Chapter-2: Review of Literature

Chapter-2.1: Geographical distribution, life style and epidemiology of esophageal cancer:

Esophageal cancer has high incidence in certain parts of the world such as Northern China, Iran, South Africa, and North & North-eastern belt of India, with up to 50-fold difference between high-risk and low risk populations (Parkin, Bray et al. 2005, Akbari, Malekzadeh et al. 2006). Worldwide estimated new esophageal cancer cases reported in male is 323000 and in female it is 133000 with cumulative risk to age 75 is 1.1 and 0.4 in males and females respectively (Ferlay, Soerjomataram et al. 2015). GLOBOCAN 2012 data suggests that In India, esophageal cancer is 6^{th} leading cancer in males and 8^{th} in females (Ferlay, Soerjomataram et al. 2015). The highest incidence of cancer of the esophagus in India has been reported from Assam after Meghalaya in the North-east region, where it is the second leading cancer in men and third leading cancer in women (Chattopadhyay, Kapur et al. 2007).

Throughout the world, it has been reported that the risk of ESCC increases as socioeconomic class declines (Brown, Hoover et al. 2001)(Menvielle, Luce et al. 2005). Several case-control studies (Tuyns, Riboli et al. 1987, Gao, Wang et al. 1994, Tzonou, Lipworth et al. 1996, Launoy, Milan et al. 1998, Yang, Wang et al. 2005), and two cohort studies (Guo, Blot et al. 1994, Gonzalez, Pera et al. 2006) have shown reduced risk of esophageal cancer associated with regular intake of fruits and green and yellow vegetables. In a study from Japan, it was found that 100gram/day increase in the intake of total fruit reported 11% decreases in the risk of esophageal squamoucs cell cancer

development (Yamaji, Inoue et al. 2008).

Additionally, genetic factors may also play a role in the risk of ESCC development as evidenced by the greater prevalence of ESCC in individuals with the so-called "Mongolian phenotype" and the high risk areas of esophaeal cancer belt as depicted in figure 8 that extends from China through the Central Asian republics of the former Soviet Union to north-eastern Iran (Kamangar, Malekzadeh et al. 2007)

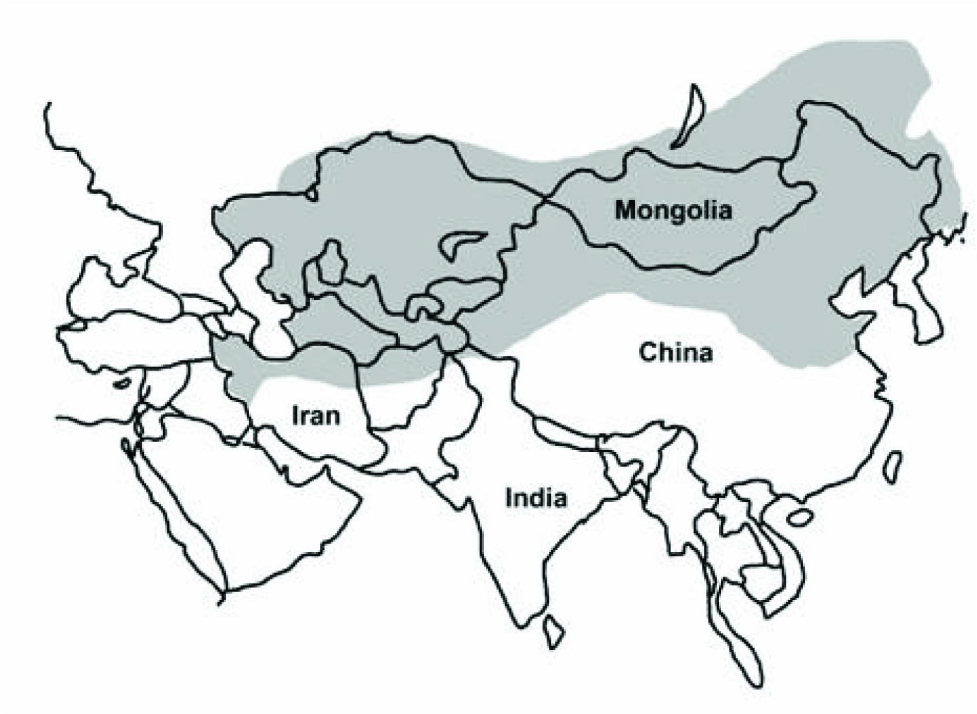


Figure 8: Esophageal cancer belt of central Asia. Image sourced from (Kamangar, Malekzadeh et al. 2007)

Tobacco smoking is a major etiologic factor for ESCC in western countries, where it increases the risk by approximately 3 – 5 folds. Drinking alcohol is quite common in these countries. Several studies have shown that a long term alcohol use may be possible cause of ESCC development in western countries (Jensen 1979, Tuyns 1983, Boffetta and

Garfinkel 1990, Brown, Hoover et al. 1994, Brown, Hoover et al. 2001, Pennathur, Gibson et al. 2013). In a recent report, it was found that among African American man who uses alcohol and tobacco, incidence of ESCC was highest (Prabhu, Obi et al. 2016). Among Asian ESCC patients, it was reported that tobacco smoking and alcohol along with dietary habits, associated with the etiology of esophageal squamous cell carcinoma (Wang, Xu et al. 2007).

In ESCC high incidence regions of Iran though all form of tobacco use independently or in combination with opium associated with the increase in the risk factor for ESCC development whereas alcohol use didn't found any significance (Nasrollahzadeh, Kamangar et al. 2008). Tobacco smoking and chewing along with alcohol in combination was found significantly associated with the development of esophageal cancer in India (Znaor, Brennan et al. 2003).

Clinical biochemical parameters like albumin, alkaline phosphatase, bilirubin, hemoglobin, RBC, WBC etc along with age at diagnosis, performance status, sex were reported to have having prognostic significance in advanced lung, gastrointestinal cancer (Proctor, Talwar et al. 2010). Albumin and C reactive protein (CRP) in combination used as Glasgow Prognostic Score (GPS) and the other biochemical parameter like c-glutamyl transferase and alkaline phosphatase were found correlated with the chronic inflammation in systemic response activity in patients with advanced lung and gastrointestinal cancer (Brown, Milroy et al. 2007) where they also reported Karnofsky Performance Status poorer in cancer patients along with low serum haemoglobin and albumin level.

Clinical biochemical parameter high lactate dehydrogenase (LDH) activity correlated

with poor response to treatment in Hodgkin's lymphoma (Smolewski, Robak et al. 2000). In esophageal carcinoma, a clinical biochemical parameter, red cell distribution width (RDW) (≧ 15%) if reported elevated at the time of initial diagnosis can predict poorer disease-free and overall survival (Wan, Chen et al. 2016). Biochemical parameter like lower alalanine transaminase (ALT) and hematological parameters like higher white blood cell count can predict response to neo adjuvant chemotherapy (NACT) in esophageal squamous cell carcinoma patients (Liu, Chen et al. 2014).

2.2 Molecular Markers in Esophageal squamous cell carcinoma

Esophageal squamous cell cancer is considered an aggressive solid tumor where response to standard treatment regime varies. For the better understanding of this disease, different molecular markers have been getting evaluated to determine their efficacy in predicting the clinical course of ESCC. There are multiple molecular markers that have been described in various literatures in context of predicting prognosis or treatment response like cancer stem cell marker aldehyde dehydrogenase 1 (ALDH1) (Minato, Yamamoto et al. 2013, Ji, Li et al. 2016), epidermal growth factor receptor like HER2 (Gibson, Abraham et al. 2003), and surrogate marker for Human Papilloma Virus (HPV) $p16^{INK4A}$ (Taghavi, Biramijamal et al. 2010).

Despite the enormous advancement in the surgical and treatment strategies to manage ESCC, the 5 year survival rate that is nearly 14-28% remains low and has not yet improved remarkably from the dismally low 5 year survival rate of 4% in the 1970s (Enzinger and Mayer 2003, Tew, Kelsen et al. 2005, Gebski, Burmeister et al. 2007). To overcome this, more emphasis has been given throughout the world on the understanding

of the biology behind the mechanistic failure of conventional chemotherapy.

The mortality due to ESCC cancers in India is among the highest due to late symptoms assessment and presentation of patients in clinics. Non availability of specialized cancer care in most part of India, particularly rural India also aggravates the situation. Despite adopting a multimodality approach that comprises of surgery, chemotherapy, radiotherapy, and personalized therapy for treating esophageal cancers, no significant improvement in overall survival has occurred over the past 4-5 decades. Since, five-year survival is directly related to stage at diagnosis, prevention and early detection efforts have the potential not only for decreasing the incidence, but also for improving the survival of those who develop this disease. Several new findings and concept are coming up in this direction.

There are two types of cancer models based in general. In contrast to the stochastic model, where every cancer cell has the potential to give rise to a new tumor, the cancer stem cell model suggests that only a few cells have the intrinsic capacity to generate new tumors (Balicki 2007). Conventional therapies used to treat tumors, don't target cancer stem cells and this may be the possible reason why after treatment, tumors returns. According to the cancer stem cell model, tumors originate in either tissue stem cells or progenitor cells, through deregulation of the normally tightly regulated process of self-renewal (Passegue, Jamieson et al. 2003, Molofsky, Pardal et al. 2004). However, in a recent report observed surprising finding that normal and cancer stem cells (CSC) like cells can arise de novo from more differentiated cell types and that hierarchical models of mammary stem cell biology should encompass bidirectional inter-conversions between stem and nonstem compartments which suggests a more complicated scenario than

previously thought (Chaffer, Brueckmann et al. 2011).

The hypothesis of the origin of cancer from stem cell is strengthened from the fact that stem like cells are discovered in many types of tumors. From the initial finding of CSCs in leukemia (Lapidot, Sirard et al. 1994, Bonnet and Dick 1997), and breast cancer (Al-Hajj, Wicha et al. 2003). CSCs have been prospectively isolated in numerous malignancies including: brain (Singh, Hawkins et al. 2004), colon (Ricci-Vitiani, Lombardi et al. 2007), head and neck (Prince, Sivanandan et al. 2007), pancreatic (Hermann, Huber et al. 2007, Li, Heidt et al. 2007), melanoma (Schatton, Murphy et al. 2008), mesenchymal tissue (Wu, Wei et al. 2007), liver (Yang, Ho et al. 2008), lung (Eramo, Lotti et al. 2008), prostate (Collins, Berry et al. 2005), and ovary (Curley, Therrien et al. 2009).

First isoform of aldehyde dehydrogenase (ALDH1) is a cytosolic enzyme responsible for oxidizing intracellular aldehydes, and conversion of retinol to retinoic acid in stem cells. ALDH1 has been previously demonstrated to be a marker for stem cells in breast cancer and non-small cell lung cancer (NSCLC), and a predictor of poor clinical outcome (Ginestier, Hur et al. 2007).

Polymorphisms in enzymes that control alcohol and acetaldehyde metabolism have also been associated with esophageal cancer risk (Lewis and Smith 2005, Yokoyama and Omori 2005). Thereafter, adjusting for alcohol intake, low activity polymorphisms in alcohol dehydrogenase-2 (*ADH2**1) and aldehyde dehydrogenase-2 (*ALDH2**2) have both been associated with a higher risk of esophageal cancer (Yokoyama and Omori 2003, Lewis and Smith 2005).

Over expression of HER2 (which derives its name from human epidermal growth factor receptor 2) defines one of these unique subtypes. The HER2/neu gene is a member of a family of genes encoding transmembrane receptors for growth factors, including the epidermal growth factor receptor (EGFR), HER2, HER3, and HER4 (Burstein 2005). The intracellular domain of HER2 has tyrosine kinase activity that regulates important aspects of the physiology, growth, and differentiation of cells (Yarden and Sliwkowski 2001, Cho, Mason et al. 2003). Extracellular domains of the HER2 protein interact with HER family members, allowing HER2 to serve as a co-receptor and to facilitate signal transduction as part of a heterodimer complex that forms after ligand binding. There is no known ligand for HER2 itself, however, suggesting that the primary role of HER2 is to modulate signals after ligand binding to other HER-family receptors.

Amplification of the HER2/neu oncogene and related genetic elements in the amplicon on chromosome 17 causes a marked increase (up to 100 times the usual level) in the expression of HER2 on the surface of breast-tumor cells. HER2 became clinically relevant with the demonstration that HER2-positive breast cancers have a worse prognosis than HER2- negative tumors (Slamon, Clark et al. 1987).

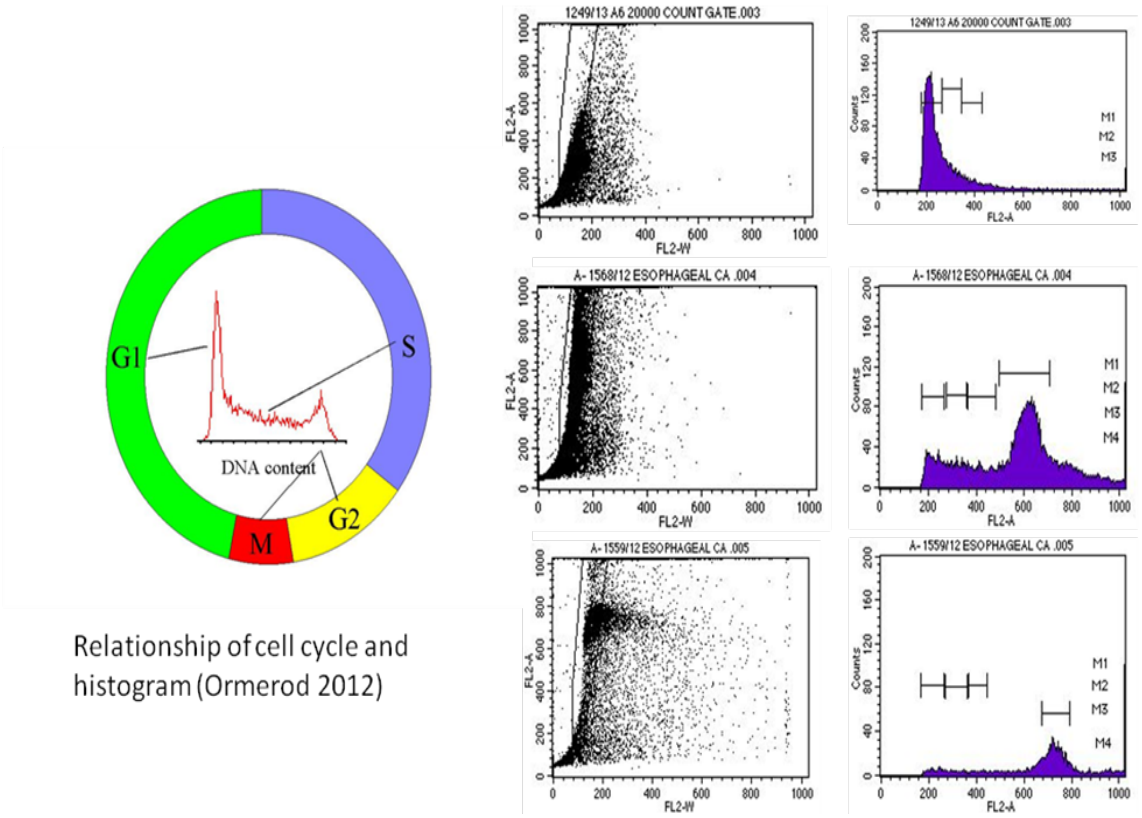
In women's having Her2-positive breast cancer, addition of 1 year adjuvant trastuzumab (a type of monoclonal antibody) significantly increases disease-free and overall survival (Slamon, Eiermann et al. 2011). Her2 molecular profiling is been explored in other cancer types like the international phase III trial known as ToGA (Trastuzumab for Gastric Cancer) aimed to determine the clinical efficacy and acceptable toxicity profile of trastuzumab in combination with first-line chemotherapy in HER2-overexpressing gastric or gastroesophageal cancer (Norguet, Dahan et al. 2012).

HPV infection has been reported to correlate with the clinical outcome in different squamous cell malignancies. p16 expression that is correlated with HPV infection was shown as marker for responder and better prognosis among head and neck squamous cell carcinoma patients who underwent radiotherapy (Lassen, Eriksen et al. 2009). Chemotherapy could be omitted in this group of patients. Similarly there is some evidence to suggest that high p16 expression correlates with favourable prognosis in esophageal squamous cell carcinoma as well (Sturm et al., 2001; Cao et al., 2014).

2.3 Cell cycle parameters in esophageal cancer:

Cancer progression commonly arises as a result of an acquired genetic instability and the subsequent evolution of that clonal populations with accumulated genetic insults. This nonlinear evolutionary cascade usually consists of multiple cell lineages with combinations of somatic genetic abnormalities. One of the most common lesions in human cancers is the development of ploidy abnormalities. Elevated 4N (G2-tetraploid) cell populations have been detected in several human neoplasias and are associated with subsequent progression in a variety of cancers (Shackney, Smith et al. 1989). Cell cycle phase analysis can predict the treatment response and prognosis in solid tumors (Lee, Endesfelder et al. 2011).

Several reports have suggested than aneuploidy and S-phase fraction can be used as prognostic factors in solid tumors and non-Hodgkin's lymphomas (Smolewski, Robak et al. 2000). S phase fraction was found indicator of poor prognosis in hodgkin's disease though DNA aneuploidy did not correlate any of the clinical outcome (Erdkamp, Breed et al. 1993). Figure 9 depicts an overview of a typical cell cycle graph.



Relationship of cell cycle and histogram (Ormerod 2012)

Figure 9. Overview of Flow Cytometer based cell cycle analysis which can provide information related to ploidy estimation and S phase fraction.