Dose-Dense Induction
with Sequential-High-Dose Cytarabine and Mitoxantone (S-HAM) and Pegfilgrastim results in a High Efficacy and a Short Duration of Critical Neutropenia in de-novo Acute Myeloid Leukemia
– A Pilot Study of the AML-CG

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Running Title: S-HAM in de-novo AML

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Abstract

Dose density during early induction has been demonstrated to be one of the prime determinants for treatment efficacy in acute myeloid leukemia (AML). The German AML-Cooperative Group has therefore piloted a dose dense induction regimen S-HAM (sequential high-dose AraC and Mitoxantrone followed by pegfilgrastim) in which two induction cycles are applied over 11 - 12 days instead of 25 – 29 days as used in conventional double induction thereby increasing dose density two-fold. Of 172 de-novo AML patients (excluding acute promyelocytic leukemia) 61% reached a complete remission, 22% a complete remission with incomplete peripheral recovery, 7% had persistent leukemia, 10% succumbed to early death - resulting in an overall response rate of 83%. Kaplan Meier estimated survival at 2 years was 75% for the whole group [patients with unfavourable karyotypes 38%; patients with favourable karyotypes 69%; patients with intermediate karyotypes 75%] after S-HAM treatment. Importantly the compression of the two induction cycles into the first 11 - 12 days of treatment was beneficial for normal hematopoiesis as demonstrated by a significantly shortened duration of critical neutropenia of 31 days as compared to 46 days after conventionally timed double induction. The clinical study is an official study of the "Kompetenznetz Akute und Chronische Leukämien" and is registered in the European Leukemia Trial Registry with the registration number LN_AMLINT_2004_230.
1. INTRODUCTION

The overall prognosis of patients suffering from AML has steadily improved over the last three decades. Nowadays, complete remissions are achieved in 60 – 70% of all patients with long-term disease-free survival and potential cure in 25 – 40% of cases. A more detailed analysis indicates that this progress has mainly been achieved in patients <60 years of age while in older patients little improvements have been obtained 1-5.

When analyzing the approaches that underlie the progress in AML therapy two major developments appear essential: the intensification of therapy and the improvement of supportive care.

In the last few years increasing insights into the biology of AML have been gained. It has become clear that AML is a not a homogeneous disease but rather a group of different subtypes. These subtypes differ not only in their biology but also in their prognosis. Therefore genetic markers have become mandatory to discriminate prognostic subgroups and to adjust treatment according to distinct risk groups 6-12. However the definition of distinct subgroups of AML is still mainly descriptive and the major challenge for clinicians and translational researchers remains how to treat these different subgroups most effectively and how to improve on their current outcome. Except for acute promyelocytic leukemia the better understanding of AML biology and the development of “targeted” therapies so far has not resulted in significant improvements in overall survival.

The standard induction therapy for AML is still a “3+7” type regimen comprising three days of Daunorubicin and seven days of standard-dose Cytosine Arabinoside (AraC) as continuous infusion 13,14. Based on cell biologic data, the German AML-CG modified the 3+7 regimen and established the TAD-9 regimen which is the combination of Thioguanine, AraC, and Daunorubicin 15. The TAD-9 regimen resulted in a high CR rate and has been part of the induction strategies of the German AML-CG since 1979. For the dosing of Daunorubicin in particular an improvement in overall survival was demonstrated for full dosing as compared to reduced dosing in patients older than 60 years 16,17.

In an attempt to improve the long-term prognosis of patients with AML, the AML-CG introduced the concept of “double induction”. This strategy is primarily focussed on patients < 60 years of age. It consists of two courses of chemotherapy irrespective of the degree of cytoreduction in the bone marrow after the first course with the second course starting on day 21 unless severe complications prohibit its application.

This strategy resulted in a significantly longer remission duration and overall survival as compared to standard induction 18. In order to further improve on these results double induction with two courses of TAD 9 was compared to a first course of TAD 9 followed by high dose AraC (HD-AraC) plus Mitoxantrone (HAM) as second course. While no significant
differences in outcome were observed for the overall group of patients a favourable effect of HAM was seen in the subgroup of high-risk patients as defined by unfavourable karyotype and/or elevated LDH level and/or residual day 16 bone marrow blasts with an OS at five years of 25% vs. 18% (p = 0.0118) 19. The subsequently performed comparison of two courses of HAM (HAM/HAM) versus the TAD 9/HAM sequence, however, showed no significant differences between HAM-HAM and TAD-HAM in terms of CR rate (71% vs. 65%), RFS at five years (35% vs. 29%), and OS at 5 years (32% vs. 30%) 20. While the escalation of drug doses thus obviously has reached a limit, further intensification of therapy by shortening the time interval between induction cycles appeared as a promising new approach. This strategy was first evaluated in patients with relapsed and refractory AML. Based on prior studies by Burke et al. and Archimbaud et al. 21,22 the HAM regimen was modified into a sequential application of two HAM courses (S-HAM). S-HAM comprises HD-AraC bid on days 1, 2, and Mitoxantrone on days 3, 4; after a rest period of only three days the identical sequence is repeated on days 8 and 9 (HD-AraC) and 10 and 11 (Mitoxantrone), respectively (Figure 1).

The S-HAM protocol was highly effective in patients with advanced disease (primary refractory or relapsed AML) with a CR rate of more than 50% but was complicated by a high early death rate from infections 23, 24,25. Subsequent supportive therapy with G-CSF, however, reduced the duration of critical neutropenia from 40 to 36 days (p=0.008) and the ED rate from 30% to 21% (not significant) 26. First results of dose dense therapy in first line therapy of de-novo AML were gained by a prospective randomised comparison of conventional versus dose dense therapy in children with AML. In the COG (Children Oncology Group) study 2891 dose dense therapy comprising Dexamethasone, Cytarabine, Thioguanine, Etoposide and Rubidomycin (DCTER) given on days 0-4 and 10-14 regardless of response was compared to the standard DCTER regimen given on days 0-4 and 14-18 or later, depending on response. Dose dense treatment resulted in a significantly longer disease-free and overall survival after 3 years of 55 % versus 37 % (p=0.0002) (DFS) and 52±6% versus 42±6 % (OS), respectively 27. In adult patients a French study showed that a similar sequential approach (however not involving high-dose AraC) resulted in a surprisingly low hematological toxicity and a lower cumulative incidence of relapse as compared to conventional induction 28.

These results prompted the AMLCG to assess the efficacy and feasibility of dose-dense therapy with S-HAM in newly diagnosed de novo AML in a phase II study. Supportive therapy with pegfilgrastim was mandatory 29. In addition, a three step escalation of treatment days was planned to obtain total doses of AraC and Mitoxantrone equivalent to the HAM-HAM arm of double induction therapy (Figure 1).
2. PATIENTS AND METHODS

2.1. Patients and Entry Criteria
Adult patients (18 + years) with first diagnosis of de-novo AML (except acute promyelocytic leukemia APL) were eligible for the study. There was no upper age limit. Patients were ineligible in case of severe organ dysfunction not explained by leukemia.

2.2. Cytogenetic Subgroups
For exact characterization of leukaemia bone marrow samples underwent a standardised processing procedure including central sample registration, preparation, and evaluation by cytomorphology, cytochemistry, multiparameter immunophenotyping, cytogenetics, fluorescence in situ hybridization (FISH), and molecular genetics. Favourable cytogenetics were defined as t(8;21) and inv(16). Unfavourable cytogenetics were defined as complex chromosomal aberrations [≥ 3 numerical or structural chromosomal aberrations without involvement of t(15;17), t(8;21) or inv(16)], aberrations of chromosomes 5 or 7, inv(3), involvement of 11q23. Intermediate cytogenetics were defined as normal karyotype or aberrations not qualifying for favourable or unfavourable cytogenetics.

2.3. Treatment Protocol
In the initial phase of the study S-HAM was given as previously used in advanced AML. This S-HAMbasic (66%) regimen – which was identical in dose and schedule to the regimen used in the advanced studies - comprized AraC 3 g/m² as a 3 hours continuous infusion, bid days 1 – 2 and days 8 – 9 (1g/m² in patients 60 + years) and Mitoxantrone 10 mg/m², 30 minutes infusion, days 3 – 4 and days 10 – 11 (Figure 1). A prephase with AraC 100mg/m² as continuous infusion over 24h for up to 7 days was allowed to reach a peripheral leukocyte count < 30.000/µl in order to avoid tumor lysis. 7 days after the completion of treatment a bone marrow aspirate was performed. If ≤ 5% blasts were seen in the aplastic marrow pegfilgrastim 6mg was applied subcutaneously. Pegfilgrastim was repeated every 10 – 12 days until leukocyte recovery (> 1000/µl leukocytes/ > 500/µl neutrophils). Supportive care was provided according to the standards of the local centers.

A dose escalation was planned in order to reach identical doses of AraC and Mitoxantrone in the S-HAM regimen as compared to the HAM-HAM regimen of conventional double induction (100%). Patients were to be treated initially with the S-HAM regimen as known from the advanced AML studies. The doses of this so called S-HAMbasic (66%) regimen amount to 66% of the doses of HAM – HAM. The planned escalation schedule is shown in figure 1. Following the S-HAMbasic (66%) regimen the second dose level [= S-HAMescal (83%)] involved one more day of AraC and Mitoxantrone on days 3 and 4,
respectively, resulting in 83% of the HAM – HAM dose. For the third dose level [S-HAM (100%)], one further day of AraC and Mitoxantrone was planned to be added on days 9 and 10, respectively, resulting in 100% of the HAM – HAM dose (Figure 1).

Postremission therapy was identical to that of the AMLCG 1999 trial with TAD-9 consolidation and monthly myelosuppressive maintenance for three years with alternating 5 day cycles of AraC plus either Thioguanine, Daunourubicin or Cyclophosphamide (AD – AT – AC – AD -...). Patients < 65 years of age with a high or intermediate risk of relapse according to cytogenetics or molecular genetics and an available donor (family or matched unrelated) were to receive an allotransplant.

2.4. Evaluation and Response Criteria
Response criteria were used as proposed by Cheson et al. 2003 30. Patients dying within the first 65 days or while still cytopenic beyond this time were considered early deaths (ED) including those that had residual leukemia. Treatment related side effects were assessed by WHO criteria.

2.5. Data Description and Statistical Analysis
The aim of the present study was to test the feasibility of a dose-dense regimen in adult patients with de-novo AML. Therefore, overall toxicity and the early death rate were of particular relevance. Given an ED rate of 21,3% in 1238 patients with de novo AML treated within the AMLCG 1999 trial 20 and given the potential increase of efficacy achievable by dose-dense S-HAM induction therapy the present trial was to be stopped prematurely if an excessive death rate was observed (> 16% at 60 days; > 23% at 90 days).

In order to put the results of the S-HAM regimen (dose-dense induction) into a perspective a descriptive comparison of the pertinent parameters with data of the AML-CG 1999 trial (conventional double induction) was made. From the large cohort of the AML-CG 1999 trial the following subgroup was selected as a comparator: Patients with de-novo AML who received HAM – HAM double induction. In this group a comparable distribution of cytogenetically defined prognostic subgroups and of the ECOG performance status was found as compared to the S-HAM patient cohort. Since the current phase II study was meant only to demonstrate the feasibility of the dose dense approach in order to allow a future randomized comparison no formal statistical comparison with the historical data was attempted.
2.6. Study Conduct

The study was carried out in accordance with the modified Declaration of Helsinki. All patients gave their informed consent after having been informed about the purpose and the investigational nature of the trial. Before initiation the study received approval of the responsible institutional review board and the ethics committees of the participating institutions. The clinical study is an official study of the “Kompetenznetz Akute und Chronische Leukämien” and is registered in the European Leukemia Trial Registry with the registration number LN_AMLINT_2004_230.
3. RESULTS

3.1. Patient Characteristics:
From August 2004 until January 2008 172 patients with de-novo AML were included into the study and were evaluable for treatment response after induction treatment. Patient characteristics are given in table 1. The median age was 54 years, which is lower than the median age of 58 years in the AML-CG 1999 trial. 40% of patients were \( \geq 60 \) years of age, 12% were older than 70 years.

The majority of patients (83%) had a good or only slightly decreased performance status (ECOG 0 and 1) – table 1. The cytogenetically defined prognostic subgroups were as follows: Favourable 9%, intermediate 69%, unfavourable 22% (table 1). In comparison to the historical control of the AML-CG 1999 trial there was no difference in the distribution (favourable: 12%, intermediate 67%, unfavourable 21%)

3.2. Treatment Delivery:
Out of 172 patients recruited into the study 168 (= 98%) patients received their induction treatment as planned. Four patients did not receive the full treatment due to prohibitive toxicity within the first week of treatment (splenic rupture, pulmonary infiltrates, liver toxicity). Three of these patients received only the first block of treatment – i.e. the equivalent of 1 cycle of HAM, 1 patient had also received 1 day of the 2\textsuperscript{nd} block when his treatment was stopped. These 4 patients are included into the following response analyses on an intention to treat basis.

After the feasibility of the S-HAM\textsubscript{basis} (66%) regimen had been demonstrated in the first 68 patients, the following patients were to receive the S-HAM\textsubscript{escal} (83%) regimen. However, when a significantly longer duration of neutropenia was observed after the S-HAM\textsubscript{escal} (83%) regimen (see 3.3.) this regimen was restricted to patients < 60 years, whereas patients 60 + years went on to receive the S-HAM\textsubscript{basis} (66%) regimen in order to spare older patients from a prolonged duration of neutropenia. In total 113 patients received S-HAM\textsubscript{basis} (66%) and 59 patients S-HAM\textsubscript{escal} (83%).

After observing a 6 day prolongation of critical neutropenia by the addition of one more day of AraC and Mitoxantrone after the S-HAM\textsubscript{escal} (83%) regimen as compared to the S-HAM\textsubscript{basis} (66%) regimen it was decided that the dose-limiting toxicity was reached and a further escalation to S-HAM (100%) was not performed.

Of 143 responding patients 98 patients have a follow-up of > 150 days, which allows an estimation of the feasibility of postremission therapy. Of those patients 75 patients (77%) received their postremission therapy as scheduled. 63 patients (64%) received TAD-9 consolidation – with a median interval between the start of S-HAM induction and start of TAD-9 consolidation of 61 days (range 39 – 154 days). 15 patients (15%) received an
allotransplant as the sole consolidation therapy – with a median interval between the start of S-HAM induction and allogeneic transplantation of 127 days (range 64 – 286 days). Another 5 patients received their allotransplant in CR1 after having had TAD-9 consolidation therapy resulting in a total rate of 20% of patients with allogeneic transplantation in CR1. Reasons for transplantation were cytogenetic high risk (n=9), molecular genetic high risk (mostly FLT3-ITD positivity (n=3), CR with incomplete peripheral recovery - thereby making chemoconsolidation impossible – (n=3), availability of an HLA identical donor in the cytogenetically intermediate risk group (in which case allo-transplantation is regularly offered to patients in the current AML-CG studies) (n=3) and “other” (n=2).

3.3. Non-Hematologic Toxicity:
Non-hematologic grade III and IV toxicities during or following S-HAM induction treatment are given in table 2. As a historical comparison the toxicity data of the AMLCG 1999 trial 20 with double induction treatment are also listed. The major toxicities were serious infections in 40% of patients following S-HAM (56% following double induction in the AML-CG 1999 trial). There was no toxicity that was more frequent during S-HAM than during standard double induction.

When comparing the two evaluable dose levels of S-HAM serious infections were more frequent after S-HAM escal (83%) than after S-HAM basis (66%) - (54% versus 32%, p < 0,01) as was lung toxicity (34% versus 15%, p < 0,01).

3.4. Hematologic Toxicity:
The median time to leukocyte recovery (> 1000/µl) was 31 days after the start of treatment with S-HAM (figure 2a). When compared to de-novo AML patients treated with double induction within the AMLCG 1999 trial (median time to leukocyte recovery 46 days) the total duration of critical leukopenia was more than 2 weeks shorter after S-HAM (figure 2a). When comparing the two dose levels leukopenia after S-HAM escal (83%) was 6 days longer than after S-HAM basis (66%) - (figure 2b, 36 days versus 30 days, p < 0,01).

3.5. Response to Treatment:
A bone marrow aspirate was successfully performed 7 days after the completion of treatment in order to morphologically assess the early blast clearance (EBC) in 159 out of 172 patients. The median percentage of residual blasts was 0% for the whole group. The EBC was higher after the S-HAM escal (83%) regimen as compared to S-HAM basis (66%) with 97% (range 0 – 20%) of patients having ≤ 10% residual blasts as compared to 86% (range 0 – 70%) (p <
0.05). In the AMLCG 1999 trial only 63% of patients had ≤ 10% blasts 7 days after the completion of the (first of two) induction cycles. Response rates for the whole group are given in table 3. An overall response rate (ORR = CR + CRi) of 83% was reached. Treatment failure occurred (ED 7%, PL 10%) in 17%. Responses according to dose level, age or cytogenetic subgroup are given in table 3. For comparison the respective results of the AMLCG 1999 trial are also provided (table 4).

3.6. Overall Survival
The mean follow-up for the 172 patients is 13 months. The overall survival of the whole group is shown in figure 3a. Overall survival according to age, dose level and cytogenetic subgroup is shown in figures 3b - c, respectively. Survival of patients < 60 years at 2 years after S-HAM was 67%, of patients 60 + years 45%. In the AMLCG 1999 trial a 2 year survival of 55% and 30% was reached for patients < 60 and 60 + years respectively. No impact of CRi versus CR on overall survival was noted.
4. DISCUSSION

The current study indicates that the novel strategy of dose-dense intensive induction therapy for de-novo AML with the S-HAM regimen is feasible, is associated with a short period of critical neutropenia and appears highly active. Hence, an early death rate of only 10%, no increase in non-hematologic toxicity as compared to the preceding AMLCG study ’99, a median duration of critical neutropenia of only 31 days and an overall remission rate of 83% was observed in 172 consecutive patients entering this phase II trial.

The approach to investigate a dose-dense therapy for de novo AML emerged from several preceding findings: (1) A historic comparison of different AMLCG studies clearly demonstrated that a shorter time interval between two induction cycles as applied during double induction therapy resulted in a higher CR rate, a longer CR duration and a longer overall survival as compared to the application of the second induction cycle after hematologic recovery. (2) In a prospective randomized comparison the intensively timed DCTER regimen as applied twice with an interval of 6 – 10 days was superior in long term outcome to the same regimens as applied with a 16 day interval in children with AML. (3) Two consecutive studies of the AMLCG in relapsed and refractory AML revealed a high anti-leukemic efficacy of S-HAM and a reduction of the duration of critical neutropenia by the supportive use of filgrastim.

Although the present phase II study primarily tested the feasibility of S-HAM in first line therapy it was tempting to compare the results with the preceding AMLCG ’99 study in which two different forms of conventional double induction therapy were evaluated. This comparison reveals a lower median age of 54 years in the current study as compared to 58 years in the AMLCG 1999 trial but a comparable distribution of cytogenetic and molecular genetics (not shown) as well as of ECOG performance status.

This comparison strongly suggests that S-HAM is obviously not associated with a higher treatment related mortality with an early death rate of only 10% as compared to 21% in the AMLCG 1999 trial. It appears comparable to the preceding study in the frequency and degree of non-hematological toxicities. After S-HAM critical neutropenia was substantially shorter with a reduction by 15 days as compared to standard double induction (31 days versus 46 days). One component of this beneficial effect might be the mandatory application of pegfilgrastim in patients with adequate blast clearance 7 days after the completion of treatment – whereas after double induction the use of filgrastim was only optional and was applied in only 60% of patients. Most likely, however, this effect results from the earlier and potentially more appropriate timing of the second block of treatment. In double induction the second block of treatment is applied on day 21 – at a time when normal hematopoiesis is often about to regenerate and might therefore be particularly vulnerable to cytotoxic agents.
In contrast, in the S-HAM regimen the second block of treatment is applied only 3 days after the first one – at a time when the normal hematopoiesis is still suppressed and less vulnerable to cytotoxic cell damage. Clearly, a two weeks shorter period of critical neutropenia is of substantial clinical relevance and most probably explains the lower rate of grade III/IV infections in S-HAM treated patients versus the historical control (40% versus 55%).

Appropriate timing between cycles appears to be of crucial relevance for treatment related toxicities as well. Hence, the aforementioned intensively timed DCTER – DCTER regimen that was effective in children with AML was too toxic for adult patients when given with an interval of 6 – 10 days. A very similar experience was made by the AML-CG when two cycles of TAD-9 were given with a 7 days interval which resulted in an excessive rate of intestinal toxicity (unpublished data). Both experiences indicate that an around 7 day interval between treatment blocks leads to intolerable toxicity in adults. In accordance with our findings the ALFA 9000 study demonstrated that a 4 days interval between two intensively timed induction cycles in adult patients was associated with surprisingly low toxicity.

The notion that a shortening of the time interval between treatment cycles might be associated with a good tolerability of therapy is also supported by experiences with the FLAMSA protocol in allogeneic transplantation. In this setting an intensive 4 day course of cytarabine, amsacrine and fludarabine is followed after an identical interval of 3 days by reduced intensity conditioning treatment with total body irradiation and cyclophosphamide with only very moderate non-hematological toxicities.

The antileukemic efficacy of the S-HAM regimen was high with an ORR of 83% as compared to the historical control of 66% after double induction (AML-CG 1999, de-novo AML patients) and treatment failure (PL + ED) of 17% as compared to 34%. The difference in antileukemic efficacy was especially pronounced in the rate of early blast clearance (EBC), because in the S-HAM setting 91% of patients had less than 10% residual blast in the bone marrow on day 18 or 19 whereas after double induction this was only the case in 63% of patients on day 16.

With respect to EBC there was a significant difference between the two dose levels with the S-HAM_{escal} (83%) being successful in 97% of patients as compared to 88% after the S-HAM_{basis} (66%) regimen - indicating the substantial antileukemic efficacy of the S-HAM_{escal} (83%) regimen. However since there was no superiority in overall survival for the higher dose level but a trend towards higher toxicity the S-HAM_{basis} (66%) regimen was chosen as the candidate regimen for a future randomized comparison.

Due to the high response rate most patients could proceed to their postremission treatment. This was the case in 83% of responders receiving chemoconsolidation after a median of 61 days after the start of S-HAM. Patients allocated to allogeneic transplantation in
CR-1 underwent this approach after a median of 127 days. This proportion of patients successfully receiving their postremission treatment and the median duration until its initiation also compare very favourably to the experiences after double induction.

As a result the overall survival following S-HAM also looked promising with a two year survival of 67% and 45% for patients < 60 and 60 + years respectively. This also compares favourably to the historical control of de-novo AML patients treated with double induction with a 55% and 30% overall survival at two years for patients < 60 and 60 + years respectively.

In conclusion the dose-dense intensive S-HAM regimen is a highly effective treatment regimen with a response rate of 83% and a low early death rate of 10% in the first 65 days which is most probably due to the short duration of critical neutropenia. In spite of these promising results a prospective randomized comparison of such a dose-reduced but intensely-timed therapy versus conventional double induction is required to prove the potential superiority of the new approach. This trial has been initiated within the next generation of the AML-CG studies (AML-CG 2008).
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Wolfgang Kern collected data
Christian Buske collected data
Stefan Bohlander collected data
Achim Heinecke performed statistical analysis
Herrad Baermann collected data
Dietrich W. Beelen analyzed and interpreted data
Wolfgang E. Berdel designed research
Thomas Büchner designed research
Wolfgang Hiddemann analyzed and interpreted data and wrote the manuscript

None of the authors has a conflict of interest to declare.
6. REFERENCES


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7. LEGENDS

Table 1: Patient characteristics

Table 2: Non-hematological Toxicities (grade III and IV) of S-HAM in Comparison to Double Induction in the AML-CG 99 trial (de-novo AML, age < 60 years, HAM-HAM double induction)

Table 3: Antileukemic efficacy of the S-HAM Regimen for the whole group, according to the respective dose level (66% = S-HAM\textsubscript{basis} (66%); 83% = S-HAM\textsubscript{escal} (83%), according to age and according to cytogenetics (CG).

Table 4: Comparison of responses after S-HAM in the AML-CG 2004 study and standard double induction in the AML-CG 1999 study in de-novo AML according to age and according to cytogenetics.

Figure 1: Schema of the S-HAM regimen and of the planned three step escalation of S-HAM within the AML-CG 2004 Study

Figure 2: A.) Duration of critical leukopenia (< 1000/µl) following S-HAM Induction and subsequent Pegfilgrastim as compared to conventional double induction (HAM-HAM) - % recovery

B.) according to dose level - S-HAM\textsubscript{basis} (66%) - (left) and S-HAM\textsubscript{escal} (83%) - (right) induction

Figure 3: A.) Overall survival of the whole group

B.) according to age (< 60 and 60 + years)

C.) according to dose level [S-HAM\textsubscript{basis} (66%) versus S-HAM\textsubscript{escal} (83%)]

D.) according to karyotype (favourable – blue, intermediate – green, unfavourable – brown)
8. TABLES

<table>
<thead>
<tr>
<th>General Characteristics</th>
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<tr>
<td>Total number of patients</td>
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<td>60 +</td>
<td>68 (39,5%)</td>
</tr>
<tr>
<td>70 +</td>
<td>20 (11,6%)</td>
</tr>
<tr>
<td>Induction as per protocol</td>
<td>168 (97,6%)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>ECOG Performance Status (n = 141)</th>
<th>N (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>50 (35%)</td>
</tr>
<tr>
<td>1</td>
<td>68 (48%)</td>
</tr>
<tr>
<td>2</td>
<td>21 (15%)</td>
</tr>
<tr>
<td>3</td>
<td>2 (1%)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Cytogenetic Subgroup (n = 151)</th>
<th>N (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Favourable</td>
<td>14 (9%)</td>
</tr>
<tr>
<td>Intermediate</td>
<td>104 (69%)</td>
</tr>
<tr>
<td>Unfavourable</td>
<td>33 (22%)</td>
</tr>
</tbody>
</table>

Table 1: Patient characteristics
<table>
<thead>
<tr>
<th>Tox. III°/IV°</th>
<th>S-HAM AML-CG 2004</th>
<th>HAM-HAM AML-CG 1999</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nausea</td>
<td>9%</td>
<td>16%</td>
</tr>
<tr>
<td>Stomatitis</td>
<td>3%</td>
<td>9%</td>
</tr>
<tr>
<td>Diarrhea</td>
<td>14%</td>
<td>14%</td>
</tr>
<tr>
<td>Hemorrhage</td>
<td>8%</td>
<td>8%</td>
</tr>
<tr>
<td>Infection</td>
<td>40%</td>
<td>56%</td>
</tr>
<tr>
<td>Cardiac Ev.</td>
<td>6%</td>
<td>5%</td>
</tr>
<tr>
<td>CNS Toxic.</td>
<td>5%</td>
<td>5%</td>
</tr>
</tbody>
</table>

Table 2: Non-hematological Toxicities (grade III and IV) of S-HAM in Comparison to Double Induction in the AML-CG 1999 trial (de-novo AML, age < 60 years, HAM-HAM double induction)
<table>
<thead>
<tr>
<th>Treatment Response</th>
<th>CR</th>
<th>CRi</th>
<th>PL</th>
<th>ED</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Whole Group</td>
<td>n</td>
<td>105</td>
<td>38</td>
<td>12</td>
<td>17</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>61</td>
<td>22</td>
<td>7</td>
<td>10</td>
</tr>
<tr>
<td>Dose</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>66%</td>
<td>n</td>
<td>72</td>
<td>24</td>
<td>10</td>
<td>7</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>64</td>
<td>21</td>
<td>9</td>
<td>7</td>
</tr>
<tr>
<td>83%</td>
<td>n</td>
<td>33</td>
<td>14</td>
<td>2</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>56</td>
<td>24</td>
<td>3</td>
<td>17</td>
</tr>
<tr>
<td>Age</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt; 60</td>
<td>n</td>
<td>69</td>
<td>17</td>
<td>7</td>
<td>12</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>66</td>
<td>16</td>
<td>7</td>
<td>11</td>
</tr>
<tr>
<td>60 +</td>
<td>n</td>
<td>36</td>
<td>21</td>
<td>5</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>54</td>
<td>31</td>
<td>7</td>
<td>7</td>
</tr>
<tr>
<td>CG</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Favor.</td>
<td>n</td>
<td>8</td>
<td>6</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>53</td>
<td>40</td>
<td>0</td>
<td>7</td>
</tr>
<tr>
<td>Inter.</td>
<td>n</td>
<td>68</td>
<td>20</td>
<td>5</td>
<td>7</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>68</td>
<td>20</td>
<td>5</td>
<td>7</td>
</tr>
<tr>
<td>Unfav.</td>
<td>n</td>
<td>18</td>
<td>5</td>
<td>6</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>55</td>
<td>15</td>
<td>18</td>
<td>12</td>
</tr>
</tbody>
</table>

Table 3: Antileukemic efficacy of the S-HAM Regimen for the whole group, according to the respective dose level [66% = S-HAM_{basis} (66%); 83% = S-HAM_{escal} (83%)], according to age and according to cytogenetics (CG).
<table>
<thead>
<tr>
<th>Age</th>
<th>AML-CG</th>
<th>ORR %</th>
<th>PL %</th>
<th>ED %</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 60</td>
<td>1999</td>
<td>72</td>
<td>11</td>
<td>17</td>
</tr>
<tr>
<td></td>
<td>2004</td>
<td>82</td>
<td>7</td>
<td>11</td>
</tr>
<tr>
<td>60 +</td>
<td>1999</td>
<td>59</td>
<td>15</td>
<td>26</td>
</tr>
<tr>
<td></td>
<td>2004</td>
<td>85</td>
<td>7</td>
<td>7</td>
</tr>
<tr>
<td>Cytogenetics</td>
<td>AML-CG</td>
<td>ORR %</td>
<td>PL %</td>
<td>ED %</td>
</tr>
<tr>
<td>Favourable</td>
<td>1999</td>
<td>74</td>
<td>6</td>
<td>20</td>
</tr>
<tr>
<td></td>
<td>2004</td>
<td>93</td>
<td>0</td>
<td>7</td>
</tr>
<tr>
<td>Intermediate</td>
<td>1999</td>
<td>71</td>
<td>12</td>
<td>17</td>
</tr>
<tr>
<td></td>
<td>2004</td>
<td>88</td>
<td>5</td>
<td>7</td>
</tr>
<tr>
<td>Unfavourable</td>
<td>1999</td>
<td>45</td>
<td>25</td>
<td>30</td>
</tr>
<tr>
<td></td>
<td>2004</td>
<td>70</td>
<td>18</td>
<td>12</td>
</tr>
</tbody>
</table>

Table 4: Comparison of responses after S-HAM in the AML-CG 2004 study and standard double induction in the AML-CG 1999 study in de-novo AML according to age and according to cytogenetics.
9. FIGURES

<table>
<thead>
<tr>
<th>S-HAMbasic (68 %)</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>AraC</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mitoxantron</td>
<td>↑↑</td>
<td>↑↑</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>S-HAMesc (83 %)</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>AraC</td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Mitoxantron</td>
<td>↑↑</td>
<td>↑↑</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td></td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>S-HAM (100 %)</th>
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<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>AraC</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mitoxantron</td>
<td>↑↑</td>
<td>↑↑</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
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<td></td>
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</tr>
</tbody>
</table>

**Check for feasibility and safety**

**Check for feasibility, safety and efficacy**

**Equal doses to HAM – HAM double induction therapy**

<table>
<thead>
<tr>
<th>HAM – HAM double induction therapy</th>
<th>Week 1</th>
<th>Week 2</th>
<th>Week 3</th>
<th>Week 4</th>
<th>Week 5</th>
<th>Week 6</th>
<th>Week 7</th>
<th>Week 8</th>
</tr>
</thead>
<tbody>
<tr>
<td>AraC</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mitoxantron</td>
<td>↑↑</td>
<td>↑↑</td>
<td></td>
<td></td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Figure 1: Schema of the S-HAM regimen and of the planned three step escalation of S-HAM within the AML-CG 2004 Study
Figure 2:  
A.) Duration of critical leukopenia (< 1000/µl) following S-HAM Induction and subsequent Pegfilgrastim as compared to conventional double induction (HAM-HAM) - % recovery  
B.) according to dose level - S-HAM\textsubscript{basis} (66%) - (left) and S-HAM\textsubscript{escal} (83%) - (right) induction
Figure 3:  
A.) Overall survival of the whole group in days  
B.) according to age (< 60 and 60 + years)  
C.) according to dose level [S-HAM\textsubscript{basis} (66%) versus S-HAM\textsubscript{escal} (83%)]  
D.) according to karyotype (favourable, intermediate, unfavourable)
Dose-dense induction with sequential-high-dose cytarabine and mitoxantone (S-HAM) and pegfilgrastim results in a high efficacy and a short duration of critical neutropenia in de-novo acute myeloid leukemia - a pilot study of the AML-CG

Jan Braess, Karsten Spiekermann, Peter Staib, Andreas Gruneisen, Bernhard Wormann, Wolf-Dieter Ludwig, Hubert Serve, Albrecht Reichle, Rudolf Peceny, Daniel Oruzio, Christoph Schmid, Xaver Schiel, Marcus Hentrich, Christina Sauerland, Michael Unterhalt, Michael Fiegli, Wolfgang Kern, Christian Buske, Stefan Bohlander, Achim Heinecke, Herrad Baurmann, Dietrich W. Beelen, Wolfgang E. Berdel, Thomas Buchner and Wolfgang Hiddemann

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