Histopathological study of Human Papillomavirus (HPV) in breast cancer patients

Bushra JAl-Badry¹, Rana Talib Al-Muswie² AND Sameera Jameet³

Science collage¹, Dentistry College²,3, Thi-Qar University, Iraq.

e-mail: ranaalmuswie@gmail.com

Abstract. A large number of epidemiological studies have evaluated the association between Human papillomavirus infection and breast carcinoma, but the results have been inconsistent. Numerous types of cytopathic effect are distinct in living cultures, but fixation and staining of the cells is essential to realize appearances of viral infection such as inclusion bodies. Identifying cytopathic effect and using it as a investigative tool needs much practice in observing stained section of numerous cell types. The present study aimed at diagnosing the histological changes in the malignant breast tissue, which caused by the HPV and comparing them with malignant breast tissue caused by other factors in order to find a way to diagnose the virus through the histopathologic effect. A total of 100 samples was included in the study, 20 from benign tumor and 80 carcinomas, including different histopathological change. HPV was detected by using the conventional PCR. The breast cancer sections were examined for the expression of the effect of Human papillomavirus. In separate series the results of using formalin-fixed, paraffin-embedded sections were compared. Overall prevalence of HPV in malignant breast tissue was difference, we found that the histopathological characteristics of HPV-associated breast cancer are very different to breast cancer caused by other factors. Specifically, we showed the presence of HPV in breast cancer associates with the adenocarcinoma. The virus caused more activity in the tumor. Tumor is acinar carcinoma with increased cellularity and proliferative. While, in none infected with virus, there is no increased in cellularity or acinar, in some arease there was dilution of ducts. The observations indicate a likely causal role of HPV in causing a different histopathological change than breast cancercaused by other factors. This helps in the diagnosing of the HPV through cytopathic effect.

Keyword: Human papillomavirus; Breast cancer; Malignant; Benign; Histological change

Introduction

Breast cancer is characterised by the uncontrolled growth of abnormal cells in the milk producing glands of the (ducts) or breast that deliver milk to the nipples. It is the most commonly diagnosed cancer in women-worldwide¹. Viruses can be caused several human cancers. Human papillomaviruses
(HPVs) are accepted as being cervical, anogenital cancers and carcinogenic in human. High-risk human papilloma viruses (HPVs) may have a causal role in some breast cancers. However, the assessment of causality of HPVs in breast cancer is extremely difficult. The main difficulty is that HPV infections are common, but HPV-associated breast cancers are uncommon. Further, HPV infections may precede the development of breast and other cancers by years or even decades.

The main candidate viruses are human papilloma virus (HPV), mouse mammary tumor virus (MMTV), Epstein-Barr virus (EBV) and bovine leukemia virus (BLV). Each of these viruses has known oncogenic potential and all have been identified in normal and malignant human breast tissues. The past priority has been to confirm the identity of these viruses in breast tumors. The new priority is to determine whether or not they are causal rather than innocuous passengers invading pre-existing malignant tissues. According to the World Health Organization (WHO) classification, BC can be classified in up to 21 distinct histological types on the basis of cell morphology, growth, and architecture patterns. Breast cancer (BC) heterogeneity can be found at different levels, from the classic histopathological characterization to the more modern molecular classification. BC special types, including the classic lobular invasive carcinoma, represent 25% of all breast cancers. The most common histological type is invasive ductal breast carcinoma of no special type. The histopathological classification has a prognostic value. Two extreme examples are the tubular carcinoma, associated with an excellent prognosis, and metaplastic cancer, whose outcome is generally unfavorable. In all studies, high-risk HPV was found in tumor tissue only and not in surrounding normal tissue, except one study done in Turkey, in which the virus was also detected in normal tissue. Although the route of transmission for the virus has not been determined, Koilocytes are large epithelial cells with a vacuolated halo surrounding a dense nucleus. They are specific and indicative of HPV infection being caused by the action of HPV E5 and E6 on coproteins.

In this paper we report on the presence of HPV in breast cancer tissues, in the nuclei of cells within the cancerous regions of breast cancer specimens and correlate the presence of HPV with the histopathological features and not presence of HPV in tissue of breast cancer in malignant and Benign of breast cancer in woman.

**Materials and Methods:**

**Study design and samples.**

100 Samples from women diagnosed between January 2018 till July at the Hospital Al-Hussien in Nassiriya (Iraq). All the patients participated in this study were diagnosed as breast cancer by clinical examination. Malignant breast tissue specimens from patients with present Human Papilloma Virus associated breast cancer were aseptically collected by one surgical team immediately after tumor resection. HPV was not present in any of the specimens studied irrespective of histology. The sample was placed into a sterile tube and transported to the PCR Laboratory, snap frozen and stored at -80 °C. Unselected formalin-fixed breast cancer specimens and cancer specimens from women who had HPV breast, were analysed using PCR. Paraffin-embedded specimens obtained during surgical treatment were selected consecutively until achieving a maximum number of specimens representing the most frequent histological types of breast cancer, and clinical presentations, and then enriched with the most rare ones in order to have representation of all histological diagnoses.
Results

Histological change

The histological structure in breast cancer was shown there are differences histological characteristics of HPV positive and HPV negative marker human breast cancers and benign tumors are shown in the sections of results. HPV positive human breast cancers are not similar to other type of breast cancer.

In this recent study, we have unambiguously demonstrated the presence of high risk HPV in breast cancer cell lines and in the nuclei of breast tumor cells. The virus caused more activity in the tumor with increased acinar, cellularity and proliferative. While, in none infected with virus, there is no increased in cellularity in some areas there were dilution of ducts.

Histological changes of Malignant, HPV associated breast cancer

Tumor is acinar carcinoma, microscope is showed, increased acinar some areas increased cellular and at the periphery adipose tissue, and proliferative breast, in another slide tumor showed scattered cellularity, also, permanent stroma and dilated ducts (Fig. 1(A,B,C,D).

Histological changes of Malignant breast cancer

The histological structure in breast cancer was shown there were scatted acinar, permanent stroma, there is no increased in cellularity in some area there was dilation of ducts and present with adipose tissue at the periphery (Fig. 2(A,B,C,D).

Histological changes of benign HPV associated breast cancer

In the present study, we have been shown that the tissues with positive HPV benign has increase acinar, some feline rich with cellularity dilated ducts with some debacles of cells in the center of the lumen (Fig. 3 A,B,C,D).

Histological changes of benign breast cancer

In our study have been shown that the benign tissues with negative HPV results, mostly only dilated duct and reduce in acinar (Fig. 4 A,B,C,).
Figure(1): Histological special types of HPV associated breast cancer (malignant). Photomicrographs breast cancer appears (A) Increased the acinar (arrow). (B) Adipose tissue. (C) Scattered cellularity. (D) Stroma (head arrow) (H&E 100X)
Figure(2): Photomicrographs breast cancer appears (A) and (B) dilation of ducts (arrow). (C) Adipose tissue. (D) permanent stroma (head arrow). (H&E.100X)
Figure 3: Benign associated HPV. Photomicrographs appears in (A) and (B) the acinar (arrow), (C) dilation in ducts (D) stroma and adipose tissue (head arrow). (H&E.100X)
Discussion

In this study, we have confirmed the presence of HPV in the of cells in breast cancer tissue caused more activity in the tumor with increased cellularity and proliferative. While, in none infected with virus, there is no increased in cellularity in some areas there was dilution of ducts.

The detection of HPV in breast cancer is consistent with previous publications reporting the presence of HPV in breast cancer world-wide with a prevalence of ranging from 4–86%\textsuperscript{14}. Given such a high proportion of studies reporting HPV in breast cancer and breast tissue\textsuperscript{15,16}. The presence of HPV in some normal breast samples (normal breast and normal surrounding breast tissue) is consistent with a tumourigenic role for HPV in some breast cancers. If HPV has a causal role in breast cancer, it is reasonable to expect that this would be an early event, similarly with cervical cancerogenesis, and that
HPV would be found in at least some normal tissue. The possibility that some viruses could be linked to breast cancer aetiopathogenesis has important public health implications. In fact the role of viruses in some types of cancer has been clearly established. One of them is HPV, as some genotypes were recognized to be carcinogenic in humans in 1995. The first authors to report the presence of HPV in breast cancer tissues were Di Lonardo. After them, many other authors have studied the presence of this virus in breast tumors, finding varying detection rates ranging from 0 to 82%. Its presence has been described in breast milk, which indicates that this virus can infect breast tissue. A point that supports HPV being a carcinogen for breast tissue is that oestrogens act to increase its oncogenic effect. Oestrogens are considered an independent risk factor for development of a clearly HPV-related neoplasm, cervical cancer, especially since a meta-analysis including 24 epidemiological studies published in 2007 linked contraceptive use with this disease. Later in-vitro and in-vivo mouse assays showed that oestrogens cooperate with E7 HPV oncogene, transforming normal epithelium to a neoplastic one.

In present study we have been shown in histological change increased acinar and cellular in HPV associated breast cancer while in breast cancer non infected with HPV has a dilation in ducts. The fourfold higher prevalence of high-risk HPVs in breast cancer as compared to normal and benign breast tissues is a consistent finding among over 25 studies conducted in different laboratories in many contrasting populations. Some authors, however, suggested that HPV could reach breast tissue through haematic dissemination, but neither this hypothesis nor the former have been confirmed.

Human papilloma viruses are mainly transmitted by cell surface contact. Therefore, how are HPVs transmitted to the breast. A suggestive clue comes from the observation by de Villiers that HPVs are present in cancers occurring in human nipple milk ducts and that these cancers have the typical histological features of HPV-induced human cancers.

Conclusion

In the Malignant tumor the virus (Human Paplloavirus ) Histological structure has been difference compare with breast cancer non an infected with HPV by increased activity of the tumor, specified increased acinar ,some of them with proliferative and high cellularity .In case of benign fibroadenoma ,the virus increased number of acinar while in the non an infected ,fibro showed least acinar and dilated duct.

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