Supplementary Appendix

Supplementary Methods

Exclusion Criteria

Patients were excluded if they had received an investigational agent within 14 days, or hydroxyurea or medications to reduce blast count within 24 hours before randomisation. Patients who had undergone an allogeneic or autologous stem cell transplant for acute myeloid leukaemia (AML) and were refractory or relapsed within 90 days before randomisation, or on active immunosuppressive therapy for graft-versus-host disease (GVHD) or GVHD prophylaxis within 2 weeks before randomisation were also excluded. Patients with known or suspected central nervous system involvement of active AML were excluded. Except non-melanoma skin cancer or cervical intraepithelial neoplasia, patients with other active or non-active malignancies (including other haematologic malignancies) within 12 months before randomisation were excluded. Patient must not have had any uncontrolled active infections or infestations and did not have a history of myocardial infarction, unstable angina, cerebrovascular accident, or transient ischemic attack within 3 months prior to randomisation.

Follow-Up

Upon treatment completion or discontinuation, patients were followed monthly during year 1, every 2 months during year 2, and every 3 months thereafter for survival, disease status for patients in continuing CR, CRp, or CRi, and subsequent AML treatment.

Supplementary Results

Table S1. Subsequent therapy

	vos/cyt (n=356)	pla/cyt (n=355)	total (N=711)
Any subsequent non-protocol AML therapy (including transplantation), % (95% CI)			
Overall	65.4 (60.3, 70.4)	71.8 (66.8, 76.5)	68.6 (65.1, 72.0)
By age, years			
<60	78.5 (70.4, 85.2)	81.5 (73.8, 87.8)	80.0 (74.6, 84.7)
≥60	58.0 (51.2, 64.5)	66.2 (59.6, 72.4)	62.1 (57.4, 66.6)
Subsequent transplantation, % (95% CI)			
Overall	30.1 (25.3, 35.1)	29.0 (24.3, 34.0)	29.5 (26.2, 33.0)
By age, years			
<60	46.2 (37.4, 55.1)	45.4 (36.6, 54.3)	45.8 (39.6, 52.0)
<60	20.8 (15.7, 26.7)	19.6 (14.6, 25.3)	20.2 (16.6, 24.2)

CI=confidence interval; cyt=cytarabine; pla=placebo; vos=vosaroxin.

Table S2. Overall Survival by Cytogenetic Risk Group

	Cytogenetic Risk*					
	Favorable		Intermediate		Unfavorable	
	vos/cyt (n=7)	pla/cyt (n=9)	vos/cyt (n=175)	pla/cyt (n=155)	vos/cyt (n=58)	pla/cyt (n=75)
Median overall survival (95% CI)	15.6 (1.0, NE)	19.6 (4.2, NE)	8.3 (7.1, 10.9)	7.1 (5.5, 9.4)	5.0 (4.0, 7.7)	3.8 (2.8, 5.0)
P value, unstratified	0.87		0.32		0.20	
P value, stratified	0.39		0.30		0.0214	
Hazard ratio estimates (95% CI)						
Unstratified	0.90 (0.24, 3.36)		0.88 (0.69, 1.13)		0.79 (0.55, 1.14)	
Stratified	0.48 (0.09, 2.63)		0.88 (0.68, 1.13)		0.63 (0.42, 0.94)	

*Per National Comprehensive Cancer Network Treatment Guidelines, AML 2014. Results of cytogenetic assessments were available at screening for 479 patients (67%).

CI=confidence interval; cyt=cytarabine; NE=not estimable; pla=placebo; vos=vosaroxin.

Table S3. Overall Survival by FLT3 and NPM1 Molecular Status

	Molecular Status*							
	FLT3 Mutated		FLT3 Wild-Type		NPM1 Mutated		NPM1 Wild-Type	
	vos/cyt (n=17)	pla/cyt (n=20)	vos/cyt (n=66)	pla/cyt (n=62)	vos/cyt (n=22)	pla/cyt (n=25)	vos/cyt (n=53)	pla/cyt (n=50)
Median overall survival (95% CI)	7.4 (2.0, 20.2)	2.2 (1.1, 3.8)	8.2 (6.7, 10.1)	9.3 (5.2, 11.3)	10.5 (6.4, 15.6)	8.0 (3.6, 23.4)	7.4 (4.5, 10.1)	5.5 (3.9, 10.0)
P value, unstratified	0.0368		0.66		0.85		0.90	
P value, stratified	0.19		0.87		0.40		0.70	
Unstratified hazard ratio estimate (95% CI)	0.47 (0.23, 0.97)		1.09 (0.74, 1.60)		0.94 (0.48, 1.85)		0.97 (0.64, 1.48)	

*Results of FLT3 and NPM1 molecular assessments were available at screening for 367 patients (52%) and 150 patients (21%), respectively.

CI=confidence interval; cyt=cytarabine; pla=placebo; vos=vosaroxin.

Table S3. Treatment-related SAEs occurring in >1 patient in either treatment arm (N=705)

	vos/cyt (n=355)	pla/cyt (n=350)
SAEs, no. (%)	116 (32·7)	58 (16·6)
Haematologic events, n (%)		
Febrile neutropenia	24 (6·8)	15 (4·3)
Anemia	0	2 (0·6)
Pancytopenia	2 (0·6)	0
Gastrointestinal disorders, n (%)		
Stomatitis	12 (3·4)	3 (0·9)
Colitis	3 (0·8)	0
Esophagitis	3 (0·8)	0
Enteritis	2 (0·6)	1 (0·3)
Infections and infestations, n (%)		
Sepsis	21 (5·9)	11 (3·1)
Bacteremia	12 (3·4)	5 (1·4)
Neutropenic sepsis	9 (2·5)	6 (1·7)
Pneumonia	9 (2·5)	3 (0·9)
Septic shock	3 (0·8)	4 (1·1)
Pneumonia, fungal	3 (0·8)	0
Cellulitis	2 (0·6)	1 (0·3)
Enterococcal sepsis	2 (0·6)	0
Perirectal abscess	2 (0·6)	0
Streptococcal bacteremia	2 (0·6)	0
Staphylococcal bacteremia	1 (0·3)	2 (0·6)
Infection, fungal	1 (0·3)	2 (0·6)
Urinary tract infection	0	3 (0·9)
Metabolism and nutrition disorders		
Dehydration	2 (0·6)	0
Respiratory, thoracic, and mediastinal disorders		
Epistaxis	0	2 (0·6)
Pulmonary hemorrhage	2 (0·6)	0

cyt=cytarabine; pla=placebo; SAE=serious adverse event; vos=vosaroxin.

The following treatment-related SAEs by MedDRA System Organ Class and Preferred Term occurred in one patient in the vosaroxin+ara-C arm: thrombocytopenia, disseminated intravascular coagulation, left ventricular dysfunction, myocardial infraction, vitreous hemorrhage, diarrhoea, ileus, nausea, enterocolitis, gastrointestinal hemorrhage, gastrointestinal inflammation, neutropenic colitis, non-cardiac chest pain, sudden death, cholestasis, cholestasis, cytolytic hepatitis, hepatic function abnormal, bronchopulmonary aspergillosis, escherichia sepsis, device related infection, enterobacter infection, enterococcal infection, enterocolitis bacterial, enterocolitis infectious, fungaemia, fungal sepsis, gastroenteritis, infection, klebsiella infection, pneumonia staphylococcal, pulmonary sepsis, urinary tract infection enterococcal, urosepsis, vulval abscess, blood pressure increased, lipase increased, failure to thrive, subarachnoid hemorrhage, renal failure, hemothorax, pneumonitis, pulmonary edema, respiratory failure, angioedema, hypotension, hypovolemic shock.

The following treatment-related SAEs by MedDRA System Organ Class and Preferred Term occurred in one patient in the placebo+ara-C arm: thrombocytopenia, neutropenia, tachycardia, diarrhoea, ileus, nausea, gastric ulcer perforation, ileitis, pyrexia, cholestasis, cytolytic hepatitis, bronchopulmonary aspergillosis, escherichia sepsis, anal abscess, clostridial infection, device related sepsis, diverticulitis, escherichia bacteraemia, lower respiratory tract infection, pharyngitis, systemic candida, transaminases increased, failure to thrive, headache, neurotoxicity, skin nodule.

Figure S1. Schema of treatment and follow-up for patients with first relapsed or refractory AML treated with vosaroxin or placebo in combination with cytarabine in the VALOR study. Up to 2 induction cycles could be administered. If a complete remission (CR) or or CR without complete platelet recovery (CRp) was achieved by day 57 of induction 1 or 2, up to 2 cycles of consolidation could be administered if safety parameters were met. If a CR with incomplete recovery of platelets or neutrophils was achieved by day 57 of induction 1 or 2, consolidation could be considered if haematologic recovery occurred by day 85. Follow-up for all patients commenced after treatment was either completed or discontinued for any reason. AML=acute myeloid leukaemia; IDAC=intermediate-dose ara-C.

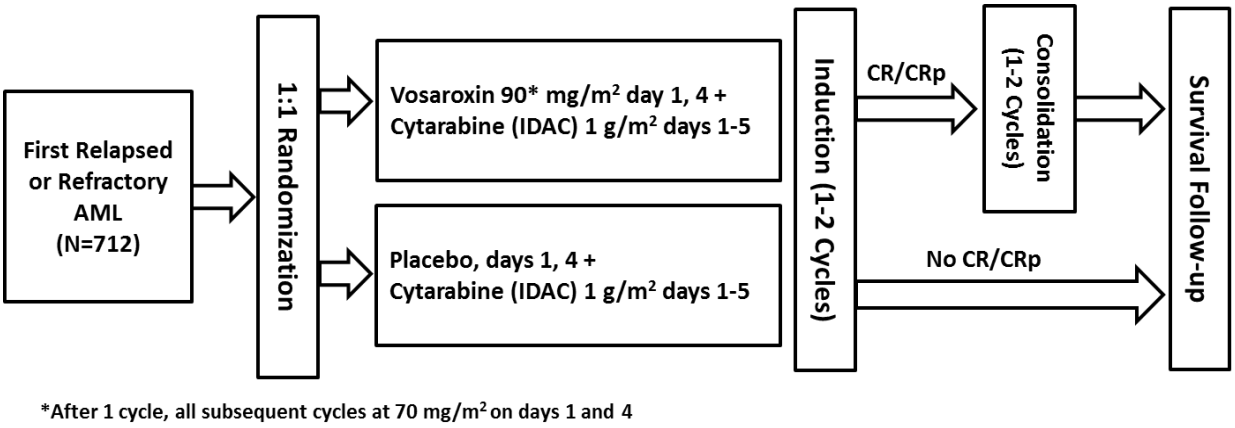

Figure S2. Patient disposition