

Use Of Immunohistochemical Method To Recognize HPV In Lung Cancer Tissue

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Abstract

Lung cancer is considered one of the most common causes of death. The major reason why lung tumours are so common is due to the much-feared HPV and also smoking. The major association between lung tumor and HPV infection has been testified, some people think that it was due to geographical and racial diversity. The main aim of this research is to discover the existence of human papillomavirus in lung cancer patients by making use of the immunohistochemical method. In this particular study, a total of thirty-five (35) lung cancer patients were studied, out of which 30 were male and 5 were female patients. The ages of patients ranged from 36-96 years, with 30 of them being smokers. HPV expressed in lung cancer tissue was especially common in smoker patients. The study proved that the human papillomavirus played a substantial role as a major risk factor in lung cancer.

Introduction

Lung cancer is one of the main reasons behind deaths of both genders all across the world, and the number of these deaths is increasing day by day (Brambilla et al., 2001). This trend of the increasing rate of death is predominantly due to the fact that patients with lung cancer are discovered at a later stage of cancer development. The mortality rate can be reduced to a great extent if early detection of lung cancer can be developed somehow. The proper treatment procedure for lung cancer can only be conceivable if early detection of the cancerous cell can be achieved somehow. This is a very serious issue as the lung is a possible place of carcinoma and malignancy, and it could even further turn into a fatal disease (Mittal KR et al., 1993). Lung cancer particularly grows in two separate compartments of the lung: one is the peripheral (respiratory) part of the lung, and the other is the central (transporting air) portion of the lung.

Discussion

There are at least four major categories of cancer that have been identified, including small cell cancer, non-small cell cancer, large cell cancer, and many other types that are not that frequent. Lengthy exposure of the epithelium results in the lining of the lungs with numerous carcinogenic elements, primarily due to smoking cigarettes, which can eventually result in frequent genetic variations of the cells, which would ultimately result in numerous concerns related to health. These

various variations could result in morphological concerns, including the development and propagation of tumour cells (Cina SJ et al., 2001). Subsequently, lung tumours could also possibly turn into alveoli or major bronchus. Carcinoma is generally positioned in the central part of the lung, whereas large cell cancer is characteristically positioned in the peripheral part of the lung (Nillson K et al., 2006). HPV is involved in the pathogenesis of certain individual carcinomas, like carcinomas of the cervix and oropharynx (Syrjänen KJ et al., 2000). The human papillomavirus is usually linked to the development of cervical tumours. Human papillomavirus of type 16 and type 18 kinds are repeatedly related to severe inside the cell layers, in addition to malignant neoplasm (Syrjänen KJ et al., 1979). DNA of Human papillomavirus was discovered after a large number of biopsies within diverse separated suitcases of lung carcinoma (Nadji SA et al., 2007).

Twenty different types of cases with lung tumours are assimilated in this study in order to make a method of immunohistochemical to categorize Human papillomavirus in Lung Cancer tissue removed from a patient. All These patients used in the study had thoracotomy along with bronchoscopic biopsy as well. All of these thirty-five (35) lung cancer patients' blocks had been inserted with paraffin wax and also fixed with formalin. First of all, I will categorize patients on the basis of age group, on the basis of gender, and on the basis of smokers and nonsmokers. 30 male and five female patients were involved in this study. The ages of patients ranged from 36-96 years, with an average age of 63.55 years. Thirty-one patients out of these were smokers. The ages of these lung cancer patients are characterized by certain age groups: from 30-40, there is only one case, which is a male patient. From 40-50 years, there are two male cases, from 51-60 years, there is one female case, nine male cases, and two female cases, from 61-70 there were five male and two female cases. From 71 to 80 years, there were nine male cases and one female case, while the age group greater than 80 years had only one female case. The total sum of male Patients was thirty (30), and female patients were five (5). The number of patients who smoked is around thirty-one, including 25 male and six female patients. During this study, there were around five cases of adenocarcinoma, twenty-five cases of squamous cell carcinoma, and five cases of small cell carcinoma. During this research, a Formalin-fixed inserted chunk of benign skin was used as a control for the diagnosis of Human papillomavirus in lung cancer tissues.

In this study, The immunohistochemistry staining method used was labelled streptavidin-biotin technique, which was practically designed for Human Papilloma Virus discolouration. The paraffin-embedded and formalin-fixed tissue were subjected to microtome sections to sections in thickness. All of these sections were dehydrated in an oven for 60 minutes at 60 °C temperature. After that, taxation was done by dipping each slide in xylene baths for 5 minutes. After that, rehydration was done by repeated dilutions of alcohol in two-time alterations of 100% ethanol, 90% ethanol, and 80% ethanol for three minutes for each of these cases. For further rehydration, it was placed in a phosphate buffer saline clean bath at room temperature for almost thirty minutes. After that, the slides are cleaned by means of deionized Water, After that, they were transferred in a microwave oven in citrate buffer two times for a duration of five minutes. The extra solution is to get rid of it with awareness and by means of quick movement. Put 100 µl Anti-Human Papilloma (HPV), which is a monoclonal antibody solution, and coat the tissue slice. Incubation was done for almost sixty minutes

at 37 °C temperature in a humidified hall. After that, 100 µl Biotinylated link Antibody was placed in all slides to coat the tissue slice. Incubation for almost thirty minutes was done again at room temperature. Then, all slides were put in 100 µl Streptavidin. Incubation for almost twenty minutes in a humid chamber was done at room temperature. Then, adequate drops of DAB+ were put in and incubated for five minutes. Washing the slides gently for five minutes in water would do the required job. One of the slides was treated with a primary antibody to use as a positive control, and the second slide was not incubated with a primary antibody, so it could be used as a negative control.

The typical factor for positive immuno-reactions is dark brown in colour. Each type of cancer was specified with a certain score based on the amount of nuclear staining (Han CP et al. 2008). The immuno-reactive score was then calculated by taking the product of the intensity of positive scores of stained cells and their extent. The minimum score was set at 0, while the maximum score was set at 12 (Cuello, 1993). Variations of Human papillomavirus among age groups, gender, smoking status, and cell types were calculated using Pearson's Chi-square test. Research showed that HPV immuno-expression was reported in seven out of thirty-five cases of lung cancer, and only six cases were negative. This immuno-detection rate looks positively well-connected to patients who indulge in smoking. The immunoexpression of Human papillomavirus is also related to age groups in a significant way.

The results of this study have confirmed that eight out of thirty-five cases of lung carcinoma express Human papillomavirus, which was revealed by using immunohistochemical cytoplasmic staining, which seems to be brown in colour while staining. Out of those twenty-seven cases, seven were squamous cell carcinoma cases, and one case was adenocarcinoma. At the same time, zero cases out of six cases were diagnosed with small cell carcinoma. This lung cancer showed almost 20 per cent positive for the Human papillomavirus. The immune histochemical examination of the result shows that HPV is prevalent in seven out of thirty-two cases who are also smokers and in one out of eight cases for nonsmokers. Research showed that there was no correlation between the detection of Human papillomavirus and the smoking history of the patient. Generally, the rate of Human papillomavirus infection is lower in nonsmokers than in smokers.

Our study nearly agreed with the study conducted by (Kato et al., 2012), but it is not in line with the study conducted by (Ya-Wen Cheng et al., 2007), which showed that the Human papillomavirus was found in large numbers of nonsmoker patients than smokers. Immunoexpression of HPV revealed that not any of the female cases were exposed to HPV, but eight out of thirty males were exposed to Human papillomavirus. This difference in stats of HPV prevalence among gender groups was not so significant, which is in accordance with the study performed by (Yang Fei et al., 2010) Human papillomavirus was found to be in the case of male patients higher than in female p-value greater than 0.05. whereas research by (Cheng YW et al., 2007) showed that women with HPV expressed more than their male counterparts. The Immuno expression of Human papillomavirus, in relation to different age groups, revealed that no HPV exposure was shown in the age group between thirty and forty years, only one case between age 41 and 50 out of three cases, four cases between age 51 and 60 out of thirteen cases, two cases between the age group of 61 and 70 out of ten cases, one case

between age group 71 and 80 years out of twelve cases, and finally no case was reported in age group greater than 80 years. The difference among these age groups shows the effect of HPV exposure in a very different way. The immunohistochemical method for detection of this issue seems to be slightly higher in the age group between 51 and 60 years, which is also in correlation with the study performed by (Cheng YW et al., 2007).

Conclusion

The study concluded that the prevalence of the Human papillomavirus has been found to be nearly 20% among lung cancer cases, mostly in squamous cell carcinomas. Small cell carcinoma tissues did not demonstrate any sign of Human papillomavirus when they were studied using the immunohistochemical method. Detection of human papillomavirus in lung carcinoma can easily be achieved by using a well-correlated immunohistochemical technique. Detection and treatment of these symptoms is one thing, but prevention is far better than cure. One has to realize the threats of lung cancer and how it can affect a person both physically and mentally. Studies confirmed that smoking is one of the possible reasons for lung cancer, but the rate of active smokers is increasing day by day. A person has to realize that despite the invention of all these modern medical procedures, a person can still suffer if lung cancer is not diagnosed earlier. So prevention is always better than cure.

References

Ullman R, Bongiovanni M, Halbweld I, Petzmann S, GoggKammerer M, Sapino A, Papotti M, Bussolati G, Popper HH.(2003) Bronchiolar columnar cell dysplasia – genetic analysis of anovelpreneoplastic lesion of the peripheral lung. Virchows Arch, ; 442: 429-436

Cina SJ, Lancaster-Weiss KJ, Lecksell K, Epstein JI. (2001). Correlation of Ki-67 and p53 with the new World Health Organization/International Society of Urological Pathology Classification System for UrothelialNeoplasia. Arch Pathol Lab Med, 125: 646- 651

Brambilla E, Travis WD, Colby TV, Corrin B, Shimosato Y, (2001). The new World Health Organisation classification of lung tumour. EurRespir J 18: 1059-68

Mittal KR, Demopoulos RI, Goswami S. (1993) Proliferating cell nuclear antigen (cyclin) expression in normal and abnormal cervical squamous epithelia. Am J SurgPathol,;17: 117-22

Jensen-Taubman S, Steinberg SM, Linnoila RI. (1998) Bronchiolization of the alveoli in lung cancer: pathology, patterns of differentiation and oncogene expression. Int J Cancer, 75: 489-496

Nillson K, Landberg G. (2006). Subcellular localization, modification and protein complex formation of the CDK-inhibitor p16 in Rb-functional and Rb-inactivated tumour cells. Int J Cancer, 118: 1120-1125

Syrjänen KJ and Syrjänen S: (2000). Papillomavirus infection in human pathology. John Wiley & Sons, New York, pp1-10; 355-378.

Nadji SA, Mokhtari-Azad T, Mahmoodi M, et al: (2007). Relationship between lung cancer and human papillomavirus in the north of Iran, Mazandaran province. Cancer Lett 248: 41-46

Syrjänen KJ (1980). Epithelial lesions are suggestive of a condylomatous origin found closely associated with invasive bronchial squamous cell carcinomas. Respiration 40: 150-160,

Kamoi S, AlJuboury MI, Akin MR, et al. (2000) Immunohistochemical staining in the distinction between primary endometrial and endocervical adenocarcinomas: another viewpoint. Int J Gynecol Pathol;21:217-223.

Syrjänen KJ. (1979) Condylomatous changes in neoplastic bronchial epithelium. Report of a case. Respiration 38: 299-304.