Combination therapy for dendritic keratitis with human leucocyte interferon and trifluorothymidine

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SUMMARY Sixty-one patients were treated in a randomised double-masked clinical trial either with a combination of human leucocyte interferon 10 million IU/ml and trifluorothymidine (TFT) in a 1% solution or with a combination of albumin placebo and trifluorothymidine. The healing time of dendritic keratitis was significantly lower with the combination of interferon and TFT than with the combination of placebo and TFT. For partial healing the reduction was 49% and for complete healing 42%.

Herpes keratitis is still considered to be one of the most serious diseases of the outer eye. With the advent of virostatics¹ in 1962 there has been progressive improvement in the treatment of dendritic keratitis. At present interferon treatment is at the centre of interest. It was discovered in 1957, but as it was very difficult to produce in large quantities experiments^{2,3} were limited.

Studies with interferon or interferon inducers alone⁴⁻⁹ in controlling dendritic keratitis gave disappointing results. In combination with virostatics^{4,10} interferon was reported to be more effective than virostatics alone.

We have treated patients with dendritic keratitis and superficial epithelial lesions with interferon or placebo in combination with trifluorothymidine in a double-masked randomised trial.

Material and methods

Patients with dendritic keratitis or morphological variants were treated in a randomised arrangement with eye drops containing either human leucocyte interferon (HLI) 10 million IU/ml and trifluorothymidine (TFT) in a 1% solution, or albumin placebo and trifluorothymidine in a 1% solution.

Patients with metaherpes,¹¹ stromal keratitis, or who had previously received steroid treatment were excluded from the trial.

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All patients received 5 drops of TFT daily. In addition they received every morning after careful eye examination one drop of HLI¹² or placebo in a recumbent position. After 10 minutes a second drop was given, and the patients remained recumbent for another 10 minutes to ensure that the HLI or placebo kept in contact with the surface of the outer eye.

Before treatment we isolated herpes simplex virus from the cornea and conjunctiva by minimal wiping. Virus identification was done at the National Institute of Public Health in Bilthoven, The Netherlands. During transport and storage eye swabs were kept in GLY-medium, i.e., Hanks's balanced salt solution containing 0.5% gelatin, 0.5% lactalbumin hydrolysate, 0.1% yeast extract, and antibiotics. The specimens were inoculated onto HEP-2 and on to GaBi cells (human diploid fibroblasts) in tubes. Cultures with characteristic cytopathic effect were identified as herpes simplex virus by serum neutralisation in GaBi cells. Typing was done in GaBi cells in microplate culture, with the use of rabbit antisera monospecific for type 1 and type 2, respectively.

We used 2 criteria for healing: (1) partial healing, which we defined as closure of the epithelial wound only, i.e., no staining with fluorescein; and (2) complete healing, which we defined as closure of the epithelial wound without any epithelial oedema or cystic changes in the area of the previous dendrite.¹³ We gave the HLI or placebo until the third day of partial healing; the TFT was continued till complete healing.

Fig. 1 Cumulative frequency graph for partial healing time of dendritic keratitis treated with interferon/TFT or placebo/TFT.

Results

Eight patients were excluded from the study, 2 because of omissions in complying with the examination protocol, one because he developed metaherpes; this patient received the placebo/TFT combination. Three patients were excluded because all herpes simplex virus isolations were negative. Two other patients developed severe stromal keratitis; both received the placebo/TFT combination.

Of the remaining 53 patients (33 males and 20 females) 25 were treated with HLI and TFT (15 males and 10 females) and 28 were treated with placebo and TFT (18 males and 10 females).

In Fig. 1 the number of days for partial healing of

Fig. 2 Cumulative frequency graph for complete healing time of dendritic keratitis treated with interferon/TFT or placebo/TFT.

dendritic keratitis is given in a cumulative frequency graph. The average time for partial healing of the interferon/TFT treated group was 3.3 days and of the placebo/TFT treated group 6.5 days.

Table 1 is the table of analysis of variance for the data of partial healing. The treatment effect was statistically highly significant ($p < 0.01$).

In Fig. 2 the number of days for complete healing of dendritic keratitis is given in a cumulative frequency graph. The average time for complete healing of the interferon/TFT treated group was 6.6 days and of the placebo/TFT treated group 11.3 days. The treatment effect of complete healing was also statistically highly significant ($p < 0.01$).

Table 1 Analysis of variance of partial healing time

Nature of effect	Source	Sum of squares	DF	Mean sum of squares
Main factors	Therapy (T)*	136.94	1	136.94
	Sex (S)	2.32	1	2.32
	Eye (E)	0.93	1	0.93
Interactions	T×S	2.37	1	2.37
	T×E	3.53	1	3.53
	S×E	1.20	1	1.20
Residual		111.68	46	2.43
Total		258.97	52	

DF=degrees of freedom. * $p < 0.01$.

Fig. 3a

Fig. 3b

Fig. 3c

Fig. 3d

Fig. 3 These photographs show the healing process of dendritic keratitis after treatment with the interferon/TFT. (a) Before treatment. (b) One day after treatment. (c) Two days after treatment. (d) Three days after treatment.

The interval in days between partial and complete healing of dendritic keratitis for the interferon/TFT treated group was 3.3 days and for the placebo/TFT treated group 4.8 days. This difference was statistically significant ($p < 0.01$).

The healing process after treatment with interferon/TFT is illustrated in Fig. 3, a-d.

Discussion

Only those corneal herpetic lesions were studied in this trial in which active virus replication was present in the epithelial cells. We therefore excluded metaherpes, which is considered to be a trophic herpetic disease, and also stromal forms in which immunological reactions rather than virus replication are the important factor.

Many authors⁴⁻⁹ have concluded that interferon or interferon inducers alone did not significantly reduce the healing time of dendritic keratitis as compared with the natural healing time.

Our results indicate that the combination of interferon and TFT gave a lower healing time for dendritic keratitis than TFT alone. In addition the 2 forms of treatment led to a quicker healing time than leaving the ulcers untreated. We have previously recorded a natural partial healing time of 13.8 days ($SD=4.1$) and a natural complete healing time of 20 days ($SD=3.6$).¹⁴ In the present trial for TFT alone the comparable data are 6.5 days ($SD=1.5$) and 11.3 days ($SD=2.6$) respectively. The percentage gain for TFT alone is 53% for partial healing and 44% for complete healing. If interferon is used in combination with TFT, these data are 3.3 days ($SD=1.1$) and 6.6

days (SD=1.4) respectively. Here the percentage gain as compared with the natural healing time is 76% for partial healing time and 67% for complete healing time.

Because there is a difference in the interval between partial and complete healing of the interferon/TFT treated group and the placebo/TFT treated group, we may assume that the virus still exists in the period between partial and complete healing.

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