain the factors involved in ALK1 transcriptional regulation, we report here the sequence and characterization of its promoter region. We have sequenced a 1247 bp fragment of genomic DNA containing the theoretical transcription start site and part of the first exon. This sequence was subjected to an *in silico* analysis with different algorithms, to find putative transcription factor binding sites. The 5' region of ALK1 lacks TAATA and CAAT boxes, but contains a G/C rich region with multiple consensus sequences for Sp1 transcription factor. To assess whether Sp1 was a basal transcription factor for ALK1, different promoter constructs were generated and inserted into the pGL2 reporter. Schneider cells lacking Sp1 were transiently transfected with these constructs in the absence or presence of Sp1. Without Sp1 no significant ALK1 promoter activity could be detected, whereas Sp1 cotransfection increased the promoter activity up to 15 fold. These results suggest that Sp1 is involved in the basal transcription of ALK1. Current studies in progress are addressing the identification of other transcription factors able to upregulate ALK1 gene expression, which may be used to compensate for HHT2 haploinsufficiency.

Acknowledgements: We thank Dr. Jonathan Berg for providing the ALK1 genomic fragment (Berg et al. The activin receptor-like kinase 1 gene: genomic structure and mutations in hereditary hemorrhagic telangiectasia type 2. Am J Hum Genet. 1997 61:60-7).

074
ALK1 AND ALK5-DEPENDENT TGF-β SIGNALING IN L6E9 MYOBLASTS

S. Velasco, J.M. López-Novoa, A. Rodríguez-Barbero

Instituto Reina Sofía de Investigación Nefrológica, Dpto de Fisiología y Farmacología, Universidad de Salamanca, Salamanca, Spain

Transforming growth factor-β (TGF-β) is a growth factor that regulates multiple biologic processes. TGF-β elicits cellular responses via specific type I and II receptors and their effectors, termed Smads. In most cells, TGF-β signals via TGF-β type II receptor and activin receptor-like kinase (ALK) 5 to induce Smad2/3 phosphorylation. However, it has been shown that in endothelial cells TGF-β activates two distinct types I receptor pathways, that is, ALK5-inducing Smad2/3 phosphorylation and ALK-1-promoting Smad1/5 phosphorylation. ALK1 activation stimulates cell proliferation and migration, whereas ALK5 activation inhibits these responses. Interestly, endoglin has been involved in the regulation of both pathways. In other cell types where the action of TGF-β has been also demonstrated, the TGF-β/ALK1 signaling pathway has not been investigated. To gain more insight into the molecular mechanism by which TGF-β regulates cellular function we used L6E9 rat myoblasts. We found that the ALK1/Smad1/5/Id1 pathway is present and functional in L6E9 myoblast. ALK1 was detected at the level of mRNA and protein by PCR and Western blot. TGF-β1 induced Smad1 phosphorylation and nuclear translocation determined by Western blot and immunocytofluorescence. Furthermore, TGF-β1 induced Id1 protein expression. As expected the classical TGF-β1 pathway ALK5/Smad2/3/PAI-1 is also present and functional in L6E9 myoblasts. Endoglin expression in these cells promotes TGF-β1-induced Id1 expression and reduces PA1-1 activity. Furthermore, endoglin promotes cell proliferation and reduces collagen expression. Our results indicate the presence of both signaling pathways in non endothelial cells and a pivotal role for endoglin in balance of ALK1 and ALK5 signaling.

075
ENDOGLIN-HAPLOINSUFICIENCY MODIFIES BIOLOGICAL PROPERTIES OF MURINE ENDOTHELIAL CELLS

A. Rodríguez-Barbero, M. Pericacho, M. Prieto, J.M. López-Novoa

Instituto Reina Sofía de Investigación Nefrológica, Dpto. de Fisiología y Farmacología, Universidad de Salamanca, Salamanca, Spain

Endoglin is a transmembrane protein abundantly expressed in endothelium and involved angiogenesis. The aim of the present study was to evaluate the role of endoglin on cell proliferation, migration and extracellular matrix synthesis in endothelial cells from endoglin heterozygous ($Eng^{+/-}$) mice. We generated primary cultures of mice aor-

tic endothelial cells (MAEC) from endothelial sprouts derived from aorta rings from *Eng*$^{+/-}$ C57BL/6 and wild type mice. Western blot analysis revealed that endoglin expression was reduced by 50% in *Eng*$^{+/-}$ MAEC. The effects of endoglin heterozygosity on cellular proliferation were tested by determining the increase in cell number. *Eng*$^{+/-}$ proliferated significantly slower than *Eng*$^{+/+}$ MAEC. Flow cytometry analysis revealed a greater percentage of *Eng*$^{+/-}$ cells arrested in G0/G1. Endothelial cell migration was analysed by two independent techniques. *Eng*$^{+/-}$ MAEC, in a scratch wound assay, migrated a shorter distance than *Eng*$^{+/+}$ cells. Similarly, the number of migrated cells, in a Boyden chamber, was lower in *Eng*$^{+/-}$ than *Eng*$^{+/+}$ MAEC. Collagen and fibronectin synthesis, assessed by [^{3}H]proline incorporation and Western blot respectively, was significantly greater in *Eng*$^{+/-}$ compared to *Eng*$^{+/+}$ MAEC. Given that VEGF is a central mediator of endothelial cell proliferation and migration, its basal levels were quantified by ELISA. They were significantly reduced in *Eng*$^{+/-}$ versus *Eng*$^{+/+}$ cells. The mitogenic and chemotactic effects of VEGF on endothelial cells are NO-dependent and necessitate normal eNOS expression. *Eng*$^{+/-}$ MAECs displayed significantly reduced eNOS levels relative to *Eng*$^{+/+}$ cells. These data reveal the importance of endoglin in modulating important biological properties related to angiogenesis.

076
ENDOGLIN HAPLOINSUFFICIENCY MODIFIES BIOLOGICAL PROPERTIES IN DERMAL FIBROBLASTS

M. Pericacho, M. Prieto, J.M. López-Novoa, A. Rodríguez-Barbero

Instituto Reina Sofía de Investigación Nefrológica, Dpto de Fisiología y Farmacología, Universidad de Salamanca, Salamanca, Spain

Endoglin is a TGF-β co-receptor expressed mainly on endothelial cells involved in vascular remodelling, angiogenesis and fibrosis. Mutations in the coding region of CD105 gene are associated with HHT1, a vascular disorder characterized by multisystemic vascular dysplasia. The main body of endoglin studies have been realized in endothelial cells, however endoglin is expressed in other cell types and implicates in many physiological processes. Thus, we hypothesized that endoglin expression in other cell types may have different properties. The aim of the present study was to evaluate the role of endoglin on cell proliferation and migration in dermal fibroblasts from endoglin heterozygous (Eng$^{+/-}$) mice. We generated primary cultures of murine dermal fibroblasts, and characterized them by immunocytofluorescence by the absence of cytokeratin and the expression of structured smooth muscle actin filaments, a characteristic of miofibroblasts, activated form of fibroblasts. Western blot analysis revealed that endoglin expression was reduced by 50% in Eng$^{+/-}$ fibroblasts. The rate of cell growth, analyzed by cell cycle studies, PCNA expression and crystal violet assays, was faster in Eng$^{+/-}$ fibroblasts than in control cells. In the same way, migration stimulation was observed in Eng$^{+/-}$ cells measured by both, a multichannel wounding and a transwell system. In endothelial models shown by other authors, Eng$^{+/-}$ cells have deficient migration and proliferation. Whereby our data reveal the importance of endoglin in modulating proliferation and migration in this model, and could show a cell-type-dependent role of endoglin.

077
ENDOGLIN OVEREXPRESSION IN SPINDLE CARCINOMA CELL LINE SUPPRESSES TUMOROGENICITY IN SCID MICE

E. Pérez-Gómez,[1] C. Vary,[2] C. Bernabéu,[3] M. Quintanilla[1]

[1]Instituto de Investigaciones Biomédicas Alberto Sols, Consejo Superior de Investigaciones Científicas (CSIC)-Universidad Autónoma de Madrid (UAM), Madrid, Spain; [2]Center for Molecular Medicine, Maine Medical Center Research Institute, Scarborough, Maine; [3]Centro de Investigaciones Biológicas, CSIC, Madrid, Spain

The multistage skin cancer model in mice provides continuing insight on fundamental aspects of tumor evolution. In this model, the squamous to spindle cell carcinoma conversion is the latest stage in tumorigenesis, an event associated with shedding of membrane-bound endoglina (mEng) and production of soluble endoglina (sEng). Thus, intact mEng can not be detected in spindle carcinoma cell lines, such as CarC, but these cells produce sEng which is releases into the extracelular medium. In order to study the effect of mEng expression on the phenotype of spindle carcinoma cells, we have obtained CarC cell transfectants stably expressing a human L- or S-endoglin. Expression of L-endo, but not of the S-endo, reduced Smad 2, 3 signalling but did not affect the TGF-β cell proliferative response. When the cell lines were transplanted onto immunocompromised SCID mice, L-endo transfectants but not S-endo transfectants showed longer tumor latencies with respect to control cells. Furthermore, L-endo CarC tumors did not express mEng protein while the shorter mEng isoform was easily detected in S-endo CarC tumors by Western-blotting and inmunohistochemistry. These results suggest that L-endoglin is able to suppress the tumorogenicity of spindle carcinoma cells. The structural motif responsible for this effect appears to reside in the cytoplasmic tail, the only domain which is different between L- and S-isoforms.

078
ENDOGLIN HETEROZYGOUS MICE DEVELOP MORE SEVERE COLITIS THAN THEIR NORMAL LITTERMATES

M. Jerkic,[1,2] S. Suliman,[1,3] K. Paulson,[1,3] S.J. Mentzer,[4] M.A. Konerding,[5] M. Letarte[1,2,3]

[1]Molecular Structure and Function Programme, Hospital for Sick Children; [2]Heart and Stroke Lewar Center of Excellence; [3]Department of Immunology, University of Toronto, Canada; [4]Laboratory of Immunophysiology, Brigham & Women's Hospital, Boston MA; [5]Anatomisches Institut, Johannes Gutenberg-Universität Mainz, Germany

We investigated whether *Endoglin* heterozygous (*Eng*$^{+/-}$) mice, predisposed to HHT1 and telangiectasia, would be more susceptible to a model of inflammation associated

with leukocyte extravasation. We treated mice with 3% dextrane sulfate sodium (DSS) in their drinking water for 5 days, to trigger an inflammatory bowel disease. Mice were examined daily and sacrificed at either day 5, or between days 8-12 and 19-21 and the entire colon processed for histology. Corrosion cast and scanning electron microscopy (SEM) of colonic vessels were also performed on separate groups of mice at days 8-9. Both $Eng^{+/-}$ and $Eng^{+/+}$ mice developed severe colitis, peaking at days 8-10. Symptoms were worse in $Eng^{+/-}$ mice: weight loss, diarrhea and fecal blood. Histological analysis showed more leukocyte infiltration and crypt damage in $Eng^{+/-}$ mice. SEM revealed dilated colonic vessels in both groups of mice. Our studies reveal that $Eng^{+/-}$ mice develop more severe colitis than control littermates. We will characterize the infiltrates to ascertain if HHT mice display functionally impaired subsets of leukocytes. We also need to determine if the increased level of infiltration in the colon of HHT mice can be attributed to more focal structural dilations of microvessel segments (microangiectasia) that would facilitate leukocyte transmigration.

079
VASCULAR ENDOTHELIAL GROWTH FACTOR AND TRANSFORMING GROWTH FACTOR IN CHILDREN WITH HEREDITARY HEMORRHAGIC TELANGIECTASIA

P. Giordano, G.C. Del Vecchio, G. Di Bitonto, M. Altomare, E. Altavilla, A. Nigro, G.M. Lenato,[1] D. De Mattia

Dipartimento di Biomedicina dell'Età Evolutiva, [1]HHT Inter-departmental Centre, University of Bari, Italy

High levels of vascular endothelial growth factor (VEGF) and transforming growth factor (TGF) have been reported in adults affected by hereditary hemorrhagic telangiectasia (HHT). *Aim*. To estimate VEGF and TGF-β1 serum levels in children with HHT and to correlate them to the presence or absence of arteriovenous malformations (AVM) and to genetic mutations. *Materials and Methods*. We have enrolled 17 children with HHT (14 F, 3 M, median age 9 y) and 17 healthy control subjects. We have researched AVM with imaging methods and we have estimated ENG and ALK-1 with molecular analysis. VEGF and TGF-β1 serum concentrations were determined by standard ELISA. We have subdivided patients into two groups based on the presence or absence of AVM. Data were expressed as median, minimum, maximum and percentage and analysed by using test U of Mann-Whitney and Chi square test. *Results*. AVMs were present in 11 patients, in 5 AVMs were absent and in 1 were not evaluated. There was not significant difference into VEGF levels between patients (median 218 pg/mL, minimum 50 pg/mL, maximum 500 pg/mL) and controls (median 290 pg/mL, minimum 71 pg/mL, maximum 500 pg/mL) and into TGF-β1 levels between patients (median 11,2 ng/mL, minimum 10 ng/mL, maximum 20 ng/mL) and controls (median 14,6 ng/mL, minimum 7,2 ng/mL, maximum 20,8 ng/mL). Among patients serum levels of VEGF in those with AVM (median 250 pg/mL, minimum 85 pg/mL, maximum 500 pg/mL) were significantly higher than those without AVM (median 165 pg/mL, minimum 50 pg/mL, maximum 200 pg/mL). No difference for TGF-β1 levels was found in these subgroups of patients. Finally no difference was noted for VEGF and TGF-β1 levels among 6 patients with ENG and 9 patients with ALK-1; in 2 patients mutation was not note. *Conclusions*. VEGF is increased not in all children with HHT but in those with AVM.

080
EVALUATION OF IMMUNE PARAMETERS IN CHILDREN WITH HHT

E. Jirillo,[1,2,3] L. Amati,[2] GM Lenato,[3] P. Giordano,[3,4] G. Pasculli,[3,5] F. Resta,[3] Guastamacchia E.,[3] C. Sabbà[3,5]

[1]Department of Internal Medicine, Immunology and Infectious disease, [2]National Institute of Gastroenterology, Castellana Grotte, [3]HHT Interdepartmental Center, [4]Unit of Pediatrics, [5]Department of Internal Medicine and Public Health. University of Bari, Italy

Previously, our group evidenced certain immune deficit parameters in adults with HHT.[1,2] In particular, our data demonstrated the occurrence of a Th1-type response ablation and of innate immune response depression. In order to evaluate whether these deficits are congenital, we successuvely investigated immune responsiveness in children with HHT. Patients were divided into ENG-mutated, ALK1-mutated and non-HHT-epistaxis-affected subgroups. Regarding T-cell subsets, B-cells and NK-cells, frequencies ranged within normal values in all subgroups when compared to healthy age-matched controls. Moreover, T-regulatory cell frequencies did not show any significant alterations. In addition, percentages of cytokine (IL-10, TNF-β)-positive lymphocytes and monocytes were normal in all subgroups. The only statistically significant parameter involved was related to polymorphonuclear cell oxidative burst which was significantly reduced in ENG-mutated children, but not in their ALK1-mutated counterparts. Overall, these findings suggest that immune deficits in HHT are not congenital but rather acquired with age, more likely due to a more precocious immune senescence.

Reference

1. Amati L, et al. Curr Pharm Des 2006;12:1201-1208
2. Cirulli A, et al., Curr Pharm Des 2006;12:1209-1215

081
UPTAKE OF GENETIC TESTING/COUNSELLING FOR HEREDITARY HEMORRHAGIC TELANGIECTASIA (HHT) IN AT-RISK RELATIVES OF INDIVIDUALS WITH ENG AND ALK-1 MUTATIONS

R. Klatt

Division of Clinical & Metabolic Genetics, The Hospital for Sick Children, University of Toronto, Canada

Clinical genetic testing for HHT has been available since November 2003. One of the main benefits of testing, if informative in a proband, is to clarify uncertain diagnoses in at-risk relatives. Anecdotally, however, it appears that a significant number of at-risk relatives do not pursue gene-

tic counselling and testing. *Methods*. We performed a retrospective review of genetics charts of 53 at-risk individuals (22 males and 31 females) from 12 families where the proband was initially counselled and tested in the Genetics Clinic at the Hospital for Sick Children, an academic medical centre. We encouraged the uptake of genetic counselling and testing in at-risk relatives by suggesting that probands share a copy of the detailed counselling letter they received with first-degree relatives and by providing the contact information for a genetic counsellor specializing in HHT. We also received a number of referrals for at-risk relatives, mainly through the Toronto HHT Centre. We did not attempt to contact relatives directly. *Results*. Three of 22 siblings of an affected individual with a disease-causing mutation in either ENG or ALK-1 pursued genetic counselling and testing. Sixteen of 27 children of an affected person with a mutation in ENG or ALK-1 pursued genetic testing and counselling. Four individuals were more distantly related. We will also report on genetic test results and symptomology in the relatives. *Conclusions*. Uptake of genetic testing in at-risk relatives was low, particularly in siblings. More effective strategies to promote the benefits of genetic testing and counselling for at-risk relatives are needed.

082
HEREDITARY HEMORRHAGIC TELANGIECTASIA IN CHILDREN: A SINGLE CENTER STUDY

N. Bevers, J.J. Mager, W. Balemans, C.J.J. Westermann

Department of Pulmonology, St. Antonius Hospital, Nieuwegein, The Netherlands

Background. Hereditary hemorrhagic telangiectasia (HHT) affects approximately 1:5000 in the Netherlands. Diagnosis in children may be difficult, because manifestations of the disease develop with increasing age. *Objective*. This study describes general characteristics and manifestations of HHT in patients under 18 years of age. *Methods*. From the database of the national referral centre in the Netherlands we selected all individuals under the age of 18 years from 64 HHT families. We studied 198 children. *Results*. The analysis revealed 51 patients (mean age 11,6 years; 55% boys) with a genetic diagnosis. ENG mutation (HHT1) was identified in 41 children (mean age 11,5 years; 54% boys), of whom 17 had defenite HHT according to the Curaçao criteria. Nine patients with ALK-1 mutation were identified (mean age 12,7 years; 56% boys); 2 of them had a defenite clinical diagnosis. One child had a SMAD4 mutation. Overall, in 27 children (mean age 12,9 years; 41% boys) a defenite clinical diagnosis of HHT could be made. Capillairy microscopy was often helpful in establishing the criterion *telangiectases*. PAVM were detected in 21 patients and occurred more often in HHT1 than HHT2. CAVM were detected in 3 of 35 children screened for CAVM and all 3 were suffering from epilepsy. *Conclusions*. Establishing a clinical diagnosis of HHT in children is more difficult in HHT2 than HHT1. Without DNA-testing, the diagnosis would have been missed in 24 of 41 HHT1 and 7 of 9 HHT2 children. The study confirms that PAVM occur more often in HHT1.

083
DESCRIPTION AND PRELIMINARY RESULTS OF A NEW EXERCISE STRESS TEST FOR MEASURING FUNCTIONAL CAPACITY OF PATIENTS WITH DIFFUSE PULMONARY ARTERIOVENOUS MALFORMATIONS

J.J. Murphy, P. Pierucci, K. Henderson, J.T. Fahey, D. Chyun,[1] R.I. White Jr

Yale University School of Medicine; [1]Yale University School of Nursing, New Haven, CT, USA

Background. To describe a new method for functional assessment of patients with diffuse pulmonary arteriovenous malformations (PAVM), using a novel exercise stress test. *Methods & Materials*. Over the past three years, 25 cardiopulmonary exercise stress tests (EST) were performed on 17 individuals (9 pediatric & 8 adult) with diffuse PAVM. The ESTs were performed in the Pediatric Cardiology Exercise Lab using a progressive incremental protocol for a cycle ergometer. Baseline heart rate, electrocardiogram, and oxygen saturation data were obtained with the individual seated on the cycle. The exercise intensity started at 30 watts and increased by 30 watts every two minutes, while continuous heart rate and oxygen saturation data was obtained. The EST was ended when the individual's heart rate reached 90% of predicted maximum or when the individual could not continue due to leg fatigue, shortness of breath, or arrhythmia. During the recovery period, heart rate and oxygen saturation data were monitored until the heart rate and saturation were within 10% of the baseline.
Results. Exercise limitation was associated with rapid rise in heart rate and fall in oxygen saturation. No complications were associated with EST and in the patients serial testing, the results were reproducible.

Table.

	Age <18 N=9 (4 male)	*Age ≥18 N=8 (2 male)*
Baseline HR	107±12	86±12
Baseline O_2 sat	0.80±10	0.89±09
Min O_2 sat	0.78±13	0.70±14
Min to peak	5.8±1.6	8±2
Max watts	87±23	124±36
Weight percentile	0.53±27	
Height percentile	0.63±.31	

Conclusions. Cardiopulmonary exercise stress testing provides a noninvasive, functional assessment of individuals with diffuse PAVM without exposure to radiation.

084
16 DETECTOR SPIRAL COMPUTED TOMOGRAPHY IN THE PRETHERAPEUTIC EVALUATION OF HEREDITARY HEMORRHAGIC TELANGIECTASIA; ENHANCED SPATIAL RESOLUTION WITH RADIATION DOSE REDUCTION

C. Reduzzi,[1] V. Vespro,[1] C. Danesino,[2] C. Olivieri,[2] F. Pagella,[3] E. Matti,[3] L. Lanzarini,[4] S. Ceccomancini,[4] E. Buscarini,[5] M. Grosso,[6] G. Pongiglione,[7] E. Boccardi,[8] the HHT-net, R. Dore[1]

[1]Radiology Institute, Fondazione Policlinico S Matteo, Pavia; [2]Medical Genetics, University of Pavia, Pavia; [3]ENT Unit, Fondazione Policlinico S Matteo, Pavia; [4]Cardiology Institute Fondazione Policlinico S Matteo, Pavia; [5]Gastroenterology Unit Ospedale Maggiore, Crema; [6]Radiology Unit, Santa Croce Hospital, Cuneo; [7]Cardiovascular Department, Istituto G. Gaslini Children's Hospital, Genova; [8]Interventional Neuroradiology Unit, Niguarda Hospital, Milan, Italy

Screening algorithm for pulmonary arteriovenous malformations (PAVMs) in hereditary haemorrhagic telangiectasia (HHT) is based on the combined use of contrast echocardiography and anteroposterior chest radiograph, followed by chest CT if at least one test is positive. An alternative is to screen directly by chest CT. Some authors described CT techniques for evaluating PAVMs in HHT patients, with conventional, or spiral CT units, but none described scanning and post-processing techniques with 16-64 multidetector CT units. We describe protocols of 16 detector spiral CT used in the pretherapeutic evaluation of PAVMs. Main characteristics are: sub-millimetric collimation that allowed demonstration of millimetric vessels; no intravenous contrast medium; high spatial resolution in all axis with isotropic multiplanar reconstructions mainly in coronal and sagittal views that enhance topographic relations; scanning requires only one breath-hold of about 10 sec; automatic modulation of radiation doses tailored for each patients; reduction of the radiation doses to less than half than normal use didn't invalidate diagnostic value.

085
ANATOMIC LOCATION AND MORPHOLOGY OF PULMONARY ARTERIOVENOUS MALFORMATIONS IN HEREDITARY HEMORRHAGIC TELANGIECTASIA EVALUATED IN MULTIDETECTOR COMPUTED TOMOGRAPHY

C. Reduzzi,[1] L. Reduzzi,[2] V. Portugalli,[2] V. Vespro,[1] C. Danesino,[3] C. Olivieri,[3] F. Pagella,[4] E. Matti,[4] L. Lanzarini,[5] S. Ceccomancini,[5] E. Buscarini,[6] M. Grosso,[7] G. Pongiglione,[8] E. Boccardi,[9] the HHT-net, R. Dore[1]

[1]Radiology Institute, Fondazione Policlinico S Matteo, Pavia; [2]Radiology Unit, Ospedale Maggiore, Crema; [3]Medical Genetics, University of Pavia, Pavia; [4]ENT Unit, Fondazione Policlinico S Matteo, Pavia; [5]Cardiology Institute Fondazione Policlinico S Matteo, Pavia; [6]Gastroenterology Unit Ospedale Maggiore, Crema; [7]Radiology Unit, Santa Croce Hospital, Cuneo; [8]Cardiovascular Department, Istituto G. Gaslini Children's Hospital, Genova; [9]Interventional Neuroradiology Unit, Niguarda Hospital, Milan, Italy

Purposes. to verify number, size and morphological patterns of pulmonary arteriovenous malformations (PAVMs) related to anatomical location in patient affected of hereditary haemorrhagic telangiectasia (HHT) with positive contrast echocardiography test and evaluated with multidetector Computed Tomography (MDCT). *Methods*. two CT unit with 16 detectors; 0,75/1.5 mm scan collimation, without intravenous contrast medium; post-process maximum intensity projection (MIP) images in axial, coronal and sagittal view. We considered lobar and segmental location, peripheral (30 mm or less from pleural line) or central site (more than 30 mm), size of the aneurysms, diameter of the vascular pedicles. *Patients*. among subjects with clinical and genetic evidence of HHT, 56 patients had positive US test and CT evidence of PAVMs. *Results*. 123 PAVMs have been recognized; 77 in right lung, 46 in the left; 40 in upper lobes, 25 in medium lobes and lingula, 58 in lower lobes. Right upper lobe was affected more frequently (30 PAVMs) than left upper lobe (10 PAVMs). Only 11/123 PAVMs were in central location (7 in lower lobes). 40 PAVMs were less than 3 mm (21 in upper lobes, 15 in lower), 48 PAVMs more than 3 mm diameter (14 in upper lobes, 24 in lower); 34 PAVMs more than 10 mm (18 in lower lobes). *Conclusions*. Some results are unexpected on the basis of the letterature: prevalence for the right lung, lower involvement of left upper lobe, mild prevalence of the small PAVMs for upper lobes and of the medium/large PAVMs for lower lobes.

086
EMBOLIZATION OF LOCALIZED PULMONARY ARTERIOVENOUS MALFORMATIONS IN ADULTS

J.P. Pelage, T. Chinet, B. Raffesstin, J. Roume, J.H. Blondel, A. Ozanne, P. Lacombe

Pluridisciplinary Consultation, Paris HHT Center, Hopital Ambroise Paré, Boulogne, France

Aim. To demonstrate safety and efficacy of embolization to treat localized pulmonary arteriovenous malformations (PAVMs). To emphasize technical aspects of the procedure relevant to successful embolization. To define clinical indications for embolization of PAVMs. *Materials and Methods*. All consecutively seen patients treated with embolization for localized PAVMs have been enrolled in the study. Clinical presentation (dyspnea, previous history of paradoxical embolism) and characteristics of treated PAVMs (single or multiple, location, diameter of the feeding artery and angioarchitecture (simple or complex) have been recorded. Specific information including number of PAVMs, number of coils and per-procedural complications has been obtained. Clinical and imaging follow-up using mutidetector computed tomography has also been performed. *Results*. Forty-two patients (26 women and 16 men, mean age 45 y.o.) including 36 with hereditary hemorrhagic telangiectasia have been embolized. Twenty two patients (53%) presented with dyspnea and 12 (29%) had previous paradoxical embolism prior to embolization. A total of 99 PAVM (mean 2.3 per patient) have been treated using 530 coils (mean 12.6 per patient)

during 47 sessions. Sixty percent of PAVMs were localized in the lower pulmonary lobes and 81% had a simple angioarchitecture. The mean diameter of the feeding artery was 6mm. No perprocedural complication occured. During the follow-up, 2 patients (5%) presented a paradoxical embolism (stroke and brain abscess) and 5 patients only (13%) had persistent dyspnea. Using MDCT, 92% of treated PAVM were occluded. *Conclusion*. Embolization of localized PAVMs is a very effective treatment to improve dyspnea and reduce the risks of paradoxical embolism. A high technical success rate can be expected by trained interventional radiologists.

087
SCHIZENCEPHALY AND HEREDITARY HEMORRHAGIC TELANGIECTASIA–A PREVIOUSLY UNRECOGNIZED ASSOCIATION?

D.M. Goodenberger

Washington University Hereditary Hemorrhagic Telangiectasia Center, St. Louis, Missouri, USA; University of Nevada School of Medicine, Las Vegas, NV, USA

Schizencephaly is a brain development abnormality in which a gray matter-lined cleft connects the cerebral cortex with the ventricles. The pathophysiology is thought to be an abnormality of neuronal migration during fetal life, although it has also been postulated that in utero ischemic insult may be the underlying cause. A variety of associations have been noted, including drug abuse, prenatal warfarin exposure, cytomegalovirus infection, and tuberous sclerosis. Most cases appear to be sporadic, but some are familial. A subset of the familial cases appear to be caused by mutations in the homeobox gene EMX2. There are no reports of association with hereditary hemorrhagic telangiectasia (HHT). Information regarding frequency is scarce, ranging from 0.0015% to 0.08% of live births. Between 2001 and 2005, screening MRI's were performed on 149 patients in our center. 3 of 149 had schizencephaly, for a prevalence of 0.67%, which is 8.4 to 447 times that expected. The frequency of associated symptoms and complications (seizures, developmental delay, and paraparesis) were similar to those previously reported. Given that schizencephaly has a genetic basis in some patients, and that EMX2 does not account for all, we believe it is plausible that one or more of the mutations causative for HHT may cause schizencephaly, perhaps due to fetal cerebral vascular maldevelopment. Evaluation of cerebral screening data from other centers is warranted, and if our findings are confirmed, genetic correlation should follow.

088
PULMONARY ARTERIOVENOUS MALFORMATIONS, PULMONARY ARTERIAL PRESSURE, AND PAVM EMBOLISATION

C.L. Shovlin,[1,4] R.J. Davies,[1] J.S.R. Gibbs,[2,4] J.E. Jackson[3]

[1]Respiratory Medicine , [2] Cardiology, [3] Imaging, Hammersmith Hospital; [4]Cardiovascular Sciences, National Heart and Lung Institute, Imperial College Faculty of Medicine, London, UK

Introduction. Elevated pulmonary arterial pressure (PAP) is considered a relative contraindication to the treatment of pulmonary arteriovenous malformations (PAVMs) by embolisation. We aimed to examine PAP distribution and contributory factors in individuals with PAVMs, and to establish whether embolisation increases the PAP, in particular in individuals with pre-existing pulmonary hypertension. *Methods*. PAP were recorded prior to contrast injection in 143 individuals with PAVMs undergoing angiography for PAVM embolisation. 131/143 (92%) had underlying HHT. 35/143 individuals had measurements in consecutive embolisation session at mean interval 19.9 (range 2-121) months. 20/143 individuals with pre-existing elevated PAP mean had same-session pre and post embolisation measurements. *Results*. The mean PAP mean was 14.00±5.01 mmHg (range 6-40 mmHg). Of the five individuals with mean PAP >25mmHg, three had severe left ventricular disease and/or mitral valve pathology, and two had idiopathic pulmonary hypertension. PAP was significantly correlated with age ($p<0.0001$, Spearman rank; $p=0.004$, multiple regression analyses with overall $p<0.0001$, $r^2=15.89\%$). We are currently examining other potential contributory factors. Overall, there was no significant increase in PAP mean as a result of embolisation, even in patients with initial PAP mean in the upper quartile (>16 mmHg). In half of this group, embolisation led to a fall in PAP mean; the maximum rise was 4 mmHg in the same session, and 8mmHg in sequential angiography sessions 3-80 (mean 26.7) months apart. *Conclusion*. PAP increases with age in individuals with PAVMs. Significant increases as a result of PAVM embolisation were not observed in this series.

We thank the families and friends of HHT patients whose donations supported this work

089
PULMONARY ARTERIOVENOUS MALFORMATIONS ASSOCIATED WITH MIGRAINE WITH AURA

M.C. Post, R.J. Snijder,[*] J.J. Mager,[*] W. Jaarsma, W.J. Schonewille,[#] V. Thijs,[$] C.J.J. Westermann[*]

[*]Department of Cardiology, Pulmonology and [#]Neurology, St Antonius Hospital, Nieuwegein, The Netherlands; [$]Department of Neurology, University Hospital Gasthuisberg, Leuven, Belgium

Background. Recently, retrospective studies report a high prevalence of migraine in patients with PAVM. In this prospective study we evaluate the association between size and presence of PAVM and migraine. *Methods*. Two-hundred fifteen out of 228 persons (94%) who were referred to our hospital for HHT screening between 05-2004

and 11-2006 underwent a transthoracic contrast echocardiography (TTE) for detecting PAVM. All patients received a structured headache questionnaire. Two neurologists diagnosed migraine according to International Headache Society criteria. *Results*. One-hundred and ninety-nine subjects (63% female, mean age 44.7±14.2 years) completed the questionnaire. A PAVM was present in 36%. The overall prevalence of migraine was 23% (13% in male, 29% in female; p=0.01) and MA 10% (7% in male, 12% in female; p=0.25). In patients with a PAVM the prevalence of overall migraine was 21% compared to 24% in patients without PAVM (p=0.68), with no association between the size of the shunt and overall migraine. The prevalence of MA was 16% in patients with a PAVM compared to 8% in those without a PAVM (p=0.06). MA occurred in 28% in patients with a large shunt (>100 bubbles), 21% in patients with a at least a moderate shunt (>20 bubbles), and 10% in the presence of a small shunt (<20 bubbles) (p for trend is 0.005). Kappa coefficient for interobserver reliability was 0.9 ($p < 0.001$).
Conclusion: Only MA seems to be associated with the presence of PAVM, and the prevalence of MA increases with increasing size of the pulmonary shunt.

090
DIFFUSE PULMONARY ARTERIOVENOUS MALFORMATIONS IN HHT SUCCESSFULLY TREATED BY REPEATED TRANSCATHETER OCCLUSION. CASE REPORT AND LITERATURE REVIEW

M.F. Carette, A. Parrot, A. Bellocq, A. Khalil

Centre d'Accueil de la Maladie de Rendu Osler de Tenon (CAMROT); AP-HP, Tenon hospital, Paris, France

Aim. To report the case of a patient with HHT history and diffuse PAVM successfully treated by repeated TCO. *Material and Methods*. We reviewed the chart, imaging studies and biological study stressing on PaO_2, SaO_2 measurement before and during the follow-up after each TCO sessions (Table 1). *Results*. A 25-year-old woman presented at 27 weeks gestational age (GA) with sudden onset of right leg weakness during 20mn with complete regression. HHT was diagnosed on the association of epistaxis, telangiectasis, family history and furthermore still known PAVMs responsible for dyspnea. Unenhanced, chest CT and MR, showed a large right upper lobe PAVM. It was occluded by TCO (S1) to prevent stroke recurrence. After a transient improvement of dyspnea, a worsening with decreased of PaO_2 led to a cesarean section at GA of 35.5 weeks. At this time, a thoracic angio-MDCT showed multiple PAVMs and acute pulmonary embolism. In spite anticoagulation, dyspnea worsened. Two sessions of TCO were performed (S2, S3) complicated by multiple pulmonary infarcts and without improvement of PaO_2. Another evaluation, before subscription on lung transplantation list, always showing numerous smalls PAVMs, we try a new TCO (S4) occluding 12 smalls PAVMs which improved dramatically the PaO_2. So two other sessions were performed (S5, S6) leading to improve PaO_2, to stop the nasal oxygen, without pulmonary hypertension (8/1- 4 mmHg). *Conclusions*. In spite of a very bad prognosis, according to the literature review, for diffuse PAVMs, this case illustrate the possibility to treat them successfully, but with multiple sessions.

Table 1: PaO_2 and SaO_2 at Room air evolution and oxygenotherapy needed before and after each Transcatheter Occlusion (TCO) session of pulmonary arteriovenous malformation (PAVM).

TCO of 52 PAVM		*Prior occlusion*			*After occlusion*		
Session (n°)/ day.month.06 /delay in months	*Number of PAVM Occluded total lenght coils (cm)*	*PaO_2 mmHg*	*SaO_2 %*	*Nasal Oxygen (L/min)*	*PaO_2 mm Hg*	*SaO_2 %*	*Nasal Oxygen (L/min)*
S1 / 20.I / 0	1 / 141	65	93	5	78	95,5	0
S2 / 3.IV / 2.5	10 /155	51	85	13			
S3 / 10.IV / 2.5	13 / 76				43	82	10
S4 / 15.V / 4	12 / 86	43	88	10	59	91	6
S5 /12.VI / 5	8 / 63	57	89	6	68	93	3
S6 / 20.XI / 10	10 / 95	58	89	3	72	94	0

091
RESULTS AND ADVERSE EVENTS OF TRANSARTERIAL EMBOLIZATION OF PULMONARY VASCULAR MALFORMATIONS IN HEREDITARY HEMORRHAGIC TELANGIECTASIA

M. Grosso,[1] F. Pedrazzini,[1] E. Buscarini,[2] C. Olivieri,[3] C. Danesino,[3] P. Gazzaniga,[4] L. Reduzzi,[5] V. Portugalli,[5] F. Pagella,[6] G. Pongiglione,[7] E. Boccardi,[8] on behalf of HHT-NET

[1]Radiology Dept, Ospedale S Croce, Cuneo; [2]Gastroenterology Dept, Maggiore Hospital, Crema; [3]Genetic Institute, University of Pavia; [4]Cardiology Dept, [5]Radiology Dept, Maggiore Hospital, Crema; [6]ENT Institute, University of Pavia; [7]Paediatric Cardiology Dept, Ospedale Gaslini, Genova; [8]Interventional Neuroradiology Unit, Niguarda Hospital, Milan, Italy

Aim. to review results and adverse events of transarterial embolization of pulmonary vascular malformations (PAVMs) in hereditary hemorrhagic telangiectasia (HHT). *Methods*. In the context of the multidisciplinary HHT family screening protocol of HHT-NET in the period 1/2001-12/2006 28 HHT patients (14 males, mean age 50.3, range 19-75) with PAVMs were sent to our unit. Patients underwent transarterial embolization; occluding material employed were amagnetic coils or Amplatzer plug. Oxigen saturation before and after embolization procedure were recorded. All patients had enhanced multislice chest CT at 6 months and one year to judge completeness of PAVM occlusion. Appropriate clinical and CT follow-up or re-treatment was advised after the one-year check. Adverse events related to embolization , either immediate or delayed, were recorded. *Results*. 58 PAVMs were treated in 28 patients with 33 procedures. Metallic coils were used in 32 procedure, Amplatzer plug

in 1. Mean oxygen saturation before embolization was 93 (SD 4%), and after it was 97 (SD 2%). In a mean follow-up of 18.7 months (range 1-71) complete occlusion of all treated lesions without any recanalization; 3 patients underwent re-treatment for 4 newly appeared or enlarged PAVMs. All patients were discharged the day after the procedure, except for 1 patient (1/33 procedures, 3%) who had an ischemic stroke, requiring 6 further days of hospitalization. No immediate or delayed device dislocation were found. *Conclusion*. Our experience confirms effectiveness and safety of transarterial embolization for treatment of PAVMs in HHT.

092
PATIENT FORAMEN OVALE IN HEREDITARY HEMORRHAGIC TELANGIECTASIA: A PREVALENCE STUDY WITH CONTRAST ECHOCARDIOGRAPHY

P. Gazzaniga,[1] L. Lanzarini,[2] S.C. Mancini,[2] E. Buscarini,[3] G. Manfredi,[3] C. Danesino,[4] C. Olivieri,[4] L. Reduzzi,[5] R. Dore,[6] P. Blotta,[7] F. Pagella,[8] M. Grosso,[9] G. Pongiglione,[10] E. Boccardi,[11] G. Inama,[1] on behalf of HHT-NET

[1]Cardiology Dept, Ospedale Maggiore, Crema; [2]Cardiology Dept, University of Pavia; [3]Gastroenterology Dept, Crema; [4]Genetic Institute, University of Pavia; [5]Radiology Dept, Crema; [6]Radiology Dept, University of Pavia; [7]ENT Dept, Crema; [8]ENT Institute, University of Pavia; [9]Radiology Dept, Ospedale S Croce, Cuneo; [10]Paediatric Cardiology Dept, Ospedale Gaslini, Genova; [11]Interventional Neuroradiology Unit, Niguarda Hospital, Milan, Italy

Background. Contrast Echocardiography with second harmonic imaging (CE) has been shown to be a very sensitive test for the detection of PAVMs in Hereditary Hemorrhagic Telangiectasia (HHT). Patent Foramen Ovale (PFO) is a quite common finding in the healthy population and is often found in young patients with an unexplained cerebrovascular event. CE is widely employed to detect right to left shunt in patient with suspected PFO. Aim of this study was to detect the prevalence of PFO in HHT. *Methods*. In the context of our multidisciplinary HHT family screening protocol 302 consecutive subjects were screened for PAVMs with CE. A positive finding for the diagnosis of PAVMs was defined as the appearance of any bubbles in the left atrium later than 3 cardiac cycles after initial opacification of the right chambers. A PFO was diagnosed if at least one microbubble was detected in the left atrium within three cycles following the appearance of the contrast in the right atrium. Valsalva manoeuvre was not performed. All patients had multislice chest CT. *Results*. 16 patients, 5.3%, had positive CE for intracardiac right-to-left shunt; 2 of them had positive CT scan for PAVMs. *Conclusion*. In HHT families an analogous prevalence of PFO was found, during normal respiration, as compared with literature data regarding healthy subjects. Its potential misleading role in the echocardiographic screening of PAVMs will be discussed.

093
THE PREVALENCE OF HEPATIC CYSTS IN FAMILIES WITH HEREDITARY HEMORRHAGIC TELANGIECTASIA

E. Buscarini,[1] G. Manfredi,[1] C. Olivieri,[2] C. Danesino,[2] P. Gazzaniga,[3] P. Blotta,[4] P.A. Forner,[4] L. Reduzzi,[5] V. Portugalli,[5] F. Pagella,[6] M. Grosso,[7] G. Pongiglione,[8] E. Boccardi,[9] on behalf of HHT-NET

[1]Gastroenterology Dept, Maggiore Hospital, Crema; [2]Genetic Institute, University of Pavia; [3]Cardiology Dept, [4]ENT Dept, [5]Radiology Dept, Maggiore Hospital, Crema; [6]ENT Institute, University of Pavia; [7]Radiology Dept, Ospedale S Croce, Cuneo; [8]Paediatric Cardiology Dept, Ospedale Gaslini, Genova; [9]Interventional Neuroradiology Unit, Niguarda Hospital, Milan, Italy

Aim. to determine the prevalence of liver cysts in HHT families (either in non- or in affected subjects) as determined by Doppler ultrasound (US) screening, and to compare with prevalence in general population. *Methods*. In our familial screening protocol for HHT we searched in all subjects for hepatic VMs and other abnormalities including focal liver lesions; typical simple cysts with no oncology history were assessed on the basis of Doppler US as for number, and size. Abdominal US scans of 1878 cases from general non-HHT population referred to our Department during the period January to December 2006 were reviewed for the presence of hepatic cysts. Statistical associations were established by comparative analysis with prevalence of liver cysts in HHT families, in non- and in affected subjects, in affected subjects with liver VMs, with different severity grading of hepatic VMs from 0+ to 4, and in general population (χ^2 with Yates correction). *Results*. Four hundred and eigthythree subjects underwent our screening protocol; 335 proved to be affected, in 151 liver VMs were showed, 102 of them of grade 0+/2, and 49 of grade 3/4. Prevalence and characteristics of liver cysts are given in Table 1.

Table 1.

	General popul	*General HHT*	*Affected subjects*	*Non affected*	*Affected + liver*	*Liver VMs grades 0+/2*	*Liver VMs grades 3/4*
Liver cysts	85/4.5	23/4.7	18/5.37	5/3.37	8/5.2	5/4.9	3/6.1
single	72/3.8	18/3.7	14/4.1	4/2.7	5/3.31	3/2.9	2/4.0
2	8/0.4	3/0.6	2/0.5		1/0.6		1/2.0
>3	5/0.2	2/0.4	2/0.5	1/0.6	2/1.32	2/1.9	
Mean size cm	1.8	1.6	1.7	2.0	1.9	1.5	2.1

No difference proved statistically significant. *Conclusions*. Prevalence of hepatic cysts in HHT families is similar to that found in general population; liver cysts do not show a significant correlation with liver VMs.

094
MIGHT THERE BE ANY PLACE FOR SURGICAL RESECTIVE APPROACH OF INTRAHEPATIC ARTEROVENOUS MALFORMATION COMPLICATING HEREDITARY HEMORRHAGIC TELANGIECTASIA? REPORT OF A CASE

G. Orlando, C. Sabbà,[$] C. Scelzo,[*] J. Lerut,[°] G. Tisone

Transplant Unit, Surgical Clinic, S. Eugenio Hospital, Tor Vergata University of Rome, Italy; [$]Department of Internal Medicine, University Interdepartmental HHT Center, University of Bari, Italy; [°]Department of Surgery, Liver Transplant and General Surgery, Catholic University of Leuven, Belgium; [*]School of Medicine, Tor Vergata University of Rome, Italy

A 55-year old woman suffering from hereditary hemorrhagic telangiectasias (HHT) underwent a left lateral liver bisegmentectomy (segment 2 and 3) for a giant arterovenous malformation (AVM) connecting the left hepatic artery to the portal system. This lesion was responsible for progressive fatigue and invalidating effort dyspnoea. Recurrent epistaxis, diffuse telangiectasias and ecchymosis were also present. Transarterial embolization of the feeding artery was excluded for the large calibre of the vessel, whose occlusion would have entailed a high risk of biliary necrosis with sepsis. Postoperative course was uneventful and patient is doing well, three years after the operation. A comprehensive screening for HHT did not reveal any sign of recurrence of the AVM. To our best knowledge, this is the first case of hepatic-based HHT treated with liver resection. The main matter of concern is the risk of relapse. If future studies will demonstrate that the intra-hepatic relapse of AVM is the rule, we will have two options: to neglect pre-emptive surgery, as it would represent a major surgical risk factor for future transplantation; to perform surgery anyway, when patients are followed in centres having a small volume of activity where the waiting time on list is long. On the other hand, if it will be proven that the ablation of AVM is effective and is not followed by local recurrence, then the role of surgery might become prominent. This anecdotal report should promote this approach in order to define its future role in the treatment of this rare disease.

095
MANAGEMENT OF GASTROINTESTINAL BLEEDING IN CANADIAN HEREDITARY HEMORRHAGIC TELANGIECTASIA PATIENTS: A RETROSPECTIVE REVIEW

R. Saravanan, N.E. Marcon, M.E. Faughnan

Department of Medicine, Divisions of Gastroenterology and Respirology, St Michael's Hospital, University of Toronto, Canada

Background. Hereditary Haemorrhagic Telangiectasia (HHT) is associated with recurrent gastrointestinal (GI) bleeding in approximately 20% of patients. Several therapies (medical, surgical and endoscopic) have been reported in small numbers of patients, though there have been no large randomized clinical trials of these, and so effectiveness is uncertain. *Aim*. To describe the clinical presentation, management and outcomes in Canadian HHT patients with related GI bleeding. *Methods and Patients*. Retrospective study of patient demographics and clinical data retrieved from HHT Database and charts. *Results*. Complete data was available for 30 patients with HHT-related GI bleeding. Twenty-eight/30 (93%) had definite clinical HHT diagnosis. Mean age was 55yrs (range35-83yrs).There were 17/30 (57%) males. Mean hemoglobin on initial assessment was 95mg/dL (range 68-153mg/dL). Seventeen/30 (57%) had low ferritin. Twenty/30 (67%) had required blood transfusions. Patients were managed with iron replacement and transfusion as required. Endoscopic therapy (APC) was performed in 12/30 (40%) patients, 2 had colectomies, 2 had embolization of a duodenal AVM. Three patients were treated with tranexamic acid and 2 with estrogen/progesterone combination. After mean follow-up of 3.5yrs, there was one unrelated death and the mean hemoglobin was 116mg/dL(69-159 mg/dL). The only major complication of therapy was the development of ischemic small bowel in one patient on hormonal therapy. *Conclusions*. Management of HHT-related GI bleeding can be challenging. Multimodality management appears to be effective in improving anemia. Recent case reports suggest that anti-angiogeneic agents are effective in treating angiodysplasia-associated bleeding and may have a role in multimodality therapy for HHT-related GI bleeding.

096
ARE THERE MORE ABDOMINAL VASCULAR ANATOMIC VARIANTS IN PATIENTS WITH HEREDITARY HEMORRHAGIC TELANGIECTASIA?

J.A. Parra,[1] J. Bueno,[2] R. Zarrabeitia,[2] A. Pérez del Molino,[2] M.C. Fariñas,[2] M.T. Delgado[3]

[1]Hospital Universitario Marqués de Valdecilla, [2]Hospital Sierrallana, [3]Universidad de Cantabria, Cantabria, Spain

Aim. To evaluate the incidence of abdominal vascular anatomic variants in patients with HHT. *Material and Methods*. 50 patients (21 men and 29 women, mean age, 47,3 years) with 2 (one patient),3 (26 patients) and 4 (23 patients) criteria for HHT with a 2 MDCT performed to rule out vascular malformations in the abdomen. 28 out of 50 patients also had a genetic test and according to it 24 were type II and 4 type I HHT. All MDCT scans were made in arterial and portal phase with 3 mm section and 1 mm reconstruction thickness and were reviewed for two radiologists independently. *Results*. Vascular anatomic variants were observed in 35/50 (70%) patients. 22/35 (63 %) in the origin of the hepatic artery, 63 % in renal vascularization and 17 % at the portal level. The two most frequent hepatic artery anatomic variants were a left hepatic artery originated directly in the celiac trunk, and a right hepatic artery originated from AMS. Two or more right or left renal arteries were the most frequent anatomic variants in the kidney. A left portal vein originated in the right portal vein was the most frequent anatomic variant in the portal system. According to the genetic test 77 % of type II and 75 % of type I HHT present vascular anatomic variants. *Conclusions*. Abdominal vascular anatomic variants are more frequent than previously described in patients with HHT. Vascular anatomic variants are seen in both genetic types of HHT.

097
ESOPHAGO-GASTRO-DUODENAL LESIONS IN HEREDITARY HEMORRHAGIC TELANGIECTASIA

A. Pisani, M. Principi, P. Suppressa,[1] G. Pasculli,[*] A. Di Leo, A. Francavilla, C. Sabbà[*]

Department of Gastroenterology, [1]Department of Internal Medicine, Interdepartmental HHT Centre, University of Bari, Italy

Aim. to evaluate the prevalence and clinical behaviour of esophago-gastro-duodenal TAGs in a cohort of 80 HHT patients. *Methods*. Over a three-year-long period, esophago-gastro-duodenoscopy (EGDS) was performed in 80 (41 M, 39 F; mean age 49.65 yrs: range 19-75 yrs) consecutive HHT patients with definite diagnosis. The patients were screened independent of the presence of gastric symptoms or upper gastrointestinal bleeding. *Results*. EGDS was without complications in all 80 subjects, 48 of whom (60%) (Group A) had esophago-gastro-duodenal TAGs and 32 (Group B) without. All of group A had TAGs in the stomach accompanied by duodenal TAGs in 18 patients (22.5%) and esophageal TAGs in 3 patients (3.7%). Moreover, 7/48 patients had bleeding gastric TAGs during EGDS treated successfully with Argon Plasma Coagulator. An additional 9/48 patients with gastric TAGs without active bleeding at EGDS also had melena (2 also with hematemesis). These latter 16 pts (33.3%) required blood transfusions during the observation; in the 9 patients without bleeding during EGDS, probably bleeding was due to lower intestinal involvement. Regarding liver involvement in Group A, 40 pts were hepatic AVM+ and 8 pts were hepatic AVM-. Mean age was 55.7 years. In Group B, 12/32 patients (37.5%) required transfusions due to epistaxis, present in all but one patient' history. The 32 patients were: 18 hepatic AVM+ and 14 hepatic AVMs- . Mean age was 43.1 years. *Conclusions*. A strong association was found between gastric TAGs (48 pts) and hepatic AVMs (40 pts) in Group A, while in Group B, the incidence of hepatic (18 pts) was similar. Esophago-gastro-duodenal TAGs were more frequently found in older patients; in fact, Group A had a higher mean age than Group B. Thus, gastric and intestinal TAGs might develop with age.

098
PERCUTANEOUS EMBOLIZATION IN 12 PATIENTS WITH HEREDITARY HEMORRHAGIC TELANGIECTASIA AND SEVERE EPISTAXIS

C.A. de Witt, S.J. Braak, F.J.M. Disch, T.T.C. Overtoom, C.J.J. Westermann

St. Antonius Ziekenhuis, Nieuwegein, The Netherlands

Objective. Evaluation of the results of embolization in patients with hereditary hemorrhagic telangiectasia (HHT) because of sever epistaxis. A questionnaire was used about the frequency and severity of epistaxis, quality of life, haemoglobin 1 month before and 1 month/1 year after embolization. *Patients and methods*. Between November 1992 till July 2006 all patients with definite HHT and embolization were asked to participate in this retrospective study. During this period 18 patients had one or more procedures. Included were 12 patients (6 males, 6 females, aged 43–75 years, mean 55.1±8.2) with a total number of 19 embolizations. 6 Patients were excluded; one died of an indirect complication of the embolization. *Intervention*. Percutaneous transfemoral catheterization and angiography of the internal maxillary arteries. Embolization of the most distal branches with polyvinyl alcohol particles or microcoils on the site of the abnormal vessels. *Results*. In 18 embolizations the impact (daily frequency x severity in 3 grades) of epistaxis improved the first month. After one year 11 embolizations led to subjective improvement of nosebleeds. Mean haemoglobin rose from 6.3 to 7.2 mmol/L after 1 year. Quality of life did improve in 13 embolizations and was equal in 6 embolizations. *Conclusion*. The direct effect of the embolization is satisfactory, but temporarily in 58%. The indication should be made carefully, because there are possible major complications.

099
ANALYSIS OF MORPHOLOGY AND DISTRIBUTION OF NASAL TELANGIECTASIAS IN HEREDITARY HEMORRHAGIC TELANGIECTASIA PATIENTS

A. Colombo,[1] E. Matti,[1] L. Semino,[1] S. Corno,[1] G. Pongiglione,[2] M. Grosso,[3] L. Lanzarini,[4] R. Dore,[5] C. Olivieri,[6] E. Buscarini,[7] C. Danesino,[6] F. Pagella[1]

[1]Otorhinolaryngology Unit, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy; [2]Paediatric Cardiology Department, Hospital Gaslini, Genova, Italy; [3]Radiology Unit, Santa Croce Hospital, Cuneo, Italy; [4]Cardiology Unit, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy; [5]Radiology Unit, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy; [6]Genetic Institute, University of Pavia, Italy; [7]Gastroenterology Unit, Maggiore Hospital, Crema, Italy

Intranasal pattern and distribution of telangiectasias in Hereditary Hemorrhagic Telangiectasia (HHT) patients are variable. From 1996 to 2006, 161 HHT patients were evaluated at ENT unit of Fondazione IRCCS Policlinico San Matteo in Pavia. We analyzed morphology and distribution of these lesions in 65 patients with epistaxis before treatment by argon plasma coagulation. The patient group comprised 37 males and 28 females. The age ranged from 19 to 79 years (mean 52.2 years; median 54 years). All patients were diagnosed as affected by HHT according to the Curaçao criteria. No-one of the patients previously had undergone septodermoplasty. All patients underwent fiberoptic examination of nasal fossae. We classified distribution of telangiectasias defining six sites: nasal valve, anterior nasal septum, posterior nasal septum (considering as anterior limit a coronal plane tangential to the head of middle turbinates), inferior turbinates, middle turbinates, floor of nasal fossae. For every site we defined the presence or absence of telangiectasias and the pattern (punctate telangiectasias, interconnected punctate telangiectasias, large isolated telangiectasias, large interconnected telangiectasias). Only anterior nasal septum and inferior turbinates resulted affected in 100% of patients. Floor of nasal fossae, nasal valve and posterior nasal septum were the sites with reduced presence of telangiectasias. No significative difference resulted between different age groups considering morphology and distribution of telangiectasias.

100
2-YEARS FOLLOW-UP IN HEREDITARY HEMORRHAGIC TELANGIECTASIA PATIENTS TREATED BY ARGON PLASMA COAGULATION WITH LOCAL ANESTHESIA

F. Pagella,[1] A. Colombo,[1] E. Matti,[1] L. Semino,[1] G. Giourgos,[1] G. Pongiglione,[2] M. Grosso,[3] L. Lanzarini,[4] R. Dore,[5] C. Olivieri,[6] E. Buscarini,[7] C. Danesino[6]

[1]Otorhinolaryngology Unit, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy, [2]Paediatric Cardiology Department, Hospital Gaslini, Genova, Italy; [3]Radiology Unit, Santa Croce Hospital, Cuneo, Italy; [4]Cardiology Unit, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy; [5]Radiology Unit, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy; [6]Genetic Institute, University of Pavia, Italy; [7]Gastroenterology Unit, Maggiore Hospital, Crema, Italy

Epistaxis in Hereditary Hemorrhagic Telangiectasia (HHT) patients is caused by vascular dysplasia leading to telangiectasias involving nasal mucosa. Over the years, various treatment modalities for epistaxis associated with this disease have been described, but there is still no general agreement upon the best treatment. From 1996 to 2006, 115 HHT patients were treated at ENT unit of Fondazione IRCCS Policlinico San Matteo in Pavia. 91 of these patients were treated by argon plasma coagulation (APC). In early 2005 we presented preliminary results in 43 patients with diagnosed HHT according to the Curaçao criteria who underwent treatment by APC between 1997 and 2004. In that study 36 patients (83.7%) reported substantial reduction of bleeding after treatment. Duration of improvement was six months in 15 patients, from 3 to 6 months in 12 patients, and less than 3 months in 9 patients. We present our data of 2-years follow-up in those 43 patients to evaluate long term results in treatment by APC. 24 months after treatment 8 of 15 patients who reported a significative improvement at 6-month questionnaire declared a persisting reduction of bleeding. During the 24-months follow-up 7 of 43 patients requested one additional treatment by APC. 1 patients requested two additional treatments. 3 patients with epistaxis disagreed to undergo the same surgery. APC isn't a definitive treatment of epistaxis in HHT patients but can provide good short-term results reducing bleeding and need for blood transfusion. Our experience suggests that in some patients this modality of treatment can provide also good long-term results. More investigations are needed to clarify the reasons of different results between patients and the eventually predictive factors useful to better formulate indication for APC.

101
EXPERIENCE WITH YOUNG'S PROCEDURE FOR THE MANAGEMENT OF EPISTAXIS IN HEREDITARY HEMORRHAGIC TELANGIECTASIA

L.P. Johnson,[1] F. Miller,[2] T.M. Davidson[3]

[1]Division of Otolaryngology-Head and Neck Surgery, Department of Surgery, University of Utah Health Sciences Center, Salt Lake City, UT; [2]Department of Radiology, University of Utah Health Sciences Center, Salt Lake City, UT; University of California at San Diego, San Diego, CA; [3]Division of Otolaryngology-Head and Neck Surgery, University of California at San Diego, San Diego, CA, USA

Young's procedure, or surgical closure of the nostril was first described by Taylor and Young in 1961 for management of atrophic rhinitis. The procedure was first proposed as a treatment for epistaxis in hereditary hemorrhagic telangiectasia by Hosney and Innes in 1994, and by Gluckman and Portugal in that same year. This procedure has more recently been described by Valerie Lund and Howard in 1997, for management of severe epistaxis in patients with hereditary hemorrhagic telangiectasia. This presentation describes the experience using Young's procedure in four patients at the University of Utah HHT Center and in one patient at the University of California, San Diego HHT Center. The surgical technique for the procedure, as well as the patient profiles and results will be presented. The first patient underwent surgery in May, 2005 and to date, all patients have had cessation of the epistaxis and are now able to maintain a normal hematocrit without their previous transfusion dependency. Young's procedure is an effective surgical treatment for the epistaxis in hereditary hemorrhagic telangiectasia in a select group of patients in whom previous treatments have not been effective in managing the epistaxis and anemia.

102
COMBINED EMBOLIZATION AND LASER TREATMENT OF EPISTAXIS IN HHT PATIENTS

M.L. Fiorella, C. Russo, R. Fiorella

Ophtalmology and Otolaryngology Department and HHT Center, University of Bari, Italy

Severe epistaxes are common in HHT patients and several methods have been described in the literature for their control. One of the most recent treatments is selective embolization of nasal arteries; in this technique small particles are injected into the arteries in order to occlude the vascular bed as distally as possible. Endovascular embolization has been reported in the literature as being efficient, safe and eventually repeated in the case of recurrent bleeding. The aim of this study is to present the results of a combined treatment with selective embolization of nasal arteries in general anesthesia to obliterate most of the vessels followed, after 24 hours, by laser coagulation of residual telangiectases in local anesthesia. A pilot study was performed in four HHT transfusion-dependent patients affected with daily nosebleeds, and post-treatment results were evaluated by means of pre- and post-operatory haemoglobin levels and a questionnaire. All patients demonstrated a reduction in the duration and

number of episodes resulting in an increase of haemoglobin levels after the combined treatment. No negative side-effects of the treatment were observed. In conclusion, this combined technique permitted to improve patients' quality of life. Further studies are necessary to confirm this preliminary report on a limited number of cases.

103
THERAPY FOR EPISTAXIS WITH RALOXIFEN IN HEREDITARY HEMORRHAGIC TELANGIECTASIA

R. Zarrabeitia, A. Perez del Molino, C. Morales, M. Salcedo, J. Calvo, A. Fontalba, E. García-Alegría, B. Señaris, A. Fernandez-L, E.M. Garrido-Martin, F. Sanz-Rodriguez, M.E. Bernabeu, C. Bernabeu, L.M. Botella

Unidad HHT, Hospital Sierrallana & Hospital Universitario Marqués de Valdecilla, Cantabria; Centro de Investigaciones Biológicas, CSIC, Madrid; Center for Biomedical Research on Rare Diseases (CIBERER), Instituto de Salud Carlos III, Madrid, Spain

Aim. Based on previous reports about good response of epistaxis to tamoxifen, an oestrogen receptor modulator, we assessed the efficacy of raloxifen (a drug of the same group, with advantages on bone mineralization, cardiovascular and ginecological cancer prevention), in postmenopausal HHT women with osteoporosis, for epistaxis management. *Methods*. During the last year, eleven HHT postmenopausal women diagnosed of osteoporosis received daily oral treatment with 60 mg raloxifen. Effects on frequency and quantity of epistaxis (based on the scale by Sadick and cols.) and variations on haemoglobin levels were considered. *Results*. Eleven HHT women (age range 49-76), diagnosed of osteoporosis were included in the study. Two of them discontinued the treatment without justified reasons and in other patient, treatment was left after three months treatment due to hepatic enzymes elevation. Of the nine patients following the treatment, five were HHT2, three HHT1 and one without molecular diagnosis. Six of them needed oral iron supplies, before the treatment. After five months treatment on average, a decrease in frequency and quantity of epistaxis was observed in all them.(one grade down in the scale). An average raise in haemoglobin levels of 8,24% was observed. Only one patient was worse and required local ENT intervention. Preliminary studies of raloxifen results on endothelial cells show higher expression of ENG and ALK1 in raloxifen-treated compared with untreated cells. *Discussion*. Raloxifen may be useful in the management of epistaxis offering a wide spectrum of indications and benefits. Further randomised trials in HHT with selective estrogen receptor modulators should be considered.

104
QUALITY OF LIFE AND EPISTAXIS IN HHT; EFFECT OF TREATMENT

I. Bakker, F. Disch, H.J. Mager, R. Snijder, E. Tromp, K. Westermann

St. Antonius Hospital, Nieuwegein, The Netherlands

Introduction. This study investigates the relation between epistaxis and quality of life. (Quol) and the effect of treatment. *Methods*. An anonymous postal questionnaire was sent to 345 patients over 18 years with definite HHT. Patients who participated in trials for epistaxis (n=79) had been excluded. Questions about frequency and severity of nosebleeds and Quol (scale 0-10) were asked before and after treatment. *Results*. 200 patients (58%), 124 women, 47.5 years, returned the questionnaire; 33% had daily, 28% weekly, 31% monthly, 4% yearly and 5% no epistaxis. 17% had minor (few drops), 45% moderate (many drops) and 38% severe (gush) epistaxis. Quol was insufficient (grade 0-5) in 17%, moderate (6-7.5) in 17% and good (8-10) in 66%. Quol and impact (annual frequency x grade of severity) of epistaxis correlated significantly ($p<.001$). 64 (32%) patients with more severe HHT (Quol 8-10 in 43%) had an average of 7.7 local treatments. Treatment had beneficial influence on Quol ($p=.03$), and not significantly on annual frequency ($p=.21$) and impact ($p=.14$) of epistaxis. This benefit has disappeared at the time of the questionaire (mean 17.5 years later). Electrocautery (n=33), lasertreatment (n=22), dermoplasty (n=7) caused a non-significant initial improvement of Quol, annual frequency and impact of epistaxis. Significance did not improve after exclusion of bowel-HHT (n=16) and the use of anticoagulants (n=11). Also, treatment had no influence on iron-suppletion and transfusions. *Comment*. Quol and epistaxis are significantly related. Quol is affected only temporarily by treatment. Quol is high. Due to selection?

105
DENTAL CARE OF HHT PATIENTS: CLINICAL AND SURGICAL MANAGEMENT OF 78 CASES

V. De Falco[1], V. Crincoli[1], M. Scivetti[1], G. Pasculli[2], G. Davanzante[2], C. Sabbà[2], G. Favia[1]

University of Bari, Interdepartmental HHT Center; [1]Unità Operativa Complessa di Odontoiatria; [2]Dipartimento di Medicina Interna e Medicina Pubblica, Bari, Italy

Seventy-eight HHT patients recruited during the period 2003-06 from the HHT center in Bari were considered in this study. Prevention and care of dento-alveolar diseases are indicated due to the risk of systemic infective complications; osteitic foci are often the site of septic emboli which are responsible for potentially lethal intracranic infections. Forty-three patients (55%) showed carious disease, 20 (25%) periapical lesions, 24 (31%) parodontopathies and 16 (20%) dysodontiasis. Seven patients had a history of cerebral abscesses, six of whom were associated to the presence of acute and chronical dento-alveolar infections. Therefore, in our experience, we believed that surgical managment in these patients would be a safer choice rather than a doubtfully-successful conservative treatment. All patients underwent a clinical protocol including intra- and extra-oral clinical examination, dental orthopantomograms, oral hygiene protocols (scaling, root planning), care of parodontal lesions, and restorative, endodontic and surgical treatments. We confirm the relevance of dental prophylaxis and its inclusion in the routine management of HHT patients.

106
JUVENILE STROKE IN A CASE OF JUVENILE POLYPOSIS-HEREDITARY HAEMORRHAGIC TELEANGIECTASIA

S. Mazzucco, B. Bonetti, M. Ferrarini, G.M. Fabrizi, N. Rizzuto

Department of Neurological and Visual Sciences, Scetion of Clinical Neurology, University of Verona, Italy

A 28-year-old man was admitted to the hospital for acute occurrence of confusion, dysphasia and sensory disturbance at the right hand, in 2000. A brain CT demonstrated an hypodense left thalamic lesion. A subsequent brain MR disclosed an associated ipsilateral cortical frontoparietal lesion. A pituitary adenoma 1 cm of diameter was also seen. Genetic and acquired prothrombotic conditions were ruled out. A trans-esophageal echocardiogram suggested a possible extracardiac source of a right-to-left shunt. Chest TC confirmed the presence of a 3 cm diameter artero-venous fistula within the right inferior pulmonary lobe. The patient underwent percutaneous embolization of the fistula. Two years later, during a second procedure carried out in order to treat a recrudescence of the right inferior lobe fistula, a second one, previously overlooked, was discovered in the left inferior pumonary lobe and subsequently treated in a separate session. The patient was diagnosed as having a juvenile polyposis (JP) when he was eighteen after a bowel bleeding due to a benign polyp of the sigma, which was endoscopically excised. The patient's father died of colorectal cancer, like his brother; the patient had three sibs, two affected sisters and one apparently unaffected brother. The latter had had a hepileptic seizure a few years before. The patient was recently (in 2006) seen in our department for assessment of residual right-to-left shunt by means of TCD examination, which revealed a large permanent shunt. The suspect of JP-HHT was proposed and genetic testing for *SMAD 4* gene mutations is ongoing.

107
HHT FOUNDATION INTERNATIONAL: TODAY'S VISION IS TOMORROW'S REALITY

M.S. Clancy

Executive Director, HHT Foundation International

The HHT Foundation serves as the International nexus for HHT education, support and research for patients and families affected with Hereditary Hemorrhagic Telangiectaisa. The mission of the HHT Foundation International is find a cure for HHT while saving the lives and improving the well-being of individuals and families affected with HHT. To achieve this mission, the HHT Foundation International will: fund research to find better treatments and a cure; educate families and physicians about HHT so that awareness of crucial diagnosis and available treatments prevents needless disability and death; provide linkages between people affected by HHT; collaborate with multidisciplinary HHT Treatment Centers worldwide while advocating for patient access to these Centers; advocate for and support those with HHT while increasing public, private and governmental awareness of the disorder, engage the scientific and medical community so that talented individuals dedicate efforts toward advances in HHT screening, diagnosis, treatment, and research. Since 1991, the HHT Foundation has evolved into a dynamic and focused organization
that has achieved crucial milestones and an unprecedented level of accomplishments. Every one of the Foundation's activities is directed to the mission of the Foundation: Education and Support, Research and Advocacy.