COMPARISON OF EFFICACY OF INTRALESIONAL PURIFIED PROTEIN DERIVATIVE (PPD) WITH CRYOTHERAPY IN THE TREATMENT OF CUTANEOUS WARTS

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ABSTRACT

Objective: To compare the efficacy of intralesional purified protein derivative (PPD) with conventional cryotherapy in the treatment of cutaneous warts.

Study Design: Quasi experimental study.

Place and Duration of Study: Dermatology Department, Pak Emirates Military Hospital (PEMH) Rawalpindi, from Jul 2017 to Jan 2018.

Methodology: Total number of 60 patients were included in the study through Out Patient Department OPD. All patients were randomly divided in 2 groups by lottery method. Group A patients were given intra-lesional PPD (0.1ml in a single wart or targeting the largest wart in case of multiple warts, procedure was repeated fortnightly) while Group B patients were given cryotherapy with liquid nitrogen (2 freeze thaw cycles 20 sec duration each fortnightly). All patients were followed up after the end of treatment (maximum 6 sessions i.e. 3 months or complete clearance of wart whichever is earlier) and assessment of treatment efficacy was based on the response which was noted after 3 months of treatment. The response was categorized as excellent (no visible lesion), good (50-99% improvement), intermediate (<50% improvement) and poor (no response) depending upon the decrease in lesion numbers and size as measured by calibrated scale. The treatment was considered efficacious if there was good to excellent response.

Results: A total of 21 (70%) cases in PPD and 9 (30%) cases in cryotherapy were treated successfully. Efficacy of treatment was significantly higher in PPD group compared to cryotherapy group, *p*-value <0.05.

Conclusion: Intralesional purified protein derivative (PPD) was more effective in the treatment of cutaneous warts as compared to conventional cryotherapy. PPD may be adopted for treatment of viral warts to gain more satisfactory results and efficacy.

Keywords: Cutaneous warts, Cryotherapy, Intralesional treatment, Purified protein derivative.

INTRODUCTION

Wart is a benign proliferation of the skin and mucosa caused by double stranded DNA virus known as human papilloma virus (HPV)[1]. Wart can be classified into several types like common wart, flat wart, plantar wart and genital wart according to site and form of the lesion[2]. There are various modalities for the treatment of warts, almost all of which are effective in some patients; therefore, a combination of them can be used[3]. The role of immunity is apparent by the appearance and persistence of warts in immunocompromised patients. A fully functional immune system is required for clearance of HPV from the epidermis. Treatment of warts by ablative therapies frequently leads to recurrence and thus persistence[4].

Cryotherapy has been conventionally used in the treatment of warts. Liquid Nitrogen with a boiling point of –195.6°C is the cryogen of choice. Cryotherapy can be performed through an open spray technique or through direct application of a dipstick or cooled probe. Cryotherapy has been commonly performed in the outpatient department setting because of its effectiveness, safety, low cost, good cosmetic results, ease of use and lack of the need for local anaesthesia. It works by cooling the target tissue. Cryotherapy induces tissue injury by formation of ice crystals within cells, vascular thrombosis and vascular stasis and

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release of electrolytes and toxins. Rapid freezing causes sloughing of the epidermis from the dermis. While cryotherapy has many advantages, it is not free from side effects like blistering, bleeding, edema, pain, vasovagal syncope, hypo/ hyperpigmentation, ulceration etc[5,6]. A study show around 18 to 33% of patients had complete resolution of warts after three months of cryotherapy sessions[7].

Immunotherapy is a promising modality for the treatment of recurrent and resistant warts which leads to resolution without scarring and additionally boosts the host response against the causative agent. This results inabsolute resolution and reduced recurrence. Various studies have been done in the past where immunotherapy has been tried with variable success[4]. Some agents have been studied comprehensively like cimetidine[8], imiquimod[9], others being under evaluation such as Measles Mumps Rubella (MMR) vaccine[10], Mycobacterium w vaccine[11], Bacillus Calmette-Guerin (BCG) vaccine[12].

PPD (Purified Protein Derivative) or tuberculin activates the T helper 1 (Th1) cells, thus stimulating the cell-mediated immune response, Natural killer (NK) cells and production of cytokines. This release of interleukin (IL-12) as a process in enhancing the cell-mediated immune responseadds up to the mechanism of action[13]. In one study 62.2% patients showed complete clearance at injected and distant warts at the end of three months treatment with intralesional PPD[14].

A quasi experimental study to compare the efficacy of intralesional immunotherapy versus cryotherapy in treatment of viral warts over a course of three months was planned and conducted at PEMH Rawalpindi as the results of studies using PPD were promising in other parts of the world. The study was done with the idea that if PPD turned out to be more effective than cryotherapy, it may be used in the future for the treatment of recalcitrant viral warts.

METHODOLOGY

This quasi experimental study was carried out at outpatient department of dermatology, Pak Emirates Military Hospital Rawalpindi. The study was accomplished over a six months period from 3rd July, 2017 till 2nd Jan, 2018. WHO sample size calculator was used to calculate the sample size (Confidence level = 95%, Power of test = 80%). The anticipated population proportion (P1) was 62.2%[14] and population proportion (P2) was 18.2%[7]. The sample size was 60 with random allocation of 30 patients in groups A and B. The sampling technique was non-probability consecutive sampling. Patients aged 15 to 60 years, of both genders, with cutaneous warts diagnosed clinically having wart duration of minimum 1 week or a maximum of 12 months and not treated for viral wart in last 6 months were included in the study. Pregnant/ lactating ladies, patients on any systemic/ immunosuppressive therapy, patients with any known systemic illness and patients not willing for follow-up were excluded.

The study was conducted after getting approval from hospital's ethical and research committee. The rationale and benefits were made clear to the patients and they were guaranteed that the study was being done merely for data publication and research purposes and an informed consent in written form was taken. All the patients were placed randomly into two groups by lottery method. They were offered picking up a chit out of total mixed up chits (half of the chits had letter 'A'while other halfhad letter 'B') and he/she was allotted the respective group.

Patients in group A were given 0.1 ml (5TU) of intralesional PPD with an insulin syringe injected at the base of the largest wart intradermally while patients in group B were subjected to cryotherapy using liquid nitrogen (-196^{o}C) for two freeze and thaw cycles. The procedure was carried out by direct application using orange stick with cotton tip. Scales were removed gently (if needed) before the procedure using sterile blade. The session was repeated in each group at 2 weekly intervals for six times or less in case of complete clearance of wart. All patients were followed after the end of treatment (3 months) and efficacy was noted. The response was

categorized asexcellent (no lesion visible), good (50-99% resolution), intermediate (<50% resolution) and poor (no response) depending upon decrease in lesion number and size as measured by a calibrated scale. Data was recorded on a specially designed proforma.

The analysis of data was performed in SPSS version 17. Mean ± SD was used for calculation of quantitative variables. Percentages and frequencies were calculated for the qualitative variables i.e. gender and efficacy in each group. Chi square test was applied for comparison of efficacy in 2 groups and *p*-value ≤0.05 was considered statistically significant.

RESULTS

The mean age of cases in PPD group was 37.77 ± 13.53 years and mean age in Cryotherapy group was 40.83 ± 12.99 years. In PPD group there were 18 (60%) male and 12 (40%) female cases while in Cryotherapy group there were 13 (43.3%) male and 17 (56.7%) female cases. The mean duration of disease in PPD and Cryotherapy group was 5.33 ± 3.22 and 6.63 ± 3.55 months respectively. The mean number of warts in PPD group was 8.80 ± 4.81 and in Cryotherapy group were 9.00 ± 4.81 respectively.

A total of 21 (70%) cases in PPDn group and 9 (30%) cases in Cryotherapy group had efficacy of treatment which was significantly higher in PPD group compared to Cryotherapy group, (*p*-value = 0.02) (table-I).

The results were also analyzed with respect to difference in age group, gender, duration and number of warts in a patient.

In 15-35 years of age group there were 10 (71.4%) cases in PPD group and 3 (30%) in Cryotherapy group and in 36-60 years of age group there were 11 (68.8%) cases in PPD group and 6 (30%) cases in Cryotherapy group who were treated effectively. In both the age groups, the efficacy of PPD was significantly higher compared to Cryotherapy, (*p*-value <0.05 in both groups).

There were 12 (66.7%) male cases in PPD group and 3 (23.1%) in Cryotherapy group, while there were 9 (75%) female cases in PPD group and 6 (35.3%) in Cryotherapy group who were treated effectively. In both male and female cases the frequency of efficacy of PPD was significantly higher compared to Cryotherapy group, (*p*-value <0.05 in both groups).

Table-I: Comparison of efficacy of treatment in both study groups.

		Study groups	
		PPD	Cryotherapy
Efficacy	Yes	21 (70.0%)	9 (30.0%)
	No	9 (30.0%)	21 (70.0%)

p-value = 0.002

Table–II: Comparison of efficacy of treatment in both study groups with respect to duration of warts.

Duration	Efficacy	Study groups		*p*-value
		PPD	Cryotherapy	
<6 months	Yes	12 (70.6%)	3 (25%)	0.016
	No	5 (29.4%)	9 (75%)	
≥6 months	Yes	9 (69.2%)	6 (33.3%)	0.048
	No	4 (30.8%)	12 (66.7%)	

Table-III: Comparison of efficacy of treatment in both study groups with respect to number of warts.

No. of Warts	Efficacy	Study groups		*p*-value
		PPD	Cryotherapy	
1-8	Yes	12 (75%)	5 (33.3%)	0.02
	No	4 (25%)	10 (66.7%)	
9-17	Yes	9 (64.3%)	4 (26.7%)	0.042
	No	5 (35.7%)	11 (73.3%)	

In cases with duration < 6 months there were 12 (70.6%) cases in PPD group and 3 (25%) in Cryotherapy group and in cases with duration of ≥6 months there were 9 (69.2%) cases in PPD group and 6 (33.3%) cases in Cryotherapy group who had effective treatment. In both groups the frequency of efficacy of PPD was significantly higher compared to Cryotherapy, *p*-value <0.05 (table-II).

In cases with 1-8 number of warts there were 12 (75%) cases in PPD group and 5 (33.3%) in Cryotherapy and in cases with 9-17 number of

warts there were 9 (64.3%) cases in PPD group and 4 (26.7%) cases in Cryotherapy group had efficacy of treatment. In both groups the frequency of efficacy of PPD was significantly higher compared to Cryotherapy, *p*-value <0.05 (table-III).

DISCUSSION

Warts are one of the common dermatologic disorders caused by the human papilloma virus (HPV). Warts generally affect the children and young people. Various studies conclude that almost 10% of the young population has suffered from warts[15]. Some of the warts regress spontaneously, whereas others persist. They can also spread to other areas of the body resulting in physical as well as emotional distress of the patients. The therapeutic options currently available for treatment of warts include eletrocautery, cryotherapy, surgical removal, laser ablation, intralesional bleomycin, topical agents for example 5FU/salicylic acid, and immunotherapy[16]. The listed treatment options eradicate warts by direct destruction and / or by the stimulation of immunologic responses against the virus affected keratinocytes. Few agents possess intrinsic antiviral activity[17,18].

The best possible treatment option should be opted by the clinician from the multiple available therapeutic choices suitable for individual cases taking into account the patient's age, sex, past medical history and characteristics of the warts. Cryotherapy with liquid nitrogen is very useful in treating warts, particularly those on the periungual and palmoplantar areas[19]. Immunotherapy has been tried with various antigens and vaccines such as bleomycin, PPD, MMR, Candida albicans and Mycobacterium w vaccine[20].

In our study a total of 21 (70%) cases in PPD group and 9 (30%) cases in Cryotherapy group had efficacy of treatment which was significantly higher in PPD group compared to Cryotherapy group, *p*-value <0.05. This was comparable to one of our reference study in which the reduction in lesion size (good: Complete clearance, intermediate: 50–99% clearance, poor: <55% clearance), adverse effects (if any) and recurrence within six months follow-up were documented. The result showed that at the final session, 'good', 'intermediate' and 'poor' responses were observed in 77.1%, 22.9% and 0% of the PPD patients; 0%, 14.7% and 85.3% of the placebo patients and 18.2%, 33.3% and 48.5% of the cryotherapy patients, respectively (PPD versus placebo: $p<0.001$; PPD versus cryotherapy: $p<0.001$). In the PPD group no notable complication was seen. The observed rate of recurrence was 8.6%, 5.9% and 24.2% in the PPD, placebo and cryotherapy groups, respectively ($p<0.05$). This study has also concluded that immunotherapy with intralesional PPD antigen is very effective as well as safe in the treatment of recalcitrant warts[21]. Recently a study was done by Khozeimah *et al* to compare the efficacy of immunotherapy vs cryotherapy on cutaneous warts. A significant difference was observed between the therapeutic response between both groups (*p*-value=0.041). 76.7% patients were entirely cured with immunotherapy, however just 56.7% responded to cryotherapy. Complete remission with fewer sessions was observed (20.17 ± 0.65) in immunotherapy patients compared to the patients receiving cryotherapy (3.82 ± 2.481)[22]. Similarly another study was done to study the impact of PPD in treatment of cutaneous warts. The trial included patients with resistant warts for immunotherapy. Each patient was given 2.5 TU of PPD in the warts intralesionally. Four sessions were administered at a 2 weekly interval and patients were advised follow up for six months after the last session. After four sessions, forty two (76%) patients had a complete resolution while remaining 13 (24%) patients did not respond. The study concluded that immunotherapy with PPD is beneficial in the treating the cutaneous warts[23]. We found 70% efficacy in PPD group that was almost same to above study.

Likewise Nimbalkar *et al* has performed a study to assess the effectiveness and safety of immunotherapy using purified protein derivative (PPD) for treating viral warts. Forty five patients participated in the study. Each patient received 10 TU of tuberculin PPD (0.2 cc) intralesionally in

the largest wart at 2 weekly intervals. A total of six treatment sessions were conducted. Resolution of viral warts was considered as the clinical end point of the study. Photographic measurements were done for clinical assessment at baseline, prior to each treatment session and 3 weeks after treatment was completed. The result of the study revealed that a total of 62.2% patients (28 out of 45) showed complete clearance at injected and distant warts, eight patients (17.8%) showed partial clearance, and nine patients (20%) showed no improvement. They concluded that tuberculin PPD immunotherapy was found to be a safe and effective treatment modality for the treatment of viral warts[24].

In our study, there was no marked difference in the mean age for most patients were falling in the age bracket of 25-45 years. Gender difference and duration of illness also did not show any marked differences in both groups.

Despite these highly promising findings, however, it is suggested that other variables like frequency and number of treatment sessions, factors related to the antigen i.e the dosage, combination, type and antigenic potency; and the characteristics of the warts like number, duration, site, size and type need evaluation in detail in further clinical trials to convince clinicians for using intralesional PPD suitably and consistently. The adverse effects are uncommon to PPD (none seen in our study) however a person who has strong immunity against Mycobacterium may occasionally develop a sizable reaction which may cause some itching, swelling or irritation.

CONCLUSION

There was higher efficacy of intralesional PPD vs. Cryotherapy in the treatment of cutaneous warts. PPD can be adopted for treatment of viral warts.

CONFLICT OF INTEREST

This study has no conflict of interest to declare by any author.

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