International Session 8: ‘Head and neck cancer’

**IS8 – 1**

**CALRECTICULIN EXPRESSION IS REQUIRED FOR ORAL CANCER CELL PROLIFERATION AND MIGRATION AND CORRELATED WITH CLINICOPATHOLOGIC FEATURES IN ORAL SQUAMOUS CELL CARCINOMA PATIENTS**

J. Y. F. Chen
Department of Biotechnology, Kaohsiung Medical University, Kaohsiung, Taiwan

**Background:** Oral squamous cell carcinoma (OSCC) has emerged as one of the major malignant tumors of the head and neck cancers. However, little is known about the molecular mechanism behind tumorigenesis of OSCC. The aim of this study was to identify cancer-associated proteins by comparing the proteomes of OSCC and adjacent non-cancerous-matched tissues (NCMTs).

**Methods:** We used two-dimensional electrophoresis coupled with mass spectrometry to identify differentially expressed proteins. Later we carried out MTT assay and transwell-based migration assay to study proliferation and migration of calrecticulin knockdown OSCC cells, respectively. Finally, we conducted immunohistochemistry (IHC) to reveal calrecticulin expression in OSCC and NCMT sections and analyze the correlations with multiple clinicopathological parameters.

**Results:** After identification of the candidate proteins by LC/MS/MS, calrecticulin was found to be upregulated in OSCC specimens. Calrecticulin was differentially expressed in fresh tumor samples and six OSCC cell lines but not in adjacent NCMTs. Functional characterization of calrecticulin by siRNA knockdown revealed its requirement for oral cancer cell proliferation and migration. Furthermore, using oral tissue microarray and IHC, we demonstrated that the positive staining of calrecticulin in tumor specimens (99 of 103) was significantly higher than that in NCMTs (29 of 92) (P < 0.001). More importantly, we found that the intensity of calrecticulin expression was positively correlated with various clinicopathological parameters including tumor size (P = 0.0241), degree of tumor differentiation (P = 0.0007) and extracapillary spread (P = 0.01).

**Conclusions:** Together, our data suggest that calrecticulin could play a key role in oral cancer development and could be a prognostic biomarker for OSCC patients.

**IS8 – 2**

**IMPORTANCE OF HPV INFECTION AND FOXP3+ T-CELL STATUS AS PROGNOSTIC FACTORS IN TONSILAR SQUAMOUS CELL CARCINOMA**


1Departments of Oncology, 2Pathology, 3Otorhinolaryngology, 4Radiation Oncology, 5Radiology, Asian Medical Center, University of Ulsan College of Medicine, Seoul, Korea

**Background:** Human papillomavirus (HPV) status is a strong and independent favorable prognostic factor for survival in tonsilar squamous cell cancer (TSCC). However, little is known about the molecular mechanism behind HPV infection and the role of Treg status in TSCC patients. The aim of the study was to identify cancer-associated proteins by comparing the proteomes of TSCC and adjacent non-cancerous-matched tissues (NCMTs).

**Methods:** We used two-dimensional electrophoresis coupled with mass spectrometry to identify differentially expressed proteins. Later we carried out MTT assay and transwell-based migration assay to study proliferation and migration of calrecticulin knockdown TSCC cells, respectively. Finally, we conducted immunohistochemistry (IHC) to reveal calrecticulin expression in TSCC and NCMT sections and analyze the correlations with multiple clinicopathological parameters.

**Results:** After identification of the candidate proteins by LC/MS/MS, calrecticulin was found to be upregulated in TSCC specimens. Calrecticulin was differentially expressed in fresh tumor samples and six TSCC cell lines but not in adjacent NCMTs. Functional characterization of calrecticulin by siRNA knockdown revealed its requirement for oral cancer cell proliferation and migration. Furthermore, using oral tissue microarray and IHC, we demonstrated that the positive staining of calrecticulin in tumor specimens (99 of 103) was significantly higher than that in NCMTs (29 of 92) (P < 0.001). More importantly, we found that the intensity of calrecticulin expression was positively correlated with various clinicopathological parameters including tumor size (P = 0.0241), degree of tumor differentiation (P = 0.0007) and extracapillary spread (P = 0.01).

**Conclusions:** Together, our data suggest that calrecticulin could play a key role in oral cancer development and could be a prognostic biomarker for OSCC patients.

**IS8 – 3**

**A RANDOMIZED, OPEN-LABEL, PHASE II STUDY OF AFATINIB VERSUS CETUXIMAB IN RECURRENT/ METASTATIC HEAD AND NECK SQUAMOUS CELL CARCINOMA (HNSCC)**


1University of Chicago Medical Center, Chicago, IL, USA, 2Centre Léon Bérard, Lyon, France, 3Centre Val d’Aurelle, Montpellier, France, 4Hospital Universitario Vall D’Hebron, Barcelona, Spain, 5Katholieke Universiteit Leuven, Leuven, Belgium, 6Université de Poitiers, CHU de Poitiers, France, 7Centre Oscar Lambret, Lille, France, 8Boehringer Ingelheim Pharmaceuticals, Inc., Ridgefield, CT, USA, 9Boehringer Ingelheim RCV GmbH & Co KG, Austria, 10Boehringer-Ingelheim, Denmark A/S, Denmark

**Background:** In this study, afatinib (A) (BIBW 2992), an oral, irreversibly ErbB Family Blocker was compared with cetuximab (C) in patients (patients) with recurrent/metastatic (R/M) HNSCC after platinum-based therapy failure.

**Methods:** Eligible patients were randomized to A 50 mg/day or C 400 mg/m2/week loading dose, then 250 mg/m2/week, until disease progression or treatment-related AEs (Stage 1 [S1]); patients could then crossover treatment arms (stage 2 [S2]). Primary end point was tumor shrinkage (maximal reduction in sum of longest diameters [SLD] of target lesions versus baseline) according to RECIST 1.0, at end of S1. Biomarkers (p16 and EGFRVIII) are also being evaluated in this study.

**Results:** One hundred twenty-four patients (median age 58.0 years, 87.1% male) were equally randomized. S1 mean tumor shrinkage (adjusted mean change [SE] versus baseline in SLD) by investigator review (IR): A, 3.86 [3.62] mm; C, 2.37 [4.97] mm (P = 0.761); and by independent central review (ICR): A, 9.88 [4.02] mm; C, 6.77 [4.14] mm (P = 0.574). IR objective response rates (ORRs) were ITT: 16.1% A versus 6.5% C (P = 0.09); evaluable patients: 19.2% A versus 7.3% C. ICR ORRs were ITT: 8.1% A versus 9.7% C (P = 0.78); evaluable patients: 9.6% versus 11.1%. Median PFS was 15.9 weeks for A versus 15.1 weeks for C (P = 0.93) by IR; 13.0 versus 15.0 weeks (P = 0.99) by ICR. At S2, 32 patients crossed from A to C from S1. For S2, disease control rate (DCR): 38.9% (A as second treatment) versus 18.8% (C) (P = 0.04); IR, 33.3% versus 18.8% by ICR. The duration of DC 20.2 versus 20.7 weeks by IR; 17.3 versus 16.6 weeks by ICR. All tumors were EGFRVIII (−) by qPCR (A 25.2, C 26.1); p16 status: A 9 (+), 25 (−); C 8 (+), 23 (−). p16 (−) tumors showed higher RR for A versus C, by IR (20.0% versus 8.7%). One p16 (+) tumor responded to A. Most common treatment related AEs in S1: diarrhea (A, n = 61; 78.7%; C, n = 60; 20.2%) and rash/acne (A: 78.7%; C: 76.6%). Patients with AEs leading to dose reduction: 25.5% A versus 3.3% C; patients with AEs leading to discontinuation: 37.7% A versus 16.7% C.

**Conclusions:** Afatinib is the first EGFR-TKI to show comparable antitumor activity to cetuximab in platinum-refractory R/M HNSCC. DC with afatinib after cetuximab failure is potentially clinically meaningful. Both afatinib and cetuximab showed characteristic safety profiles; more afatinib patients experienced diarrhea.

**IS8 – 4**

**A PHASE II TRIAL OF THE MULTI-TARGETED KINASE INHIBITOR LENVATINIB (E7080) IN ADVANCED MEDULLARY THYROID CANCER (MTC)**


1Institut Gustave-Roussy, 2Centrum Onkologii Instytutu Gliwicze, 3M.D. Anderson Cancer Center, 4Royal North Shore Hospital, 5Azienda Ospedaliera Universitaria Senese, 6John Hopkins Medical Institutes, 7H. Lee Moffit Cancer Center, 8Royal Marsden Hospital, 9The Royal Brisbane and Women’s Hospital, 10Seattle Cancer Care Alliance, 11Istituto Nazionale dei Tumori, 12Ohio State University School of Medicine, 13University of Arkansas, 14Azienda Ospedaliero Universitaria Pisana, 15University of Minnesota, 16Biomarker and Personalized Medicine CFU, Eisai Inc, 17Oncology PCU, Eisai Ltd, 18Oncology PCU, Eisai Inc.

**Background:** Lenvatinib is an oral tyrosine kinase inhibitor targeting VEGFR-1, -2, -3, FGFR1-4, FGFR2, KIT, RET, and PDGFRβ. In phase I studies of lenvatinib partial responses (PR) were observed in thyroid as well as melanoma, endometrial, and renal cancers.

**Methods:** Patients with unresectable MTC and disease progression demonstrated by RECIST during the prior 12 months were enrolled. They may have received prior VEGFR-targeted therapy and were treated with a starting dose of lenvatinib 24 mg day.
once daily in 28-day cycles until disease progression or development of unmanageable toxic effects. Primary end point was response rate (RR) by RECIST. Tumor genetic analysis and circulating cytokine and angiogenic factors (CAF) analysis were carried out.

Results: Fifty-nine patients were enrolled (med age: 52; male: 63%) and are evaluable for response. Forty-five percent of patients received a dose reduction for management of toxicity, and 22% were withdrawn from therapy due to toxicity. The most common treatment-related adverse events were proteinuria 58% (gr 3: 2%), diarrhea 56% (gr 3: 5%), hypertension 48% (gr 3: 7%), fatigue 44% (gr 3: 5%), decreased appetite 41% (gr 3: 5%), nausea 34% (gr 3: 0), and weight decreased 32% (gr 3: 3%). No gr 4 events were reported for these event categories. Confirmed PRs were observed in 21 patients (RR: 36%, 95% CI: 24–49) based on independent imaging review (IIR) and 29 patients (49%, 95% CI: 36–62) based on investigator assessment. For patients who received prior VEGFR-directed treatment (n = 26) RR = 35% (IIR); with no prior VEGFR-directed treatment (n = 33) RR = 36% (IIR). Median PFS by IRR is 9.0 months (95% CI: 7.0–11.0) (based on minimum 8 months f/u, 46 events observed). There was no clear difference in the treatment response between RET-mutant (RET-mu) and RET-wild-type (RET-wt) patients. Low baseline levels of ANG2, sTie-2, HGF and IL-8 were associated with greater tumor shrinkage and prolonged PFS, whereas high baseline levels of VEGF and sVEGFR3 were associated with greater tumor shrinkage.

Conclusions: Lenvatinib administered orally at a dose of 24 mg once daily to patients affected with this disease. This combination showed promising efficacy with acceptable toxic material. The planned sample size was 45 patients, which was calculated by SWOG two-stage attained design based on an expected %CR of 60% and a threshold of 45%.

Results: From July 2008 to July 2010, 45 eligible subjects were accrued, including 43 males, with median age 63 years, ECOG PS 0/1 (36/9), oropharynx/hypopharynx/ larynx (26/15/4), T1/T2/T3/T4a/T4b (1/11/7/7/7) and N0/N2a/N2b/N2c/N3 (2/3/ 10/24/6). %CR was 64.4% (8 CR, 21 good PR) on central review. After a median follow-up of 1.56 years, 1 year local progression-free survival was 77.8%, with 1-year progression-free survival of 70.9%, 1-year overall survival of 93.3% and 1-year time to treatment failure of 57.6%. Grade 3 or 4 toxicity included mucositis (46.7%), dysphagia (46.7%), anorexia (42.2%), radiation dermatitis (26.7%), neutropenia (26.7%) and febrile neutropenia (4.4%). No treatment-related deaths were observed.

Conclusion: This combination showed promising efficacy with acceptable toxic effects. Further investigation in a phase III trial is planned.