Anthracycline dose intensification in young adults with acute myeloid leukemia

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Ther Adv Hematol

(2011) 3(1) 17–27

DOI: 10.1177/

2040620711427069

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Abstract: The initial treatment for acute myeloid leukemia (AML) has remained largely unchanged for nearly 40 years. Our growing understanding of the molecular pathology of AML has resulted in improved measures to risk stratify patients by recurrent cytogenetic and molecular abnormalities without marked advancement in its initial treatment. The most common regimen consists of a 3-day course of an anthracycline and a 7-day infusion of cytarabine. This regimen has been employed across the globe in various iterations for many years with modest improvements in results yet this remains the first choice for the treatment of younger adults with AML. Despite this, the chemotherapeutic agents in this regimen are only now being fully understood. Recent evidence has suggested that dose intensification of anthracycline in young adults has a significant survival benefit. In this paper we review the evidence behind the use of anthracyclines in the initial induction of AML in younger adults focusing on the choice and dose of this long used drug combination.

Keywords: acute myeloid leukemia, induction therapy, dose intensification

Introduction

Acute myeloid leukemia (AML) refers to a collection of neoplasms arising from a clonal, myeloid-committed, hematopoietic precursor whose behavior is characterized by a dramatic proliferative advantage and maturation arrest [Fialkow *et al.* 1979, 1981]. This behavior clinically manifests as an abnormal increase in myeloblasts in the bone marrow, peripheral blood, and occasionally extramedullary sites. The natural history of AML is not subtle. Overt symptoms of bone marrow failure secondary to *de novo* AML are thought to occur weeks to months after the initiation of leukemogenesis.

The incidence of AML is 5 per 100,000 in the United States and represents one of the most common forms of adult leukemia. Although AML can present at any age, the incidence of the disease increases to 12 per 100,000 in those over 65 years. The median age at diagnosis is approximately 65 years and the male to female ratio is 5:3 [Yamamoto *et al.* 2008]. Age remains the most powerful risk factor in AML but other environmental factors have been implicated. Ionizing radiation to the bone marrow, cytotoxic chemotherapy, antecedent hematologic malignancies, and benzene exposure are some of the associated risk factors for the development of AML [Austin

et al. 1988; Stone *et al.* 1994; Shuryak *et al.* 2006]. The recent description of molecular abnormalities in AML has led to an evolving system of AML classification under the World Health Organization (WHO) that incorporates morphologic, cytogenetic, and genetic data to more appropriately classify the disease based on disease-initiating commonalities and prognostic markers [Vardiman *et al.* 2009].

The genetic and epigenetic events that are responsible for leukemogenesis are heterogeneous. Like the 8;21 translocation and AML/ETO1, multiple recurring chromosomal and genetic aberrations have led to monumental advances in our understanding of this disease. Clinically, cytogenetic abnormalities and genetic lesions such as *FLT3*-ITD, *NPM1*, *CEBP- α* , and *c-KIT* mutations carry important prognostic significance [Santamaria *et al.* 2009]. However, outside of acute promyelocytic leukemia, our understanding of the genetic events in AML has yet to translate into effective, targeted therapy.

Initial therapy

The most widely used induction therapy in the United States remains the so-called ‘7 + 3’ regimen. This has traditionally combined a 7-day

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infusion of cytarabine with a 3-day administration of an anthracycline. For almost 40 years, this combination has remained the mainstay of induction therapy in AML and yielded initial response rates from 50% to 75% [Yates *et al.* 1973; Vogler *et al.* 1992; Wiernik *et al.* 1992]. Several strategies have improved the complete remission (CR) and relapse rates in AML and are employed based on risk stratification, age, and institutional preferences. Double induction along with consolidation, maintenance therapy, and allogeneic stem cell transplantation (ASCT) can improve the 4-year overall survival to 60% in the best risk AML subgroups [Mayer *et al.* 1994].

Double induction with a '7 + 3' based regimen was explored by a German group who enrolled 782 participants under the age of 60 with *de novo* AML. Here, patients were randomized to two courses of induction, irrespective of their remission status after the first induction, followed by consolidation therapy if not suitable for transplantation. There was a significant improvement in relapse-free survival for those receiving double induction; however this did not translate into an overall survival benefit [Buchner *et al.* 1999].

The addition of agents to the 7 + 3 induction therapy backbone or the intensification of cytarabine has failed to show appreciable improvements in the duration or rate of response [Weick *et al.* 1996]. The type, dose, and intensification of anthracycline have been an area of active research and is the focus of this review. Although daunorubicin (DNR) and idarubicin (IDR) have been the workhorses of this class, we cover the pertinent evidence in support of mitoxantrone, amsacrine, and adriamycin to a lesser extent. We also discuss the evidence for and against dose intensification as well as consider this evidence in the context of age and AML risk stratification.

'Standard dose' daunorubicin for induction therapy

In 1972, a new class of anthracycline antibiotic was introduced to the myeloid leukemia armamentarium. A total of 22 adult patients with AML received 60 mg/m² over 3 days of DNR, and 21 patients received a combination of prednisone, vincristine, 6-mercaptopurine, and methotrexate (POMP) in a randomized study conducted at the National Cancer Institute [Wiernik *et al.* 1972]. DNR was superior to the combination in inducing CR (50 *versus* 28%), improved median time to

response (19 days), and treatment related mortality. Similar results were being seen with cytarabine with response rates varying between 25% and 50%. During this time, impressive strides were being made with combination therapy in acute lymphoblastic leukemia making the combination of cytarabine and DNR rational.

Small clinical trials began reporting this combination using varying doses of DNR and cytarabine for two and five treatment schedules, respectively. It was not until Yates and colleagues published a pilot study describing 13 of 15 AML patients achieving a complete response who received 3 days of DNR and 7 days of cytarabine that modern induction treatment was established [Yates *et al.* 1973]. This regimen was formally studied by the CALGB in 1981. Kanti Rai led a four-arm clinical trial evaluating 352 patients who were randomized to receive DNR (45 mg/m²) and cytarabine (100 mg/m²) in a 2 + 5 or 7 + 3 schedule by bolus or infusion [Rai *et al.* 1981]. Both 7 + 3 regimens were superior ($p < 0.01$) to their 2 + 5 counterparts. The infusional 7 + 3 regimen had the most robust response at 71%. The 7 + 3 and 2 + 5 arms were equally toxic but the 7 + 3 arm yielded less deaths during induction across all ages presumably secondary to improved blast clearance. This landmark trial established 3 days of DNR at 45 mg/m² with a 7-day infusion of cytarabine at 100 mg/m² as the treatment of choice for AML for the better part of 30 years.

Jerome Yates led a second CALGB study in 1982 that confirmed the superiority of this regimen [Yates *et al.* 1982]. In this study, 653 patients were randomized to receive the conventional 7 + 3 backbone with DNR at 30 mg/m², DNR at 45 mg/m², or adriamycin (ADM) at 30 mg/m². Results from this study confirmed the superior response rates (72%) using DNR45 in patients less than 60 years of age. It also demonstrated improved response rates in this age group as compared with DNR30 (59%) and ADM30 (58%) with similar toxicities and treatment-related mortality. The ADM arm, however, did have significantly increased incidences of gastrointestinal adverse events.

Dose intensification of daunorubicin

Efforts to improve the response rate, duration of response, and overall survival had previously concentrated on dose intensification of cytarabine and the addition of conventional and molecularly

targeted chemotherapeutic agents to the 7 + 3 backbone with negative results [Weick *et al.* 1996]. Increased doses of DNR had been reported in the literature but not formally studied until recently. Schiller and colleagues published a clinical trial testing cytarabine dose intensification in which 60 mg/m² of DNR was given with response rates on the order of 67% and toxicities comparable to lower dose DNR historical controls [Schiller *et al.* 1992]. In addition, results of phase I and II studies had previously suggested that DNR doses of 70–95 mg/m² for 3 days were safe and improved the rate of CR [Appelbaum *et al.* 1984; Kolitz *et al.* 2004].

To resolve the DNR dose intensification question, the ECOG randomized 657 newly diagnosed AML patients younger than 60 years of age to receive 7 + 3 induction with DNR at 45 mg/m² or DNR at 90 mg/m². Taken as a whole, high-dose DNR, as compared with a standard dose of the drug, resulted in a higher rate of CR (70.6% *versus* 57.3%, $p < 0.001$) and improved overall survival (median, 23.7 *versus* 15.7 months; $p = 0.003$). However, a subset analysis showed that this benefit was restricted to those under the age of 50 and with low- or intermediate-risk cytogenetics [Fernandez *et al.* 2009].

Considering this new evidence, it now appears that if DNR is given for induction therapy in AML, one should administer 90 mg/m² in patients who are of young age with favorable or intermediate-risk cytogenetics. It remains unclear whether high-dose DNR should be categorically recommended to those with poor cytogenetic or molecular (FLT3-ITD) risk. It is also unclear whether other dose of DNR between 45 and 90 mg/m² are as efficacious as 90 mg/m² as this has not been investigated prospectively.

Other anthracyclines

Idarubicin in induction therapy

IDR, a lipophilic analogue of DNR, has been extensively tested as part of the 7 + 3 backbone in AML. It is structurally identical to DNR except for a change at position 4 of its chromophore ring conferring a higher lipophilic coefficient, induction of more DNA single-strand breaks in tumor cells, and an active metabolite with a longer half-life as compared with DNR *in vitro* [Zunino *et al.* 1976; Supino *et al.* 1977; Plumbidge *et al.* 1978; Speth *et al.* 1986]. After phase I testing revealed a

myelosuppressive dose-limiting toxicity of 12 mg/m², phase II studies were performed in Italy, France, and the United States demonstrating that IDR as a single agent induced CR in 13–22% of adult patients with relapsed or refractory AML [Hayat *et al.* 1984; Daghestani *et al.* 1985; Gillies *et al.* 1987]. IDR was combined with cytosine arabinoside (Ara-C) and the response rate increased to the range of 24–70% in similar groups of heavily pretreated patients [Lambertenghi-Deliliers *et al.* 1987; Berman *et al.* 1989].

Subsequently, a phase III trial at Memorial Hospital in New York City randomized 130 patients under the age of 60 with newly diagnosed AML to receive 7 + 3 with 50 mg/m² of DNR or 12 mg/m² of IDR. Analyses revealed that 48 of 60 patients (80%) achieved CR on the IDR arm compared with 35 of 60 patients on the DNR arm (58%, $p = 0.005$). Overall survival for patients on the IDR/Ara-C arm was 19.5 months compared with 13.5 months on the DNR/Ara-C arm ($p = 0.025$) at a median follow up of 2.5 years [Berman *et al.* 1991]. The authors concluded that this should now replace DNR as the standard of care for newly diagnosed AML.

To confirm the results of this single-institution trial, the Southeastern Cancer Study Group performed a multi-institution phase III trial comparing 7 + 3 with 45 mg/m² of DNR or 12 mg/m² of IDR. Here 230 patients under the age of 60 with newly diagnosed AML were randomized to the above arms. CR rates were 69% (75 of 111) on the IDR arm and 55% (65 of 119) on the DNR arm ($p = 0.031$). However, in this trial, a superior overall survival in the IDR arm did not reach statistical significance. There were no statistical differences in toxicities except for more diarrhea ($p = 0.014$), elevation of bilirubin ($p = 0.032$), serum glutamic oxaloacetic transaminase (SGOT; $p = 0.003$), alkaline phosphatase ($p = 0.01$), and blood urea nitrogen (BUN; $p = 0.007$) levels on the IDR arm [Vogler *et al.* 1992].

A third multi-institutional clinical trial was conducted that randomized 214 patients younger than 60 years of age with newly diagnosed AML to 13 mg/m² of IDR or 45 mg/m² of DNR. Again the CR rates for evaluable patients was 70% in the IDR arm and 59% DNR arm ($p = 0.08$). A subset analyses revealed that the difference in CR rates was significant in patients aged under 50 years (88% for IDR, 70% for DNR, $p = 0.035$) [Wiernik *et al.* 1992].

To systematically consolidate this data, the AML collaborative group looked at IDR in 1052 patients in five trials *versus* DNR, 100 in one trial *versus* doxorubicin, and 745 in one trial *versus* zorubicin. In the trials of idarubicin *versus* DNR, CR rates were higher with idarubicin (62% *versus* 53%; $p = 0.002$). Overall survival in these five trials was significantly better with idarubicin than with DNR (13% *versus* 9% alive at 5 years; $p = 0.03$) despite a slight, but not significant, increase in treatment-related deaths in the IDR arm. There was a trend ($p = 0.006$ for remission rate) for the benefit of idarubicin over DNR to decrease with increasing age [AML Collaborative Group, 1998].

Despite what appears to be conclusive evidence for the use of IDR over DNR in patients under the age of 60 with newly diagnosed AML, controversy remains. It is unclear whether the apparent advantage of IDR over DNR is a result of inherent properties of IDR or merely a dose intensification of IDR over DNR. The dose of 45 mg or 50 mg of DNR may be a 'less toxic anthracycline dose' than 12 or 13 mg of IDR but at the cost of improved survival. The recent evidence suggesting improved survival with dose intensification in DNR begs the comparison of 90 mg of DNR with 12 or 13 mg of IDR in patients under the age of 60. The Japanese Acute Leukemia Study Group recently compared 50 mg/m² for 5 days of DNR or 12 mg/m² for 3 days of idarubicin along with conventional-dose cytarabine [Ohtake *et al.* 2011]. Although not exactly high-dose DNR as studied in the ECOG trial, the cumulative dose of DNR was similar. A total of 1064 patients were randomized and results revealed that DNR was noninferior to IDR. CRs were achieved in 407 (77.5%) of 525 patients in the DNR group and 416 (78.2%) of 532 in the idarubicin group ($p = 0.79$). Outside of the FAB-M6 subtype in which the IDR arm enjoyed a superior CR rate, no other WHO or FAB group showed statistical differences. Overall responses were also similar as were grade 3 and 4 toxicities although the IDR group had more episodes of sepsis and longer hospitalization. The authors concluded that high-dose DNR was noninferior to IDR.

Further, the Acute Leukemia French Association 9801 (ALFA-9801) addressed this by randomizing AML patient age 50–70 to receive high doses of DNR (80 mg/m²/day $\times$ 3 days), idarubicin (IDA3; 12 mg/m²/day $\times$ 4 days), and standard doses of idarubicin (IDA4; 12 mg/m²/day $\times$ 3 days) as part of the 7 + 3 backbone. The overall

CR rate was 83% in the IDA3 arm, 78% in the IDA4 arm, and 70% in the DNR arm. However, no differences were seen in the primary endpoint, event-free survival, or overall survival [Pautas *et al.* 2010].

Mitoxantrone in induction therapy

Mitoxantrone (MTZ), an anthracenedione, is a synthetic analogue of DNR found to have similar antileukemic properties to DNR *in vitro* but with decreased cardiac toxicity observed in a beagle dog model [Henderson *et al.* 1982]. This agent has been tested in a variety of iterations to include part of the 7 + 3 backbone, in combination with etoposide, and in combination with high-dose cytarabine. A single-institution phase III trial explored the use of mitoxantrone as part of the 7 + 3 backbone. In this trial, 200 patients were randomized to receive 7 + 3 with 12 mg/m² of mitoxantrone or 45 mg/m² of DNR. A total of 63% (62 of 98) of patients treated with mitoxantrone achieved CR, compared with 53% (54 of 102) treated with DNR. The median length of survival was 328 days in patients who received mitoxantrone and 247 days in those who received DNR. Toxicities were comparable in both arms [Arlin *et al.* 1990].

A more recent trial compared the addition of MTZ to 7 + 3 with 80 mg/m² of DNR in an induction, double induction or time sequential induction schema. Here, AML patients under the age of 65 with *de novo* disease were randomized to receive conventional induction therapy, double induction with the substitution of 12 mg/m² of MTZ on days 20–21, or time sequential induction with the substitution of 12 mg/m² of MTZ on days 8–9. There were no significant differences in complete response or 5-year overall survival at about 75% and 30%, respectively. However, subgroup analysis did reveal an improvement in relapse-free survival in the both intensification arms in patients under age 50. Surprisingly, there were also no differences in treatment-related mortality or duration of consolidation [Castaigne *et al.* 2004].

Mitoxantrone and etoposide has also been explored in the relapsed or refractory setting in a phase II clinical trial. A total of 68 patients with relapsed or refractory AML received 10 mg/m² of mitoxantrone with 100 mg/m² of etoposide over 5 days. A total of 26 patients (42.6%) attained a CR and seven (11.5%) a partial remission (PR). Two cases of early death within 6 weeks were

reported and 33% of patients suffered from grade 3 infections. Outside of this, this regimen was well tolerated and thus considered active in the relapsed or refractory setting [Ho *et al.* 1988].

Mitoxantrone has been paired with high-dose cytarabine (HDAC) in a phase I/II trial of relapsed or refractory AML patients. Therapy consisted of HDAC 3 g/m² every 12 hours on days 1–4 and mitoxantrone at 12 mg/m² on days 3, 4, and 5. A total of 40 patients were enrolled in this study with 21 patients achieving a CR (53%), 1 patient achieving a PR and 5 patients were nonresponders. Thirteen patients died in aplasia due to infections, pericardiac effusion, or acute cardiomyopathy [Hiddemann *et al.* 1987]. Despite its apparent toxicities, this regimen is active and used frequently in Europe.

In general, mitoxantrone appears to be an active agent in AML and has been studied in multiple regimens. However, there is insufficient data to routinely recommend mitoxantrone as a frontline agent in AML. Moreover, no study has demonstrated the superiority of mitoxantrone over IDR or DNR. Its use appears to be restricted to patients who have failed initial induction and can tolerate further therapy.

Amsacrine in induction therapy

Amsacrine is another anthracycline with both direct DNA cleavage and topoisomerase II inhibitory properties. It has been studied to a lesser extent in AML with encouraging but sparse results [Legha *et al.* 1980; Louie *et al.* 1985]. This is highlighted by a single-institution study at Memorial Hospital where 96 patients with newly diagnosed AML were randomized to receive 50 mg/m² of DNR or 190 mg/m² of amsacrine in combination with bolus cytarabine and thioguanine [Berman *et al.* 1989]. Although this trial suggested equivalent efficacy and toxicity, no prospective study has evaluated this agent as part of the 7 + 3 backbone across multiple institutions and thus there is insufficient data to advocate its use. Although it appears active in AML, there exist other, better studied anthracyclines that could be chosen. Once patients fail DNR, IDR, and mitoxantrone it is unclear whether this agent would be of any benefit and would, in fact, likely be prohibitive secondary to the cumulative cardiac toxicity of this drug class. Considering this, it would be hard to imagine a recurring scenario in which this drug could be widely used in AML.

Gemtuzumab ozogamicin

Gemtuzumab ozogamicin (GO) has also been tested as part of the 7 + 3 regimen. GO is a calicheamicin-linked monoclonal antibody to CD33. Calicheamicin is a DNA intercalating agent similar to the other anthracyclines. The dose-limiting toxicity in phase I trials, unbound to the monoclonal antibody, was liver toxicity; however, this toxicity was attenuated in the antibody-linked delivery to AML cells. Two large trials have failed to show improved response or survival when gemtuzumab was added to 7 + 3. In the NCRI trial the addition of GO to 50 mg of DNR did demonstrate superior outcomes in the favorable cytogenetic risk group. A recently presented SWOG study of 627 AML patients randomized to receive the DNR 45/cytarabine with GO or DNR60/cytarabine without GO did not demonstrate an improved overall survival. Moreover, the patients who received GO had higher treatment-related toxicity. These results led to the voluntary removal of GO from the US market [Deangelo *et al.* 2003; Burnett *et al.* 2006, 2009; Petersdorf *et al.* 2009]. Of note the control arm used DNR 60 mg/m² and produced similar CR rates as reported with higher doses of DNR or IDR. In light of the increased toxicity, the addition of GO does not seem to be a superior approach to DNR 90 mg/m² even in the favorable risk group.

Is there a threshold effect of anthracycline?

The question regarding the anthracycline of choice and dose in AML remains open. The European Organization for Research and Treatment of Cancer Leukemia Group and the Gruppo Italiano Malattie Ematologiche dell'Adulto (EORTC/GIMEA) has attempted to answer this issue by randomizing 2157 patients with AML under the age of 60 years to receive cytarabine at 25 mg/m² as intravenous bolus followed immediately by 100 mg/m² given as a continuous infusion daily for 10 days, etoposide 100 mg/m², and on days 1, 3, and 5, DNR 50 mg/m², mitoxantrone 12 mg/m², or idarubicin 10 mg/m². The complete response rates and 5-year overall survivals were similar at approximately 70% and 30% respectively. However, disease-free survival was significantly shorter with the DNR- and mitoxantrone-containing regimens allowing the authors to recommend IDR as the anthracycline of choice [Mandelli *et al.* 2009]. The remission rates for all three induction arms were lower than what has been seen in other trials with the higher-dose IDR or DNR. This recommendation was

Table 1. Idarubicin and daunorubicin comparisons across large randomized clinical trials. [Adapted from Fernandez [2010].]

Clinical Trial	Dosing	Complete remission rate (%)	Overall survival
Daunorubicin			
ECOG	90 mg/m ² x 3 days	70.1	23.7 mo median
SWOG	60 mg/m ² x 3 days	74	35 mo median
JALSG	50 mg/m ² x 5 days	77.5	49% at 4 years
EORTC/GIMEMA	50 mg/m ² x 3 days	68.7	36% at 5 years
ALFA 9000	80 mg/m ² x 3 days	77	28% at 5 years
ALFA 9801*	80 mg/m ² x 3 days	70	NR
MRC AML15	50 mg/m ² on 1, 3, 5 days	83	42% at 5 years
Idarubicin			
JALSG	12 mg/m ² x 3 days	78	53% at 4 years
EORTC/GIMEMA	10 mg/m ² x 3 days	67	45% at 5 years
ALFA 9801*	12 mg/m ² x 3 days (or 4 days)	83 or (74)	NR
*Patients aged 50–70 years.			
NR, not reported.			

made using what is now recognized as attenuated doses of DNR and thus makes extrapolation to what is already known about high-dose (90 mg/m²) DNR difficult.

Lastly, the ALFA group performed another trial where 468 AML patients under the age of 70 were randomized to receive a modified 7 + 3 schedule that included cytarabine at 200 mg/m² with either DNR at 80 mg/m² or IDR at 12 mg/m² for 3 or 4 days. CR was achieved in 70% of patients in the DNR arm, 83% in the IDR 3-day arm, and 78% in the IDR 4-day arm ($p = 0.02$). There were more grade 3 and 4 episodes of mucositis in the IDR arms and no differences in overall survival were seen [Pautas *et al.* 2007]. Despite the apparent similarity in induction remission rates between IDR 12 mg/m² and the higher DNR dosing, we can only recommend that DNR at 90 mg/m² be used as initial therapy for induction as it is the only anthracycline to have demonstrated an improved overall survival.

So is there a threshold effect of anthracycline in induction therapy? The most recently completed trials suggest DNR doses above 60 mg/m² produce equivalently high CR rates. The EORTC/GIMEMA trial presented results consistent with the theory that lower doses of anthracycline result in inferior control of the disease. The most recent trials with higher doses of anthracycline resulted in CR rates greater than 70%. Until 90 mg/m² of DNR is prospectively compared with

IDR, both will remain viable options in the young adult with AML (Table 1).

What is clear is that patients above the age of 50, and certainly above 60, begin to lose benefit from anthracycline dose intensification. The Swiss, ECOG, and EORTC trials all demonstrate that any CR or overall survival benefit is lost as age increases in AML patients. It also appears that the unfavorable cytogenetic risk group fails to benefit from dose intensification. Thus, in patients under the age of 60, dose intensification of the anthracycline with the unfavorable risk group does not guarantee a better outcome.

Next-generation anthracycline-based therapies in AML

Multiple strategies to improve outcomes in the treatment of AML patients are currently being investigated. Among these are the addition of kinase inhibitors, epigenetic modifiers, anthracycline efflux pump inhibitors, and calicheamicin-linked antibodies to the conventional approach. Combining targeted agents with varying iterations of 7 + 3 to relevant subpopulations of AML appears promising. *FLT3*-ITD is a tyrosine kinase responsible for initiating the transcription of many proteins involved in B- and T-cell development and is the most frequently found genetic alteration in AML occurring at a rate of 25–30% [Yokota *et al.* 1997]. AML patients with this mutation in the background of normal

cytogenetics tend to have a worse disease-free survival and overall survival [Kottaridis *et al.* 2001]. Multiple inhibitors of this kinase are in experimental stages of development. Midostaurin, a multikinase inhibitor with FLT3 activity, has already shown the ability to decrease tumor burden of FLT3 mutated AML in a phase II study led by the CALGB [Barry *et al.* 2007]. A phase III clinical trial (CALGB 10603) is actively recruiting FLT3 mutated patients to be randomized to 7 + 3 with 60 mg/m² of DNR with or without midostaurin. Sorafenib, another kinase inhibitor with FLT3 activity, has been tested by the MD Anderson Leukemia Department. A total of 51 patients with untreated AML under the age of 65 received cytarabine at 1.5 g/m² by continuous intravenous infusion for 4 days, idarubicin at 12 mg/m² for 3 days, and sorafenib at 400 mg orally twice daily for 7 days. Impressively, 14 patients with FLT3-ITD mutations had a 93% response rate, much higher than historical controls in the first induction [Ravandi *et al.* 2010].

Nuclear factor-kappa B (NF- κ B) is a master transcription factor responsible for upregulating many survival proteins and is highly expressed in leukemia-initiating cells traditionally resistant to cytotoxic therapies [Guzman *et al.* 2001]. Bortezomib is a proteasome inhibitor that is known to inhibit NF- κ B. A recently completed phase I trial evaluated giving 31 patients 7 + 3 with IDR along with dose escalation of bortezomib. The authors concluded that this combination was safe with acceptable toxicities up to a dose of 1.5 g/m². A phase II study is now ongoing in the CALGB [Attar *et al.* 2008].

A major component of chemotherapy resistance is mediated via the p-glycoprotein efflux pump (P-gp). P-gp has been reported in nearly three quarters of AML patients older than age 55 years and expression has been associated with poor-risk cytogenetic findings [Leith *et al.* 1997; Van Den Heuvel-Eibrink *et al.* 1997]. A SWOG study randomized 410 patients to receive several induction regimens with or without PSC-833, a potent inhibitor of P-gp. No differences were seen in overall survival or response rate with a worse toxicity profile in the PSC-833 arm. However, a subset analysis did show a statistically superior outcome in overall survival and disease-free survival in those less than 45 years of age [Kolitz *et al.* 2004].

Lastly, novel ways to optimize delivery of the anthracycline has been investigated. CPX-351

fixes a 5:1 molar ratio of cytarabine to DNR within a liposomal membrane. Laboratory studies revealed that CPX-351 was maintained in murine bone marrow for more than 24 hours and produced increased survival of murine leukemia-bearing mice compared with conventional cytarabine and DNR [Tardi *et al.* 2009]. A phase I study has been completed which showed the safety of CPX-351 in AML patients [Feldman *et al.* 2011]. A phase II study has been reported at the American Society of Hematology annual meeting suggesting a CR rate of approximately 60% that was most impressive particularly in those with a prior history of myelodysplastic syndrome and poor-risk cytogenetics [Lancet *et al.* 2010]

Conclusions

Anthracyclines continue to be a cornerstone for the initial treatment of AML. Despite its wide use in AML for over 40 years, the dose, type, and route of anthracycline is only now becoming clearer. Current evidence suggests that dose intensification of DNR is beneficial in the younger cohort of AML patients. The higher doses are safe and produce higher CR rates. The toxicity is not increased allowing more patients to receive the appropriate consolidation/curative therapy. However, trials are needed to clarify whether this higher dose is superior to IDR. Although novel mechanisms of drug delivery carry promise, larger studies are needed to prove their superiority. Despite these advances there still remains a gap in the patients benefiting from the dose intensification strategy (older, unfavorable, FLT3-ITD mutated, etc.). In addition, these therapies must complement the new standard therapy of higher anthracyclines without increasing toxicity.

Perhaps our deeper, molecular understanding of AML has made this task more attainable. It is now clear that AML is the result of a set of genetic alterations that exhibit a similar phenotype through differing molecular pathologies. This heterogeneity will present challenges to investigators as it is unlikely future therapies (new agents and new combinations of agents) will comprehensively benefit all AML patients. Using the 7 + 3 standard we must develop, intelligent clinical trial designs which will separate the different AML subsets and provide the appropriate novel therapeutic strategies which will benefit these patients. This is something we have already learned from the anthracycline dose-intensification story.

Funding

This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

Conflict of interest statement

The authors declare no conflicts of interest in preparing this article.

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