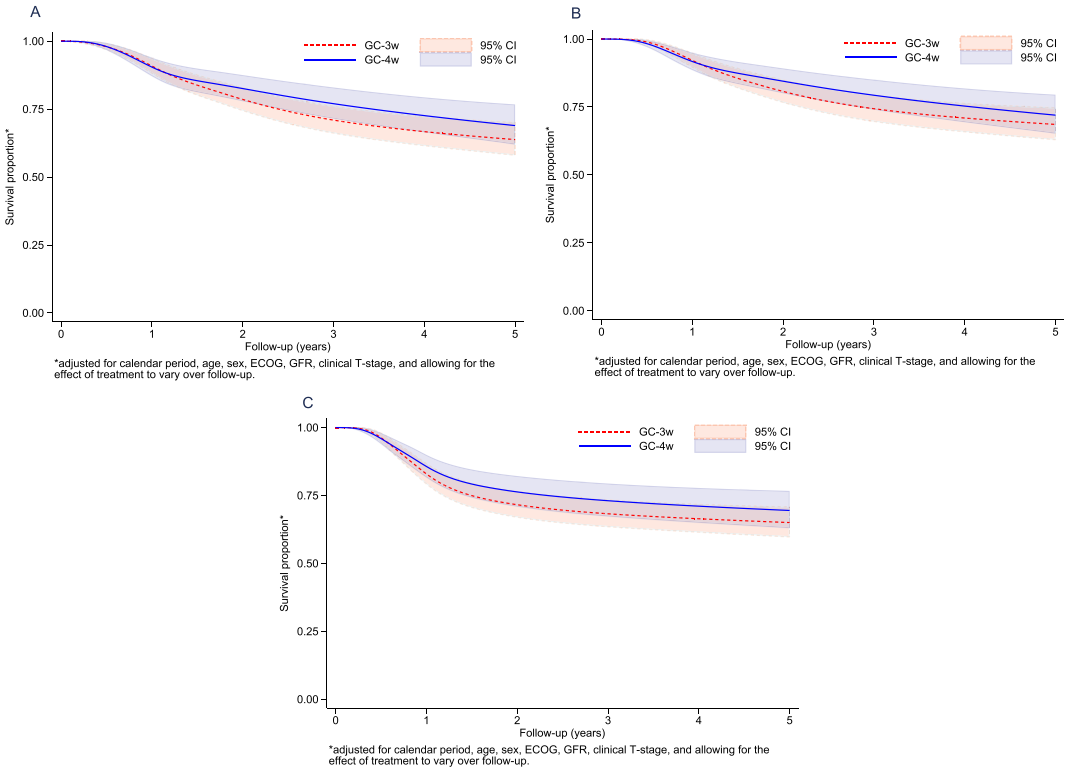

Fig. 2. Adjusted overall survival (A), adjusted bladder-cancer-specific survival (B), and adjusted relapse-free survival (C) for GC-4w versus GC-3w standardized for calendar period, age, sex, ECOG, GFR, and clinical T-stage, allowing for the effectof treatment to vary overthe follow-up period.

cumulative dose of cisplatin (280 versus 210 mg/m^2) can plausibly explain the larger proportion of pT0N0 in patients receiving GC-3w. Similarly, in the recently published neoadjuvant phase III trial VESPER, ddMVAC with a higher total dose and dose intensity of cisplatin showed significantly higher pathological downstaging compared to a 3-week schedule of GC [17]. The pathological downstaging rate for the GC-3w regimen in our trial is comparable with or exceeds the best downstaging data reported in MIBC for MVAC and GC, including dose-dense regimens [5, 11, 17–20].

Pre-treatment clinical stage (cTNM) has been shown to be an important prognostic factor for pathological downstaging (pTNM) at cystectomy [11, 21]. In the present study, the GC-4w-treated patients had more advanced tumours (cT3–cT4aN0) at baseline. After adjusting for the imbalance in the pre-treatment clinical stage, the GC-3w patients still achieved pT0N0 more frequently, although residual confounding cannot be ruled out. Moreover, patients with cT3–cT4aN0 in both cohorts had significantly lower rates of complete and partial response compared to patients with pre-treatment clinical stage cT2N0.

Interestingly, the favourable downstaging efficacy in the patient group treated with GC-3w did not however translate into a corresponding improvement in relapse-free, bladder-cancer-specific, or overall survival. Sensitivity analysis using a relative survival framework that took differences in background mortality in Sweden and Denmark into account confirmed the robustness of our data, and it yielded survival estimates that were nearly identical to those obtained when using bladder-cancer-specific mortality. Patients achieving pathological complete response showed 5-year survival of 90%, confirming that pT0N0 is a prognostic marker for favourable outcome [5, 22, 23].

The present study demonstrates the importance of a cisplatin-dose-intensive chemotherapy regimen to maximise the downstaging efficacy of the primary tumour in the bladder. However, no statistical differences were detected in relapse rate or survival parameters between GC-4w and GC-3w, indicating

a similar proportion of patients with disseminated micro-metastatic disease which presumably was *de novo* resistant to GC. Considering efficacy in eradicating distant micro metastases in MIBC, it is plausible that the sensitivity of individual tumour cells to cisplatin is more important than the final cumulated dose and dose intensity of cisplatin. To further improve the efficacy of GC as neoadjuvant treatment, it appears important to combine GC with drugs active on cisplatin resistant tumor cells rather than to further explore more dose-intense GC-combinations. This can be done by for example adding immunotherapy with immune checkpoint inhibitors (ICIs) [24–26], targeted therapies such as inhibitors of fibroblast growth factor receptor (FGFR) [27], or antibody-drug conjugates (ADCs) targeting Nectin-4 [28]. For patients with remaining residual muscle-invasive or node-positive disease the prognosis was poor (45% 5-year survival rate) an observation in line with previous studies [5, 22, 23]. Novel approaches for these patients, i.e., adjuvant precision-based treatment based on the biomarker profiles in the cystectomy specimen or in liquid biopsies, are warranted [29, 30].

The GC-3w schedule was associated with a higher degree of grade 3/4 AEs and patients treated with this regimen also more frequently discontinued treatment and experienced dose delays, mainly due to a significantly higher incidence of neutropenia. These findings indicate that G-CSF prophylaxis should be considered as a routine treatment as part of the GC-3w regimen. In the GC-4w arm, a low dose intensity was seen in gemcitabine day 15, which is in line with results from comparison of the two schedules in mUC [31]. Non-haematological grade 3/4 AEs (including decreased renal function, impaired hearing, and peripheral neuropathy) were few in both treatment groups, however grading of side effects are known to be underestimated in retrospective studies [32].

The main strengths of the present trial are the large size of the total cohort of consecutively treated patients and that criteria for neoadjuvant chemotherapy are similar in Stockholm, Sweden, and Denmark. Furthermore, the 3-week and 4-week GC schedules are standard of care in the two countries, thereby avoiding selection bias in the choice of chemotherapy regimens. The main limitation is the retrospective non-randomised approach, with the risk of bias from unknown cofounders and/or residual confounding despite careful adjustments. Median follow-up was relatively short and longer follow-up may allow for more accurate estimates on OS. Moreover, we lacked information regarding the extent of the diagnostic TUR-B and surgical cystectomy outcomes (i.e., number of lymph nodes resected, positive surgical margins, and type of urinary diversion).

In conclusion, the patient group treated with neoadjuvant chemotherapy with a more cisplatin-dose-intense 3-week regimen showed a significantly higher complete pathological response-rate compared to a commonly used 4-week gemcitabine-cisplatin schedule. The toxicity profile was manageable in both treatment regimens, but more neutropenia and premature treatment termination was observed in association to the GC-3w regimen. Relapse-free and overall survival were similar, indicating that future prospective studies should focus on identifying novel perioperative combination regimens which are active on cisplatin-gemcitabine resistant micro-metastatic disease.

ACKNOWLEDGMENTS

We thank Mikaela Hermansson Salmi, Department of Pelvic Cancer, Karolinska University Hospital, Stockholm, Sweden, for administrative support during this trial.

FUNDING

This trial was an academic study supported by grants from the Swedish Cancer Society, the Cancer Society of Stockholm, King Gustaf V Jubilee Fund, and the Stockholm County Council.

AUTHOR CONTRIBUTIONS

K Holmsten: Study concept and design, Data Collection, Data Analysis, Statistical Analysis, Manuscript Writing and Editing; L Høj Omland: Study concept and design, Data Collection, Manuscript Editing; A B Als: Data Collection, Manuscript Editing; M Agerbæk: Data Collection, Manuscript Editing; L Hammer Dohn: Data Collection, Manuscript Editing; H Lindberg: Data Collection, Manuscript Editing; N V Jensen: Data Collection, Manuscript Editing; A Carus: Study concept and design, Data Collection, Data Analysis, Manuscript Editing; M Moe: Data Collection, Manuscript Editing; A Hosseini: Data Collection, Manuscript Editing; C Radkiewicz: Data Analysis, Statistical Analysis, Manuscript Editing; H Pappot: Study concept and design, Data Analysis,

Manuscript Editing, Funding; A Ullén: Study concept and design, Data Analysis, Manuscript Writing and Editing, Funding.

CONFLICT OF INTEREST

K Holmsten has received speaker honoraria from Roche AB, Janssen; L Høj Omland has no conflicts of interest to report; A B Als has no conflicts of interest to report; M Agerbæk has no conflicts of interest to report; L Hammer Dohn has no conflicts of interest to report; H Lindberg has no conflicts of interest to report; N V Jensen has no conflicts of interest to report; A Carus has no conflicts of interest to report; M Moe has no conflicts of interest to report; A Hosseini has no conflicts of interest to report; C Radkiewicz has no conflicts of interest to report; H Pappot has received research funding from MSD and Roche, has served on an advisory board for MSD and has received speaker honoraria from BMS; A Ullén has received speaker honoraria or served on an advisory board for Pierre Fabre, Roche, Pfizer, Merck, Astellas Janssen-Cilag and MSD.

SUPPLEMENTARY DATA

Supplementary material related to this article can be found, in the online version. https://dx.doi.org/10.3233/BLC-211556.

REFERENCES

[1] ABCM-a. C. Neoadjuvant chemotherapy in invasive bladder cancer: a systematic review and meta-analysis. Lancet. 2003;361:1927-34.

[2] ABCM-a. C. Neoadjuvant chemotherapy in invasive bladder cancer: update of a systematic review and meta-analysis of individual patient data advanced bladder cancer (ABC) meta-analysis collaboration. Eur Urol. 2005;48:202-5; discussion 205-206.

[3] Winquist E, Kirchner TS, Segal R et al. Neoadjuvant chemotherapy for transitional cell carcinoma of the bladder: a systematic review and meta-analysis. J Urol. 2004;171:561-9.

[4] Griffiths G, Hall R, Sylvester R et al. International phase III trial assessing neoadjuvant cisplatin, methotrexate, and vinblastine chemotherapy for muscle-invasive bladder cancer: long-term results of the BA06 30894 trial. J Clin Oncol. 2011;29:2171-7.

[5] Grossman HB, Natale RB, Tangen CM et al. Neoadjuvant chemotherapy plus cystectomy compared with cystectomy alone for locally advanced bladder cancer. N Engl J Med. 2003;349:859-66.

[6] Yuh BE, Ruel N, Wilson TG et al. Pooled analysis of clinical outcomes with neoadjuvant cisplatin and gemcitabine chemotherapy for muscle invasive bladder cancer. J Urol. 2013;189:1682-6.

[7] Kim HS, Jeong CW, Kwak C et al. Pathological T0 Following Cisplatin-Based Neoadjuvant Chemotherapy for Muscle-Invasive Bladder Cancer: A Network Meta-analysis. Clin Cancer Res. 2016;22:1086-94.

[8] Yin M, Joshi M, Meijer RP et al. Neoadjuvant Chemotherapy for Muscle-Invasive Bladder Cancer: A Systematic Review and Two-Step Meta-Analysis. Oncologist. 2016;21:708-15.

[9] Goel S, Sinha RJ, Bhaskar V et al. Role of gemcitabine and cisplatin as neoadjuvant chemotherapy in muscle invasive bladder cancer: Experience over the last decade. Asian J Urol. 2019;6:222-9.

[10] von der Maase H, Hansen SW, Roberts JT et al. Gemcitabine and cisplatin versus methotrexate, vinblastine, doxorubicin, and cisplatin in advanced or metastatic bladder cancer: results of a large, randomized, multinational, multicenter, phase III study. J Clin Oncol. 2000;18:3068-77.

[11] Zargar H, Espiritu PN, Fairey AS et al. Multicenter assessment of neoadjuvant chemotherapy for muscle-invasive bladder cancer. Eur Urol. 2015;67:241-9.

[12] Witjes AJ, Lebret T, Comperat EM et al. Updated 2016 EAU Guidelines on Muscle-invasive and Metastatic Bladder Cancer. Eur Urol. 2017;71:462-75.

[13] Brierley JDG, M.K.; Wittekind C. TNM Classification of Malignant Tumours, 8th Edition. Wiley-Blackwell, 2016.

[14] Moch HH, P.A.; Ulbright TU, Reuter VR. WHO Classification of Tumours of the Urinary System and Male Genital Organs (IARC WHO Classification of Tumours) 4th Edition.2016.

[15] Lambert PC RP. Further development of flexible parametric models for survival analysis. The Stata Journal. 2009;9(2):265-90.

[16] Dickman P CE. Estimating and modeling relative survival. The Stata Journal. 2015;15(1):186-215.

[17] Pfister C, Gravis G, Fléchon A et al. Randomized Phase III Trial of Dose-dense Methotrexate, Vinblastine, Doxorubicin, and Cisplatin, or Gemcitabine and Cisplatin as Perioperative Chemotherapy for Patients with Muscle-invasive Bladder Cancer. Analysis of the GETUG/AFU V05 VESPER Trial Secondary Endpoints: Chemotherapy Toxicity and Pathological Responses. Eur Urol 2020.

[18] Plimack ER, Hoffman-Censits JH, Viterbo R, et al. Accelerated methotrexate, vinblastine, doxorubicin, and cisplatin is safe, effective, and efficient neoadjuvant treatment for muscle-invasive bladder cancer: results of a multicenter phase II study with molecular correlates of response and toxicity. J Clin Oncol. 2014;32:1895-901.

[19] Zargar H, Shah JB, van de Putte EEF et al. Dose dense MVAC prior to radical cystectomy: a real-world experience. World J Urol. 2017;35:1729-36.

[20] Iyer G, Balar AV, Milowsky MI et al. Multicenter Prospective Phase II Trial of Neoadjuvant Dose-Dense Gemcitabine Plus Cisplatin in Patients With Muscle-Invasive Bladder Cancer. J Clin Oncol. 2018;36:1949-56.

[21] Fairey AS, Daneshmand S, Quinn D et al. Neoadjuvant chemotherapy with gemcitabine/cisplatin vs. methotrexate/vinblastine/doxorubicin/cisplatin for muscle-invasive urothelial carcinoma of the bladder: a retrospective analysis from the University of Southern California. Urol Oncol. 2013;31:1737-43.

[22] Rosenblatt R, Sherif A, Rintala E, et al. Pathologic downstaging is a surrogate marker for efficacy and increased survival following neoadjuvant chemotherapy and radical

cystectomy for muscle-invasive urothelial bladder cancer. Eur Urol. 2012;61:1229-38.

[23] Petrelli F, Coinu A, Cabiddu M et al. Correlation of pathologic complete response with survival after neoadjuvant chemotherapy in bladder cancer treated with cystectomy: a meta-analysis. Eur Urol. 2014;65:350-7.

[24] Necchi A, Anichini A, Raggi D, et al. Pembrolizumab as Neoadjuvant Therapy Before Radical Cystectomy in Patients With Muscle-Invasive Urothelial Bladder Carcinoma (PURE-01): An Open-Label, Single-Arm, Phase II Study. J Clin Oncol. 2018;36:3353-60.

[25] Powles T, Kockx M, Rodriguez-Vida A, et al. Clinical efficacy and biomarker analysis of neoadjuvant atezolizumab in operable urothelial carcinoma in the ABACUS trial. Nat Med. 2019;25:1706-14.

[26] Siefker-Radtke A. KEYNOTE-866: Phase 3 study of perioerative pembrolizumab or placebo in combination with neoadjuvant chemotherapy in cisplatin-eligible patients with muscle-invasive bladder cancer. Annals of Oncology. 2019;30(suppl_5):v356-v402. 10.1093/annonc/mdz249 2019.

[27] Loriot Y, Necchi A, Park SH, et al. Erdafitinib in Locally Advanced or Metastatic Urothelial Carcinoma. N Engl J Med. 2019;381:338-48.

[28] Rosenberg JE, O'Donnell PH, Balar AV et al. Pivotal Trial of Enfortumab Vedotin in Urothelial Carcinoma After Platinum and Anti-Programmed Death 1/Programmed Death Ligand 1 Therapy. J Clin Oncol. 2019;Jco1901140.

[29] Bjerggaard Jensen J. Treatment Of Metastatic Bladder Cancer at the Time Of Biochemical reLApse Following Radical Cystectomy (TOMBOLA). ClinicalTrials.Gov, Available at: https://clinicaltrials.gov/ct2/show/NCT04138628 Accessed Oct 2, 2020.

[30] Christensen E, Birkenkamp-Demtröder K, Sethi H et al. Early Detection of Metastatic Relapse and Monitoring of Therapeutic Efficacy by Ultra-Deep Sequencing of Plasma Cell-Free DNA in Patients With Urothelial Bladder Carcinoma. J Clin Oncol. 2019;37:1547-57.

[31] Als AB, Sengelov L, Von Der Maase H. Gemcitabine and cisplatin in locally advanced and metastatic bladder cancer; 3- or 4-week schedule? Acta Oncol. 2008;47:110-9.

[32] Bamias A, Hegele A, Medioni J, et al. Vinflunine in the treatment of relapsed metastatic urothelial cancer: A systematic review and meta-analysis of real-world series. Crit Rev Oncol Hematol. 2019;140:80-7.