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# Treatment of Acute Nonlymphoblastic Leukemia in Adults with Daunorubicin-DNA Complex: A Preliminary Report

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***Summary.*** *Forty-four adult patients under 60 years of age with acute nonlymphoblastic leukemia were randomized for induction treatment with one of the following three regimens: R 1 = courses of daunorubicin on day 1 + ARA-C on days 1–5; R 2 = courses of daunorubicin on days 1 and 2 + ARA-C on days 4–8; R 3 = courses of daunorubicin-DNA complex on days 1–2 + ARA-C on days 4–8.*

*Out of 14 patients, 9 went into remission on R 1, 6 out of 14 on R 2, and 8 out of 16 on R 3. The preliminary results suggest that daunorubicin-DNA complex has the same efficacy for inducing remission as daunorubicin alone, if the same time intervals and dosages are used.*

## Introduction

Daunorubicin is one of the most effective drugs for induction of remission in acute nonlymphoblastic leukemia (Jacquillat et al., 1966; Bernard et al., 1969; Crowther et al., 1970; Weil et al., 1973; Udén et al., 1975). Its use for maintenance of remission is limited by its cardiotoxic effects, which are clearly related to the cumulative dose (Lefrak et al., 1973; von Hoff et al., 1977).

In 1972 (Trouet et al., 1972), it was suggested that daunorubicin could be changed into a so-called lysosomotropic antitumoral drug by linkage to DNA. Experiments with cultured fibroblasts indicated that daunorubicin-DNA complex is taken up by endocytosis into the lysosomes, where DNA is digested by lysosomal enzymes. Thus, free daunorubucin becomes available for action on nuclear DNA. It was also shown that cardiotoxicity was decreased, probably because the cardiac muscle cells have low endocytotic activity and therefore do not take up the complex (Langslet et al., 1974).

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Results in L 1210 leukemia (Trouet et al., 1972, 1974, 1975) indicate that the therapeutic effect of the complex is higher than that of free daunorubicin. However, conclusive results showing higher efficacy in human leukemia have so far not been presented, although preliminary results of nonrandomized studies indicate that the complex may be as effective as the free drug (Sokal et al., 1973; Cornu et al., 1974; Michaux et al., 1975). In agreement with the experimental evidence, it also appears that cardiotoxicity in patients is reduced by using the complex (Sokal et al., 1973; Cornu et al., 1974; Michaux et al., 1975).

The aim of the present study is to compare the efficacy and toxicity of daunorubicin-DNA complex to free daunorubicin in a randomized study. This is a preliminary report of an investigation, which was started one and a half years ago. Results of other such studies have to our knowledge not been reported until now in this symposium (Ferrant et al., 1978).

## Materials and Methods

*Induction Treatment.* Forty-four adult patients under 60 years of age, with newly diagnosed, previously untreated, acute nonlymphoblastic leukemia were randomized for treatment with one of three different regimens (Fig. 1). Regimen 1 (R 1) was identical with the regimen previously used by the Leukemia Group of Central Sweden (Lindemalm et al., 1978), regimen 2 (R 2) was identical with regimen 3 (R 3) with only one exception, R 2 contained free daunorubicin while R 3 contained daunorubicin-DNA complex. R 3 was created in collaboration with Dr. Trouet (Trouet, personal communication, 1976), who had experimental evidence that two courses of daunorubicin-DNA complex on two consecutive days followed by a one-day free interval between these courses and the five-day course of ARA-C might be superior to a program timed as R 1. For a correct comparison between the complex and free daunorubicin, R 2 was designed.

*Maintenance Therapy.* Patients in remission were maintained on monthly courses of chemotherapy. Patients induced on R 1 received every other month one course of daunorubicin + ARA-C, similar to

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the induction course, only with the exception that ARA-C was given only once a day. Every other month, the patients received thioguanine 2 mg/kg/day, and ARA-C 1 mg/kg/day for 5 days. Patients induced on R 2 received a similar maintenance treatment – the same dosages of drugs, but a one-day free interval between daunorubicin and ARA-C. Patients induced on R 3 received a treatment identical with R 2, except that daunorubicin-DNA complex was substituted for daunorubicin. Half of the patients were randomized to immunotherapy with BCG + allogeneic leukemic cells. However, these patients were evenly distributed between the different induction regimens, since separate randomization was made for patients on R 1, R 2, and R 3, respectively (Fig. 2).

*Preparation and Administration of Daunorubicin-DNA Complex.* DNA (herring sperm DNA, type VII, Sigma, St. Louis, USA) was dissolved to a concentration of 2.34 mg DNA/ml saline. The mixture was heated to 95° C and then filtered on a 0.8 μm Millipore filter. This solution was stored for no longer than 6 months at the respective hospital pharmacy. About 12 h before use, the solution was autoclaved at 120° C for 15 min and was then left to cool slowly until use, not later than 24 h after autoclaving. Daunorubicin (Cerubidin®, Leo-Rhodia) was dissolved in saline to a concentration of 20 mg/ml and was then added to the DNA solution to 100 volumes of DNA solution. The administration was performed for 4–5 h at a rate of about 100 ml/h.

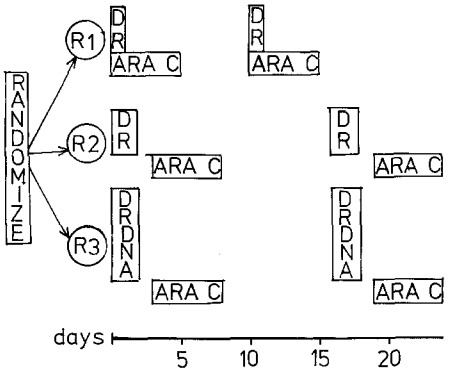


**Fig. 1.** Induction program

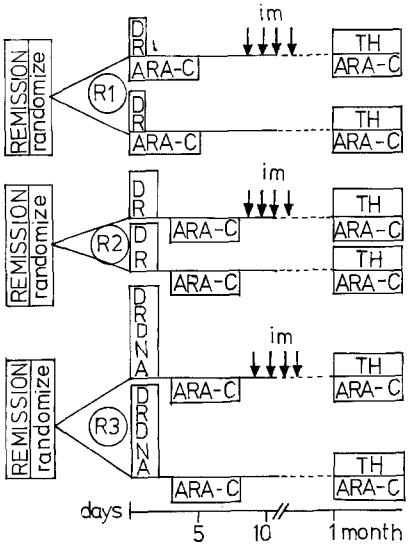


**Fig. 2.** Maintenance program

## Results

*Remission Induction.* A complete remission was induced in 9 of 14 patients on R 1, 6 of 14 on R 2, and 8 of 16 on R 3. There was a tendency for a larger number of induction courses (Fig. 3) and a somewhat longer time to remission (Fig. 4) on R 1 than on R 2 and R 3. However, the tendency to induce remission was higher on R 1 than on R 2 and R 3 although no statistical significant difference was found.

*Duration of Remission and Survival.* As yet, no definite conclusion can be drawn as to the duration of remission and survival. Of nine patients who entered remission on R 1, five are still in remission 187–544 days after diagnosis, two of six patients who entered remission on R 2 are still in remission 278–426 days, and on R 3 four of eight patients are still in remission after 100–271 days.

Although it is too early to draw conclusions, there appears to be no clear difference between R 2 and R 3, and R 1 may be superior to the other two regimens.

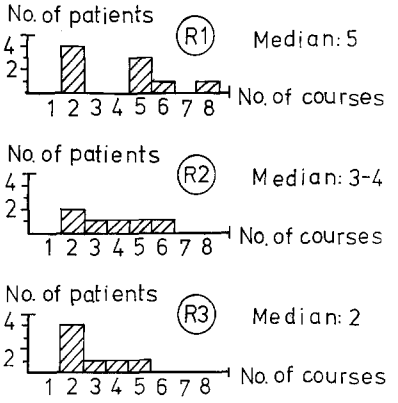


**Fig. 3.** Number of chemotherapy courses to remission on three different chemotherapy programs

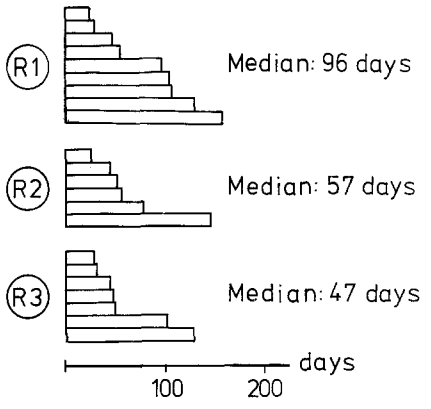


**Fig. 4.** Time to remission on three different chemotherapy programs

*Toxicity.* No patient has shown cardiac insufficiency or heart dilatation, but so far no patient has received a dose of the complex exceeding 10 mg daunorubicin/kg. Other side effects appear to be similar to the side effects for free daunorubicin.

## Discussion

The purpose of the present investigation was, first, to study whether the daunorubicin-DNA complex was comparable or hopefully better than free daunorubicin for induction of remission in acute nonlymphoblastic leukemia, and, second, to study whether the complex might be superior in maintaining remission with less or no cardiotoxicity. Only the first aim can be discussed, since no patient has so far received a cumulative dose of daunorubicin-DNA complex exceeding 10 mg daunorubicin/kg.

Although the material is small, the results permit the suggestion that daunorubicin-DNA complex is not superior to free daunorubicin for induction of remission. The results of R 2 and R 3 are practically identical and the result of R 1 has a slightly better tendency. This program differed in two respects from R 2 and R 3. First, the infusion time was shorter; 200 ml of daunorubicin solution was infused for 45 min as compared with 500 ml during 4–5 h on R 2 and R 3. The main difference was, however, that daunorubicin was given only one day in a chemotherapy course of five days, while on R 2 and R 3 daunorubicin or daunorubicin-DNA complex, respectively, was given on two consecutive days with a free interval of one day before the five-day course of ARA-C. It is possible that this difference in timing and dosage between R 1 as opposed to R 2 or R 3 will eventually show that R 1 is preferable. In our view, it therefore remains to be studied whether daunorubicin-DNA complex would not be as effective as free daunorubicin in R 1 if timed similarly.

Other reasons for the tendency to less favorable results on R 2 and R 3 are unlikely. The age of the patients, – one of the most important prognostic factors (Udén et al., 1975), – was in fact higher for patients on R 1 than on R 2 or R 3, and the proportion of patients on chemoimmunotherapy, which may have a favorable influence on survival (Gahrton et al., 1976; Reizenstein et al., 1977; Lindemalm et al., 1978) was not given to a higher proportion of patients on R 1 than on R 2 or R 3. Thus, our preliminary impression is that the efficacy of daunorubicin-DNA complex for induction of remission in acute nonlymphoblastic leukemia is most probably equivalent to that of daunorubicin in the free form. However, the timing and dosage may not be optimal in the present program, so in our next program the intension is to randomize for only two regimens, i.e., for R 1, – which in this study and our previous studies (96 consecutive patients under 60 years of age has resulted in a 58% frequency of remission (Leukemia Group of Central Sweden, unpublished data) – and for a regimen with daunorubicin-DNA complex + ARA-C in dosages and timing similar to those in R 1.

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