**Supplementary experimental procedures**

**Gene Expression data analyses and building of the Iron Score**

The list of 62 genes involved in the regulation of iron biology was established using previously published data (Miller et al., 2011).

Expression of these genes was interrogated in normal B cells (n=5 centroblasts, n=5 centrocytes) and in DLBCL samples (n=73) using data published by the group of Compagno (Compagno et al., 2009). Affymetrix gene expression data are publicly available *via* the online Gene Expression Omnibus (http:// www.ncbi.nlm.nih.gov/geo/) under accession number GSE12195.

Significance analysis of microarray analysis was applied to the 62 selected probe sets in the different samples with 1000 permutations, a fold change of two and a false discovery rate of 0% (t-test).

Gene expression microarray data from two independent cohorts of patients diagnosed with DLBCL and treated by R-CHOP were used. The first cohort comprised 233 patients (Lenz R-CHOP cohort) (Lenz et al., 2008) and the second one comprised 69 patients (Melnick R-CHOP cohort) (Shaknovich et al., 2010). Pre-treatment clinical characteristics of patients were previously published by the groups of G. Lenz and of R. Shaknovich. Affymetrix gene expression data are publicly available *via* the online Gene Expression Omnibus (http:// www.ncbi.nlm.nih.gov/geo/) under accession number GSE10846 and GSE23501. Another cohort of formalin-fixed paraffin-embedded (FFPE) tissue from newly-diagnosed DLBCL patients treated with R-CHOP (n =72) was used as validation cohort (FFPE R-CHOP cohort; GSE53786) (Scott et al., 2014). An other cohort of 181 newly diagnosed DLBCL patients treated by CHOP (Lenz CHOP cohort; GSE10846) was also used (Lenz et al., 2008). Gene expression profiling was performed using Affymetrix HG-U133 plus 2.0 microarrays for the two cohorts of patients. The data were analyzed with Microarray Suite version 5.0 (MAS 5.0), using Affymetrix default analysis settings and global scaling as normalization method. The trimmed mean target intensity of each array was arbitrarily set to 500.

In each cohort, the statistical significance of overall survival of the expression of each probe set of the iron list was calculated by the log-rank test and survival curves plotted using the Kaplan-Meier method (Maxstat R package) (Hothorn and Lausen, 2012) as previously described (Herviou et al., 2018). Survival curves were plotted using the Kaplan-Meier method in the platform Genomiscape (Kassambara et al., 2015). Probe sets with a common prognosis value in the two cohorts were selected. To gather their prognostic information within one parameter, the Iron Score of DLBCL was built as the sum of the beta coefficients weighted by ± 1 according to the patient signal above or below the probe set Maxstat value (Herviou et al., 2018; Hothorn and Lausen, 2012; Kassambara et al., 2012). Multivariate analysis was performed using the Cox proportional hazards model.

**Westernblot**

The total cell lysates were obtained with RIPA 1X lysis buffer (#9806, Cell Signaling®) according with the supplier recommendations. Protein lysates migrate on 10% polyacrylamide gel (NP-0301, Life technologies®), in MOPS 1X running buffer (NP0001, Life technologies®) or MES 1X running buffer NP-0002, Life technologies®) and proteins are transferred to nitrocellulose membrane (IB301001, I-Blot Transfert Starck, Life technologies®).

The primary antibodies mouse anti-γH2AX (Ser139) (clone JBW301, Merck Millipore, Darmstadt, Germany), rabbit anti-BCL-xl (#2764T, Cell Signaling, 1/1000), rabbit anti-c-Myc (#5605S, 1/1000, Cell Signaling®), rabbit anti-MCL1 (sc-819, Santa Cruz®), rabbit anti-bcl2 (#4223S, 1/1000, Cell Signaling) were incubated in TBS-Tween 20 0.1% (Tris-Buffered Saline, pH 7.4) with 5 % non-fat milk or Bovine serum albumin (Sigma-Aldrich, A7906).

Protein levels are objectified by labeling with an anti-β-actin mouse monoclonal antibody (Sigma, A5441, St Louis, MO, USA 1/1000). Primary antibodies are visualized with secondary anti-rabbit antibodies (Sigma®, A9169) or anti-mouse antibodies (Jackson, 115-036-068) coupled to peroxidase allows the development by chemiluminescence by Western Lightning ECL (NEL121001EA, Perkin Elmer®). Quantification of protein levels was performed with Image J® software (National Institutes of Health, Bethesda, MD, USA).

**Caspase activation assay**

Caspase activity measurement was performed with Caspase Glo 3/7 assay (G8091 Promega) and Caspase Glo 9 assay (G8210 Promega), after 24H of treatment with or without caspase inhibitors, in triplicate. Caspase-Glo assay was performed according to the manufacturer's protocol. Briefly, Caspase-Glo reagent was added 1:1 to sample volume into each well in a 96-well white plate and incubated at room temperature for 30 min protected from light, and negative controls were used to determine basal caspase activity.

CellTiter-Glo Luminescent Cell Viability was used in the same plate for normalization in relation to cell viability. Assay from Promega use a Centro LB 960 luminometer (Berthold Technologies, Bad Wildbad, Germany).

**Measurement of ROS production**

Reactive oxygen species (ROS) were staining using a CM-H2DFDA probe (C6827, Invitrogen) and measured with the FL1 channel of flow cytometer (LSRFortessa cytometer, BD Bioscience). Briefly, cells were treated as indicated in Fig 5. Then cell were incubated with 5µM CM-H2DFDA at 37°C for 20 minutes in PBS and washed with RPMI 1640 medium and incubated during 1 hour at 37°C. For controls GSH (0.5 or 5mM) and oxygen peroxide (10mM) cells were treated 1 hour before staining.

Lipid peroxidation were measured using a BODIPY-C11 581/591 probe (D-3861, Invitrogen) and measured with the FL1 channel of flow cytometer, cell were incubated with 2µM BODIPY-C11 581/591 at 37°C for 30 minutes in PBS and washed with RPMI 1640 medium and incubated during 1 hour at 37°C.

**Measurement of labile iron pool**

The LIP was quantified on its ability to bind to cell-permeable chelators, such as calcein acetoxymethyl ester (CA-AM). Upon entering viable cells, CA-AM undergoes hydrolysis by esterases to calcein and becomes fluorescent. Its fluorescence is quenched upon binding to cellular LIP, in a stoichiometric fashion (Prus and Fibach, 2008).

The addition of a high-affinity chelator, such as deferasirox, removes iron from its complex with CA, increases the fluorescence emitted by the cells. The difference in the cellular fluorescence before and after incubation with a high-affinity chelator (ΔF) reflects the amount of LIP. Cells were washed twice with saline and incubated at a density of 1x 106 cells per ml for 15 min at 37°C with 0.5 µM CA-AM (Sigma, St. Louis, MO). Then, the cells were washed twice and treated with Deferasirox (100 µM), Ironomycin (10µM) or left untreated.

Calcein fluorescence was measured with the FL1 channel of flow cytometer (LSRFortessa cytometer, BD Bioscience).

**Measurement of intracellular iron (II)**

Intracellular Fe2+ were measured in live cells with Biotracker 575 Red Fe2+ probe. Briefly, cells were washed twice with Hanks balanced salt solution and stained with 5µM of Biotracker 575 Red Fe2+ Dye (#SCT030, Merck) (stock solution : 1mM in DMSO), and incubated 1 hour at 37°C in an incubator. Cells were washed and resuspended in HBSS, fluorescence was measured with the FL2 channel of flow cytometer (LSRFortessa cytometer, BD Bioscience).

MFI of PE fluorescence were compared between CD20+ cells and CD20- cells.

**Intracellular GSH levels quantification.**

Cells were treated with or without AM5 (0.25 μM) or erastin (4µM, positif control of GSH depletion) as indicated, then harvested and counted. The intracellular GSH level was measured using a commercial kit (GSH/GSSG-Glo™ Assay, #V6611, Promega) according to the manufacturer’s protocol.

GSH/GSSG ratio was calculated with the following formula:

$${GSH}/{GSSG}ratio = \frac{(Net total glutathione RLU – Net GSSG RLU)}{[Net GSSG RLU/2]}$$

**Cytotoxicity assay based on LDH release**

Cells were seeded in a 48 well plates and treated with increasing concentration of Ironomycin, in presence of recommended controls: control without cells, vehicle only control and maximal LDH release control (obtained with the addition of 2μl of 10% Triton X-100 per 100μl for 10–15 minutes before collecting the samples for LDH detection).

The LDH release level was measured using a commercial kit (LDH-Glo™ Cytotoxicity Assay #J2380, Promega) according to the manufacturer’s protocol.

Percentage of cytotoxicity was measured with the following formula:

$$\% Cytotoxicity=100 ×\frac{(Experimental LDH Release – Medium Background)}{(Maximum LDH Release Control – Medium Background)}$$

**DNA repair foci—immunofluorescence microscopy**

**Gamma-H2AX staining:**

After deposition on slides using a Cytospin centrifuge, cells were fixed with 4% paraformaldehyde, permeabilized with 0.5% Triton in phosphate-buffered saline (PBS) and saturated with 5% bovine milk in PBS. The mouse anti-γH2AX (Ser139) antibody (clone JBW301, Merck Millipore, Darmstadt, Germany) was diluted 1/300 in 5% bovine milk in PBS, and deposited on cytospins for 90 min at room temperature. Slides were washed twice and antibody anti-mouse immunoglobulins conjugated to Alexa 488 (diluted 1/500 in 5% bovine milk in PBS) were added for 45 min at room temperature. Slides were washed and mounted with Vectashield and 1% DAPI. Images and fluorescence were captured with a ZEISS Axio Imager Z2 microscope (× 63 objective), analyzed with Zen software and FIJI (ImageJ) software. The number γH2AX foci was counted in at least 300 nuclei.

**Phospho-RPA2 (T21) staining**:

Cells are cultured 24 hours and treated with EdU during 30’. After cytospin preparation, cells were fixed with 4% paraformaldehyde and cells were washed with PBS and then incubated twice for 3 min at room temperature with CSK buffer (10 mM Pipes, pH 7.0, 100 mM NaCl, 300 mM sucrose, and 3 mM MgCl2) containing 0.7% Triton X-100 (CSK) and 0.3 mg/ml RNase A when specified (CSK+R). After pre-extraction, cells were washed with PBS and fixed with 2% PFA and permeabilized with 0.2% Triton in phosphate-buffered saline (PBS) and saturated with 5% bovine serum albumin in PBS-0.1% Tween 20. The rabbit phospho-RPA2 T21 antibody ([EPR2846(2)], ab109394, Abcam) was diluted 1/300 in 5% bovine serum albumin in PBS-0.1% Tween 20, and deposited on cytospins for overnight at 4°C. Slides were washed twice and antibodies to rabbit immunoglobulins conjugated to Alexa-Fluor 488 (A11008, molecular probe, Thermofisher) (diluted 1/500 in 5% bovine serum albumin in PBS-0.1% Tween 20) were added for 45 min at room temperature. After washes in PBS-T and PBS, Click-it reaction was performed using CuSO4 (3mM), ascorbate sodium salt (10mM) and Alexa Fluor 555 Azide, Triethylammonium Salt (30µM) (A20012, molecular probe, Thermofisher).

After washes in PBS-T and PBS, coverslips were incubated 5 min with 2 μg/ml DAPI in PBS and dipped in water and mounted on glass slides using ProLong Gold (P36930, Life technology, Thermofisher). Images and fluorescence were captured with a ZEISS Axio Imager Z2 Apotome microscope (× 63 objective), analyzed with Zen Software (version3.6, company, town state) and FIJI software. The number of pRPA2 T21 foci was counted in at least 300 nuclei.

**LC3BII foci—immunofluorescence microscopy**

After deposition on slides using a Cytospin centrifuge, cells were fixed with 4% paraformaldehyde, permeabilized with 0.5% Triton in phosphate-buffered saline (PBS) and saturated with 5% bovine milk in PBS. The rabbit anti-LC3B (D11) antibody (Cell Signaling, Bioké, Leiden, The Netherlands, #3868) and mouse anti-alpha-tubulin (Sigma-Aldrich, St. Louis MO, USA, T9026) were diluted 1/300 in 5% bovine milk in PBS, and deposited on cytospins for 90 min at room temperature. Slides were washed twice and antibody anti-rabbit and mouse immunoglobulins conjugated to Alexa 488 and Texas Red, respectively (diluted 1/500 in 5% bovine milk in PBS) were added for 45 min at room temperature. Slides were incubated 5 min with 2 μg/ml DAPI in PBS and dipped in water and mounted on glass slides using ProLong Gold (P36930, Life technology, Thermofisher). Images and fluorescence were captured with a ZEISS Axio Imager Z2 microscope (× 63 objective), analyzed with Zen software and FIJI (ImageJ) software. The number LC3B-II foci was counted in at least 300 cells.

**DNA fiber spreading:**

DNA fiber spreading was performed as described (Breslin et al., 2006), (Jackson and Pombo, 1998). Briefly, DLBCL cells were labeled sequentially with 10 μM of IdU (Iododeoxyuridine) and 100 μM of CldU (Chlorodeoxyuridine) with or without treatment for 30 min. A thousand cells were transferred on a glass slide (SuperFrost) and lysed with spreading buffer (200 mM Tris.HCl ph7.5; 50 mM EDTA; 0.5 % SDS) by gently stirring with a pipette tip. The slides were tilted slightly and the surface tension of the drops was disrupted with a pipette tip. The drops were let run down the slides slowly, air-dried, fixed in methanol/acetic acid 3:1 for 10 minutes and allowed to dry. Slides are washed with H2O and denatured with 2.5M HCl for 1 hour and washed with blocking solution (PBS 1% tween 20, 1% BSA) for 1 hour.

Glass slides were processed for immunostaining using IgG1 mouse anti- BrdU for IdU detection (BD, clone B44, #347580, 1/100), IgG2a rat anti-BrdU for CldU detection (clone BU1/75, 1/100, Eurobio AbCys, ABC117-7513), mouse anti-ssDNA (1/50) antibodies and corresponding secondary antibodies conjugated to different Alexa-Fluor dyes (Goat anti-Mouse IgG1 AF546 1/100 #A21123, Chicken anti-Rat AF488 1/100 # A21470, Goat anti-Mouse IgG2a AF647 1/50 #A21241, Molecular probe). The nascent DNA fibers were visualized using immunofluorescence microscopy (Leica DM6000). The acquired DNA fiber images were analyzed using FIJI software and statistical analysis was performed with GraphPad Prism V7.

**Drug combination study**

The interaction between the drugs tested in vitro was investigated with a concentration matrix test, in which increasing concentration of each single drug were assessed with all possible combinations of the other drugs. For each combination, the percentage of expected growing cells in the case of effect independence was calculated according to the Bliss equation (Combes et al., 2019; Greco et al., 1995):

𝑓𝑢C = 𝑓𝑢𝐴.𝑓𝑢𝐵

where 𝑓𝑢C is the expected fraction of cells unaffected by the drug combination in the case of effect independence, and 𝑓𝑢𝐴 and 𝑓𝑢𝐵 are the fractions of cells unaffected by treatment A and B, respectively. The difference between the fraction of living cells in the cytotoxicity test and the 𝑓𝑢C value was considered as an estimation of the interaction effect, with positive values indicating synergism and negative values antagonism.

**Primary DLBCL Cells**

Lymph node samples were collected after patients’ written informed consent in accordance with the Declaration of Helsinki and institutional research board approval from Montpellier University hospital. Cells are obtained from lymph nodes or blood of 5 patients with DLBCL. Cells from blood are obtained by density gradient separation and cells from lymph node are obtained with a tissues dissociator and qualified by Flow cytometry.

Cells are cultured in Gibco ® Iscove’s MDM (Glutamax) medium (#31980-022) with 20% FBS with antibiotitcs-antimicotics (Gibco Penicillin-streptomycin-amphotericin B 100X, #15240-096) at a density of 0.5x10^6 Cell/mL with 50ng/mL of histidine-tagged CD40L (R&D System, 2706-CL) and 5µg/mL of anti-histidine antibody R&D System, MAB050), Gibco ® pyruvate 100X, # 1136-039. Cells are seeded 24H after thawing and treated with various compounds during 72H.

Total cells were counted with trypan bleu and stained with the panel CD45 V500 (BD, #560777), Kappa FITC (Dako, F0434), CD19 PE-Cy7 (BD, #341113), Lambda PE (Dako, R0437), CD3 APC-H7 (BD, #641415), CD10 APC (BD, #332777) and CD20 V450 (BD, #655872) and analyzed by flow cytometry (Canto II cytometer, BD Pharmigen). Malignant cells were gated on CD19+, CD45+, CD20+, Kappa or lambda and non-tumorous population cells were gated on CD45+,CD19-, and T cells on CD45+, CD3+, CD19-.