ANTICANCER RESEARCH *44*: 3269-3276 (2024)
doi:10.21873/anticanres.17145

Transcriptional Induction of *SERPINE1* and Fibrinolysis Inhibition as Predominant Effects of Glucocorticoids on the Cancer Coagulome

FLORIANE RACINE[1], CHRISTOPHE LOUANDRE[2], JULIEN DEMAGNY[3], CORINNE GODIN[1,2], ZUZANA SAIDAK[1,2] and ANTOINE GALMICHE[1,2]

[1]*UR7516, CHIMERE, Université de Picardie Jules Verne, Amiens, France;*
[2]*Service de Biochimie, Centre de Biologie Humaine, CHU Amiens, Amiens, France;*
[3]*Service d'Hématologie, Centre de Biologie Humaine, CHU Amiens, Amiens, France*

Abstract. *Background/Aim: How tumors regulate the genes of the coagulome is crucial for cancer-associated thrombosis and the occurrence of venous thromboembolic complications in patients with cancer. We have previously reported potent yet complex effects of glucocorticoids (GC) on the expression of three genes that play a key role in the regulation of thrombin/plasmin activation (F3, PLAU, and SERPINE1). This study aimed to extend the investigation of GC effects to the whole tumor coagulome and assess the resulting impact on the ability of cancer cells to activate thrombin and plasmin. Materials and Methods: Cancer RNA expression data were retrieved from various sources. Additionally, oral squamous cell carcinoma (OSCC) cells exposed to GC in vitro were analyzed using QPCR, enzymatic assays measuring thrombin and urokinase-type Plasminogen Activator (uPA) activity, and D-dimer concentrations. Results: Our findings highlight the potent and specific stimulatory effect of GC on SERPINE1 expression across different types of cancer. Consistently, GC were found to inhibit uPA proteolytic activity and reduce the concentrations of D-dimers in OSCC in vitro. Conclusion: Fibrinolysis inhibition is a key consequence of cancer cell exposure to GC, possibly leading to the stabilization of the fibrin clot in cancer.*

Correspondence to: Antoine Galmiche, MD, Ph.D., Service de Biochimie, Centre de Biologie Humaine (CBH), CHU Amiens Sud, Avenue Laennec, 80054 Amiens Cedex, France. E-mail: galmiche.antoine@chu-amiens.fr

Key Words: Cancer-associated thrombosis, coagulome, glucocorticoids, *SERPINE1*/Plasminogen Activator Inhibitor-1.

This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY-NC-ND) 4.0 international license (https://creativecommons.org/licenses/by-nc-nd/4.0).

Venous thromboembolism (VTE) is a major cause of morbidity and mortality in cancer patients (1, 2). Among the mechanisms that contribute to VTE, the establishment of a local hypercoagulable state, typically accounted for by the over-expression of tissue factor (TF) by tumor cells, is thought to be an essential step (1). Importantly, TF is a key regulator of thrombin activation and constitutes an essential trigger for the formation of the fibrin clot (1). How cancer cells regulate their expression of *F3*, the gene that encodes TF, has been a research question for investigators aiming to decipher the process of cancer-associated thrombosis (CAT) (1). In addition to *F3*, an array of genes regulates the activation of the proteases involved in coagulation and fibrinolysis. The recent application of systems biology, especially using transcriptomics, has allowed for a more precise, pan-cancer description of the human tumor coagulome, *i.e.*, the extended set of genes with direct or indirect effects on the regulation of coagulation and fibrinolysis (3, 4). This increasingly precise description of the tumor coagulome has unveiled great differences between different tumor types and among individual tumors (4). Some primary tumor types, such as glioblastoma multiforme (GBM) and pancreatic adenocarcinoma (PAAD), are characterized by high expression of *F3*. Oral squamous cell carcinoma (OSCC), one of the most frequent tumors of the upper aerodigestive tract, have been shown to express the highest levels of *F3* and *PLAU*, coding for the main regulators of coagulation and fibrinolysis TF and urokinase-type plasminogen activator (uPA), respectively (5, 6). The simultaneous high expression of the main regulators of coagulation and fibrinolysis makes OSCC a great model for the study of the coagulome, and in particular to address the biological determinants that shape its composition.

Glucocorticoids (GC) are steroid hormones produced by the adrenal gland, secreted during situations of stress. GC are involved in several biological processes, often related to their

ability to interact with the receptor *NR3C1*, a member of the nuclear receptor superfamily (7, 8). Synthetic GC agonists are often used in the clinic for multiple purposes that include the control of deleterious inflammatory reactions, direct targeting of tumor cells, as well as against the secondary effects induced by some anticancer treatments (9). Natural GC and synthetic agonists of the GC receptor can regulate a large variety of genes (7, 10). In a previous study, we have shown that GC are transcriptional regulators of three key coagulation-related genes in OSCC (11). While *F3*/TF and *PLAU*/uPA expression decreased upon cell exposure to dexamethasone, we noted a major direct positive transcriptional effect of dexamethasone on *SERPINE1*/plasminogen activator inhibitor-1 (PAI-1) expression. From these observations, we concluded that the expression of coagulation-related genes likely involves a dual regulation, with both direct transcriptional regulation and indirect effects possibly explained by the anti-inflammatory action of GC (11). By analyzing RNA-seq data from human tumors in The Cancer Genome Atlas (TCGA), we have suggested the existence of a potential GR-PAI-1 axis linked to increased levels of fibroblasts and endothelial cells in OSCC and PAAD (11). While interesting, our previous study did not address the effect of GC on the coagulome, and it did not assess the functional effects of GC on coagulation and fibrinolysis. In this present work, we further examined the effects of GC by extending our analysis to a set of 85 coagulation-related genes. In addition to providing a broader characterization of the effects of GC on the tumor coagulome, we explored the functional consequences of GC regulation in cancer cells in culture using enzymatic tests measuring coagulation / fibrinolysis and D-dimer concentrations.

Materials and Methods

In silico gene expression analysis. Our analysis was centered on the 85 genes in the Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway “Coagulation and complement cascades” (hsa04610). RNA-seq expression [log2(FPKM+1)] from the NCI-60 was retrieved through CellMiner (12). The GR-activity score was calculated as described previously (11), based on the study by West *et al*. (13). For each coagulation-related gene, a correlation coefficient was calculated between specific gene expression and the GR-activity score. Gene expression data (RNA SeqV2 data normalized using RNA-Seq by Expectation Maximization: RSEM) were retrieved for 19 cancer types from TCGA, using cBioportal (14). The GR-activity score was calculated using pan-cancer z score values. Data from a study exploring the impact of the glucocorticoid hydrocortisone (2.75 μM, 8 h) on global gene expression in five human non-small cell lung carcinoma (NSCLC) cell lines (A549, H2122, H460, H1975 and H1944) were retrieved from the Gene Expression Omnibus (GSE159546) (15). Fold change in gene expression with hydrocortisone treatment was determined for the 5 cell lines (log2).

Cell culture and reagents. The human OSCC cell line PE/CA-PJ41 was purchased from European Collection of Authenticated Cell Cultures (ECACC, Salisbury, England). Cells were cultured in Dulbecco’s Modified Eagle Medium (DMEM) supplemented with 10% fetal bovine serum (FBS), 2 mM glutamine and antibiotics (streptomycin and penicillin). Human recombinant tumor Necrosis Factor-α (TNFα, ref. 210-TA) was purchased from R&D Systems Biotechne (Noyal Chatillon sur Seiche, France). Dexamethasone (D4902), aldosterone (A9477), GW4064 (G5172), vitamin D3/cholecalciferol (679101), bexarotene (SML0282) and T3 thyroid hormone (T2877) were purchased from Sigma Aldrich (Saint-Quentin-Fallavier, France).

Quantitative PCR. Total RNA was extracted in RLT buffer and purified using RNeasy minikit (74104, Qiagen, Courtaboeuf, France). The samples were reverse transcribed using High-Capacity cDNA Reverse Transcription kit (4368814, ThermoFisher, Villebon-sur-Yvette, France). cDNAs were amplified using gene-specific probes for *F3* (Hs01076029_m1), *PLAU* (Hs01547054_m1), *SERPINE1* (Hs00167155_m1) and *GAPDH* (4333764F) (all from ThermoFisher). We used TaqMan Fast Advanced Master Mix (4444557, ThermoFisher) on a QuantStudio™ 7 Pro Real-Time PCR System (ThermoFisher).

Fibrinolysis and coagulation assays. Thrombin and uPA activities were measured using colorimetric substrates (ab234620 from Abcam (Amsterdam, the Netherlands) and AG4000 from Merck Millipore (Molsheim, France). Assays were conducted using cell culture supernatants from OSCC cells cultured in medium with 0.5% FBS, exposed to TNFα and dexamethasone, as indicated. For the thrombin coagulation assay, we added 10% human plasma (Human pooled plasma, Dominique Dutscher, Bernolsheim, France) immediately before the measurements. The enzymatic activity was assessed using a spectrophotometer at 414 nm (measured every 5 min over 2 h).

D-dimer measurements. D-dimer levels were assessed using the clinically-approved INNOVANCE® D-Dimer test (Siemens, Courbevoie, France). OSCC cells were cultured in DMEM containing 0.5% FBS, exposed to TNFα and dexamethasone as indicated. Coagulation was triggered by adding 10% human plasma 24 h before D-dimer measurements.

Statistical analysis. Student’s *t*-test was used to compare the different conditions using GraphPad Prism (5.00.288 version, Dotmatics, Boston, MA, USA). Correlation analyses were done using R version 4.3.2 (16). $p<0.05$ was considered as threshold for significance.

Results

Glucocorticoid receptor activity correlates with the expression of coagulome genes in cancer cells and human tumors. To perform a systems analysis encompassing the key genes of the coagulome, we retrieved RNA-seq data for genes from the KEGG pathway “Coagulation and complement cascades” (hsa04610; 85 coagulation-related genes) from the NCI-60 database, which contains data from 54 solid cancer cell lines (eight types of primary tumors). We calculated the GR-activity score for each cell line and performed a Pearson’s correlation analysis between the GR-activity scores and expression of the 85 coagulation-related genes (Figure 1A). We observed a significant correlation for eight genes ($R \geq |0.25|$ and $p<0.05$): *SERPINE1* (r=0.40), *SERPING1* (r=0.37), *TFPI* (r=0.35),

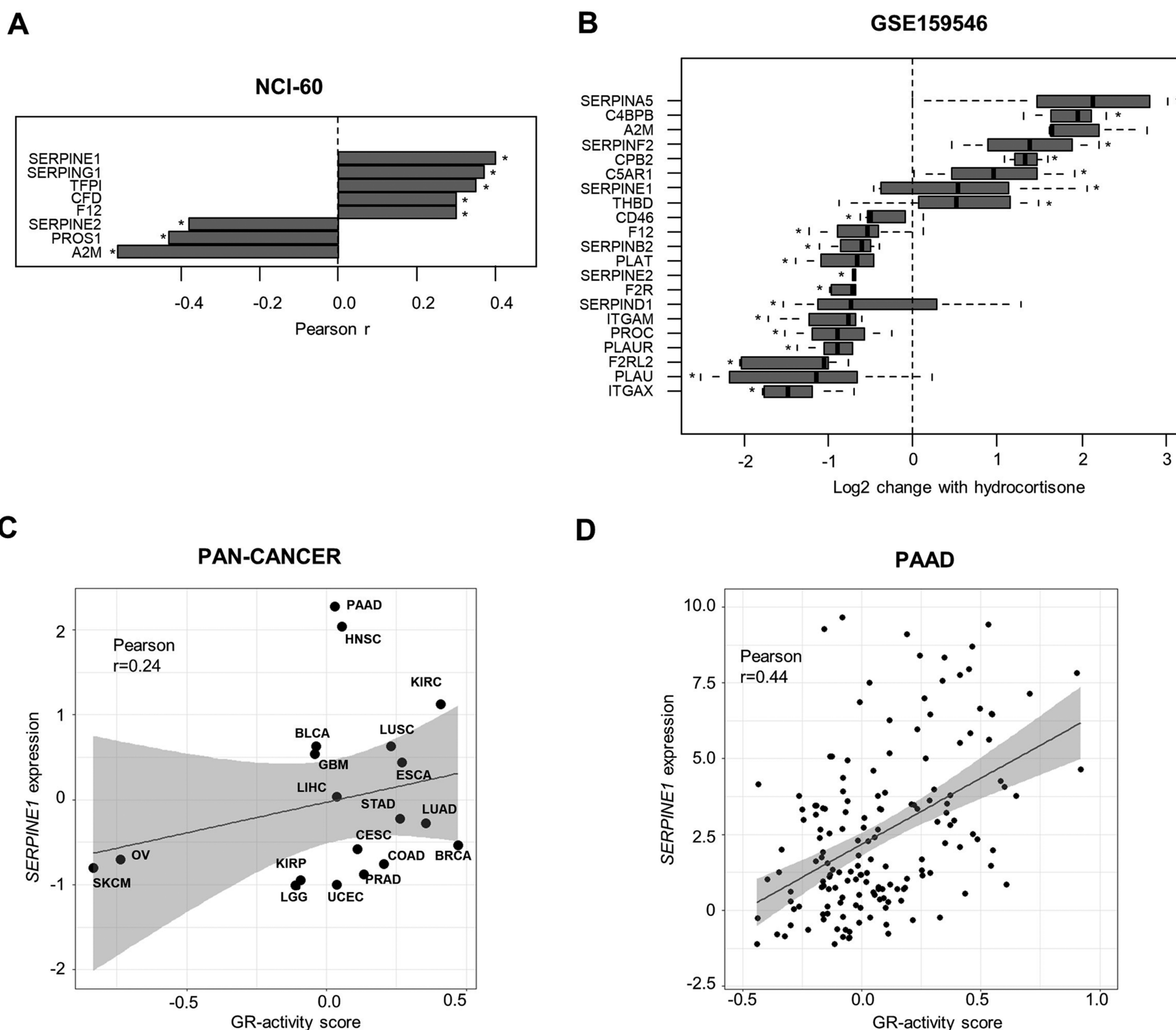

Figure 1. *Glucocorticoid regulation of coagulome-related genes in human tumors. (A) Pearson correlation between the GR-activity score for 54 solid cancer cell lines from the NCI-60 and their expression of the 85 coagulation-related genes from KEGG. (B) RNAseq data from the Gene Expression Omnibus (GSE159546) were used to evaluate the changes in expression of coagulation-related genes from KEGG (n=85). The log2 change in expression was calculated for five NSCLC cell lines treated with hydrocortisone (2.75 μM, 8 h). The top 21 genes either up-regulated or down-regulated are shown. (C, D) Pan-cancer correlation analysis of the GR-activity score and SERPINE1 expression, using gene expression data from 19 common human cancers from TCGA (C) and PAAD-TCGA (D).*

CFD (r=0.30), *F12* (r=0.30), *SERPINE2* (r=–0.38), *PROS1* (r=–0.43), *A2M* (r=–0.56). *SERPINE1* was the gene with the highest positive correlation score (r=0.40). We next retrieved data from GSE159546 (15) to evaluate the expression of the 85 coagulation related-genes in NSCLC cells exposed to hydrocortisone (2.75 μM, 8 h). We calculated the fold-change in expression of the 85 coagulation-related genes in the five cell lines treated with hydrocortisone (Figure 1B). Consistent variations in expression were noted for 21 genes (median log2 change >|0.5|) (13 down-regulated and 8 up-regulated). *SERPINE1* was among the genes that were up-regulated, confirming our previous observation in NCI-60. We then performed a pan-cancer analysis to correlate the GR-activity score and *SERPINE1* expression across different types of human tumors. We used bulk RNA-seq data available for 19 types of tumors in TCGA. A modest positive correlation was observed between GR-activity score and *SERPINE1* expression in the pan-cancer analysis (Pearson r=0.24) (Figure 1C). The correlation was however much stronger in tumor types characterized by an overall high expression of *SERPINE1*, such as pancreatic adenocarcinoma (PAAD) (Pearson r=0.44) (Figure 1D).

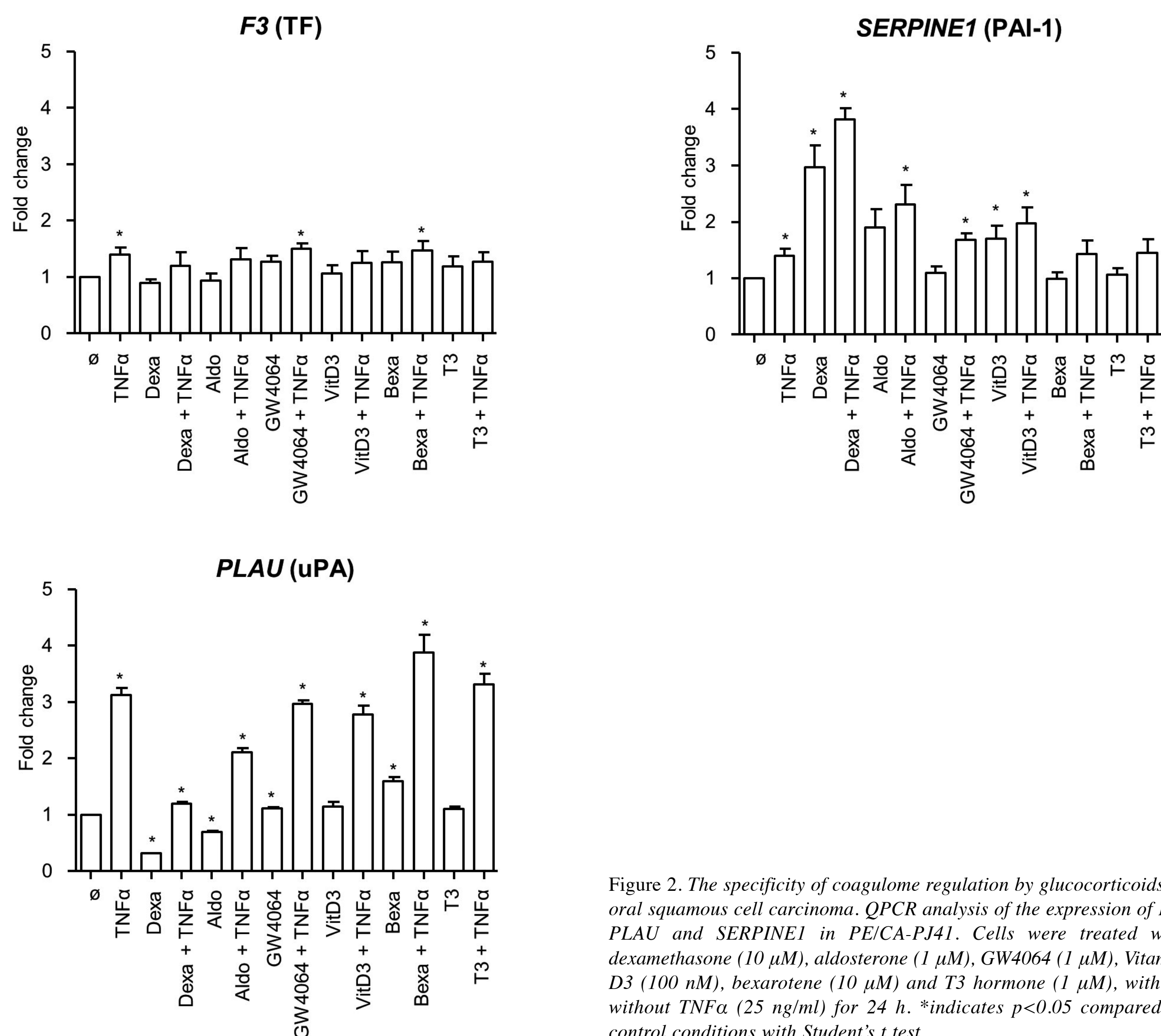

Figure 2. *The specificity of coagulome regulation by glucocorticoids in oral squamous cell carcinoma. QPCR analysis of the expression of F3, PLAU and SERPINE1 in PE/CA-PJ41. Cells were treated with dexamethasone (10 μM), aldosterone (1 μM), GW4064 (1 μM), Vitamin D3 (100 nM), bexarotene (10 μM) and T3 hormone (1 μM), with or without TNFα (25 ng/ml) for 24 h. *indicates p<0.05 compared to control conditions with Student's t test.*

Positive transcriptional regulation of SERPINE1 by GC in OSCC in vitro. In order to experimentally address the regulation of the coagulome by GC and examine its specificity, we tested the effect of five agonists active on other members of the nuclear receptor superfamily. The human OSCC cell line PE/CA-PJ41 was exposed *in vitro* to dexamethasone, aldosterone (mineralocorticoid receptor agonist), GW4064 (FXR agonist), Vitamin D3 (vitamin D receptor agonist), bexarotene (RXR agonist) and T3 hormone (thyroid hormone receptor agonist) (17) (Figure 2). PE/CA-PJ41 cells were treated with these drugs for 24 h at active concentrations, either alone or in combination with the inflammatory cytokine TNFα (25 ng/ml). Using QPCR, we observed that dexamethasone and other members of the nuclear receptor superfamily had little effect on the expression of *F3* (Figure 2). We noted that dexamethasone, and to a lower extent aldosterone, decreased the expression of *PLAU* mRNA, especially in cells exposed to TNFα. Conversely, dexamethasone sharply increased *SERPINE1* expression (an almost 3-fold induction, *i.e.,* greater than all other agonists used). In these experimental conditions, no biologically significant effect on cancer cell viability was detected (data not shown).

The effect of dexamethasone on coagulation and fibrinolysis was determined by measuring the activity of thrombin and uPA in the cell supernatants using specific peptide substrates. PE/CA-PJ41 cells were exposed to dexamethasone (1 μM) and TNFα (25 ng/ml) for six days. The supernatants were collected to measure the activity of

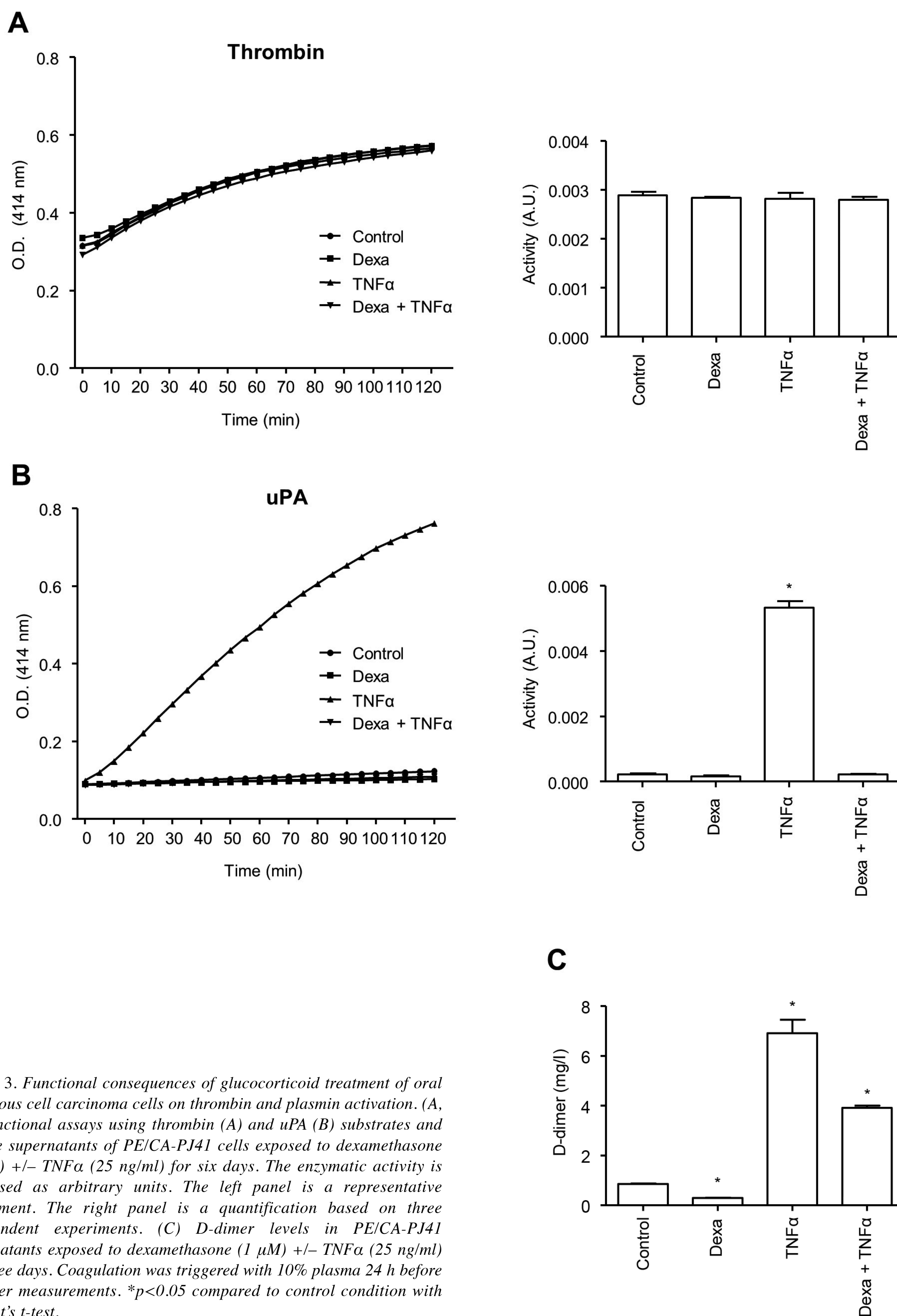

Figure 3. *Functional consequences of glucocorticoid treatment of oral squamous cell carcinoma cells on thrombin and plasmin activation. (A, B) Functional assays using thrombin (A) and uPA (B) substrates and culture supernatants of PE/CA-PJ41 cells exposed to dexamethasone (1 μM) +/– TNFα (25 ng/ml) for six days. The enzymatic activity is expressed as arbitrary units. The left panel is a representative experiment. The right panel is a quantification based on three independent experiments. (C) D-dimer levels in PE/CA-PJ41 supernatants exposed to dexamethasone (1 μM) +/– TNFα (25 ng/ml) for three days. Coagulation was triggered with 10% plasma 24 h before D-dimer measurements. *p<0.05 compared to control condition with Student's t-test.*

thrombin and uPA. We found no biologically-significant effect of TNFα or dexamethasone on thrombin activity in the supernatants of OSCC cells (Figure 3A). Conversely, TNFα produced a sharp increase in the enzymatic activity of uPA in the culture supernatants of OSCC cells. Interestingly, dexamethasone abolished the increase in uPA activity induced by TNFα (Figure 3B). In a parallel experiment, we measured the concentrations of D-dimers. We found that GC greatly reduce the appearance of these fibrin degradation products (Figure 3C), further suggesting that GC inhibit fibrinolysis in OSCC cells.

Discussion

The tumor coagulome is an essential tumor-intrinsic determinant of VTE complications. While it is possible to study the tumor coagulome at an unprecedented resolution with systems approaches, a major challenge remains in identifying the biological events and molecules that could dynamically shape it (4). Here, we extended the conclusions of our previous study (11) by documenting the effects of glucocorticoids on the cancer coagulome beyond the three key genes that were explored previously and in a number of different cellular contexts. In agreement with our previous findings of a direct positive transcriptional regulation of *SERPINE1* explained by the interaction of the active GR with the promoter of *SERPINE1* (11), we noted a consistent correlation between *SERPINE1* and GR activity in different transcriptional datasets (NCI60 and TCGA, covering multiple types of primary tumors). We tested the effects of GC on OSCC cells in an experimental setting allowing for the simultaneous examination of thrombin/uPA activity upon GC exposure, in control and inflammatory conditions (mimicked by TNFα). We observed a strong inhibition of the proteolytic activity of uPA and a simultaneous reduction of D-dimer concentrations, the most commonly measured product of fibrin degradation, in the OSCC culture supernatants. These findings suggest that GC block plasminogen activation and fibrinolysis and may therefore stabilize fibrin clots formed in the vicinity of cancer cells.

Our study is to the best of our knowledge the first to examine the functional consequences of GC exposure on cancer cell-associated thrombin/plasminogen activation. The radical inhibition of plasminogen activation by uPA that we observed is in accordance with a robust and specific induction of *SERPINE1*/PAI1, further supporting the concept of a GR/*SERPINE1* axis (11). This GR/*SERPINE1* axis might be of clinical relevance, considering that GC might increase the occurrence of fibrin clots and the risk of VTE in cancer patients. Importantly, GC are not only endogenous hormones, but they are also among the most commonly used therapeutics in cancer patients addressed for surgical or radiotherapeutic treatment, *i.e.*, procedures that increase the expression of *F3*/TF, as was for example recently shown in GBM (18). The risk of VTE induced by GC might be especially high for tumors such as OSCC that are characterized by the coexistence of a strong pro-fibrinolytic activity counterbalancing a hypercoagulable gene expression profile (19). For these tumors, thorough prospective studies addressing the effects of GC on biomarkers of coagulation/fibrinolysis/VTE risk are warranted. Interestingly, an increased risk of thromboembolic accidents has been noted with GC-treatment in the non-oncological context. A high risk of VTE was for example reported in Cushing's syndrome, *i.e.*, chronic hypercortisolism (20) and in patients with COVID-19 infection receiving oral GC (21). Furthermore, treatment of healthy subjects with GC for 10 days was found to induce a procoagulant state with increased PAI-1 blood levels (22). The conclusions of these studies and ours open up the possibility that a GR-*SERPINE1* axis might be of general relevance, beyond the oncological context.

Finally, we hypothesize that our observations may be important beyond the thromboembolic risk. *SERPINE1*/PAI-1 has consistently been reported as one the strongest biomarkers and prognostic factors for various human cancers (23, 24). The formation of a fibrin network surrounding cancer cells, recently compared to a "bird's nest" (25), could provide a scaffold for cell migration and favorable conditions for cancer cells to resist chemotherapeutic agents (25). Besides the control of the composition and turnover of the tumor extracellular matrix, proteases of the coagulation and fibrinolysis cascades also interact with specific protease-activated receptors (PARs) present on the surface of different cell types (4). We suggest that GC might potentially modulate the active interplay that takes place between the coagulation/fibrinolytic cascades and the tumor microenvironment (4). The GR-*SERPINE1* axis might for example exert potent effects on tumor-associated macrophages (26). GC are well known to modulate the activation and function of cells of the immune system (27). The existence of the GR-*SERPINE1* axis, as supported here with a combination of systems and functional approaches, calls for a broad re-examination of the consequences of GC signaling in tumor physiology and anti-tumor immunity, with particular attention paid to the tumor coagulome.

Conclusion

Overall, our findings suggest that the inhibition of fibrinolysis is a key consequence of cancer cell exposure to GC, possibly leading to the stabilization of the fibrin clot in cancer. Future studies are warranted to address the role of *SERPINE1* induction in this context.

Conflicts of Interest

The Authors have no conflicts of interest to declare in relation to this study.

Authors' Contributions

Floriane Racine: Writing – original draft, investigation, data analysis. Christophe Louandre: Methodology, Investigation; Julien Demagny, Methodology, Investigation; Corinne Godin: Methodology, Investigation; Zuzana Saidak: Writing – original draft, validation, methodology, Investigation, data analysis, Writing - review and editing; Antoine Galmiche: Writing – original draft, validation, conceptualization, funding acquisition.

Acknowledgements

The Authors would like to thank the Amiens University Hospital and the Ligue contre le Cancer, comité de l'Aisne for the financial support.

Funding

This study was financed by the Ligue contre le Cancer, comité de l'Aisne (COAGULCAN).

References

1 Falanga A, Schieppati F, Russo L: Pathophysiology 1. Mechanisms of thrombosis in cancer patients. Cancer Treat Res 179: 11-36, 2019. DOI: 10.1007/978-3-030-20315-3_2
2 Khorana AA, Cohen AT, Carrier M, Meyer G, Pabinger I, Kavan P, Wells P: Prevention of venous thromboembolism in ambulatory patients with cancer. ESMO Open 5(6): e000948, 2020. DOI: 10.1136/esmoopen-2020-000948
3 Saidak Z, Soudet S, Lottin M, Salle V, Sevestre MA, Clatot F, Galmiche A: A pan-cancer analysis of the human tumor coagulome and its link to the tumor immune microenvironment. Cancer Immunol Immunother 70(4): 923-933, 2021. DOI: 10.1007/s00262-020-02739-w
4 Galmiche A, Rak J, Roumenina LT, Saidak Z: Coagulome and the tumor microenvironment: an actionable interplay. Trends Cancer 8(5): 369-383, 2022. DOI: 10.1016/j.trecan.2021.12.008
5 Lottin M, Soudet S, Fercot J, Racine F, Demagny J, Bettoni J, Chatelain D, Sevestre MA, Mammeri Y, Lamuraglia M, Galmiche A, Saidak Z: Molecular landscape of the coagulome of oral squamous cell carcinoma. Cancers (Basel) 14(2): 460, 2022. DOI: 10.3390/cancers14020460
6 Racine F, Soudet S, Sevestre MA, Galmiche A, Saidak Z: The coagulome of oral squamous cell carcinoma: examining the role and regulation of coagulation in oral cancers using a systems approach. Curr Opin Otolaryngol Head Neck Surg 31(2): 73-77, 2023. DOI: 10.1097/moo.0000000000000870
7 Kadmiel M, Cidlowski JA: Glucocorticoid receptor signaling in health and disease. Trends Pharmacol Sci 34(9): 518-530, 2013. DOI: 10.1016/j.tips.2013.07.003
8 Cohen DM, Steger DJ: Nuclear receptor function through genomics: lessons from the glucocorticoid receptor. Trends Endocrinol Metab 28(7): 531-540, 2017. DOI: 10.1016/j.tem.2017.04.001
9 Lin KT, Wang LH: New dimension of glucocorticoids in cancer treatment. Steroids 111: 84-88, 2016. DOI: 10.1016/j.steroids.2016.02.019
10 Weikum ER, Knuesel MT, Ortlund EA, Yamamoto KR: Glucocorticoid receptor control of transcription: precision and plasticity via allostery. Nat Rev Mol Cell Biol 18(3): 159-174, 2017. DOI: 10.1038/nrm.2016.152
11 Racine F, Louandre C, Godin C, Chatelain B, Prekovic S, Zwart W, Galmiche A, Saidak Z: The tumor coagulome as a transcriptional target and a potential effector of glucocorticoids in human cancers. Cancers (Basel) 15(5): 1531, 2023. DOI: 10.3390/cancers15051531
12 Reinhold WC, Sunshine M, Liu H, Varma S, Kohn KW, Morris J, Doroshow J, Pommier Y: CellMiner: a web-based suite of genomic and pharmacologic tools to explore transcript and drug patterns in the NCI-60 cell line set. Cancer Res 72(14): 3499-3511, 2012. DOI: 10.1158/0008-5472.CAN-12-1370
13 West DC, Kocherginsky M, Tonsing-Carter EY, Dolcen DN, Hosfield DJ, Lastra RR, Sinnwell JP, Thompson KJ, Bowie KR, Harkless RV, Skor MN, Pierce CF, Styke SC, Kim CR, de Wet L, Greene GL, Boughey JC, Goetz MP, Kalari KR, Wang L, Fleming GF, Györffy B, Conzen SD: Discovery of a glucocorticoid receptor (GR) activity signature using selective GR antagonism in ER-negative breast cancer. Clin Cancer Res 24(14): 3433-3446, 2018. DOI: 10.1158/1078-0432.CCR-17-2793
14 Gao J, Aksoy BA, Dogrusoz U, Dresdner G, Gross B, Sumer SO, Sun Y, Jacobsen A, Sinha R, Larsson E, Cerami E, Sander C, Schultz N: Integrative analysis of complex cancer genomics and clinical profiles using the cBioPortal. Sci Signal 6(269): pl1, 2013. DOI: 10.1126/scisignal.2004088
15 Prekovic S, Schuurman K, Mayayo-Peralta I, Manjón AG, Buijs M, Yavuz S, Wellenstein MD, Barrera A, Monkhorst K, Huber A, Morris B, Lieftink C, Chalkiadakis T, Alkan F, Silva J, Győrffy B, Hoekman L, van den Broek B, Teunissen H, Debets DO, Severson T, Jonkers J, Reddy T, de Visser KE, Faller W, Beijersbergen R, Altelaar M, de Wit E, Medema R, Zwart W: Glucocorticoid receptor triggers a reversible drug-tolerant dormancy state with acquired therapeutic vulnerabilities in lung cancer. Nat Commun 12(1): 4360, 2021. DOI: 10.1038/s41467-021-24537-3
16 R Core Team. R: A Language and Environment for Statistical Computing. Vienna, Austria, R Foundation for Statistical Computing, 2022. Available at: https://www.R-project.org/ [Last accessed on May 17, 2024]
17 Zhao L, Zhou S, Gustafsson JÅ: Nuclear receptors: recent drug discovery for cancer therapies. Endocr Rev 40(5): 1207-1249, 2019. DOI: 10.1210/er.2018-00222
18 Jeon HM, Kim JY, Cho HJ, Lee WJ, Nguyen D, Kim SS, Oh YT, Kim HJ, Jung CW, Pinero G, Joshi T, Hambardzumyan D, Sakaguchi T, Hubert CG, McIntyre TM, Fine HA, Gladson CL, Wang B, Purow BW, Park JB, Park MJ, Nam DH, Lee J: Tissue factor is a critical regulator of radiation therapy-induced glioblastoma remodeling. Cancer Cell 41(8): 1480-1497.e9, 2023. DOI: 10.1016/j.ccell.2023.06.007
19 Haen P, Mege D, Crescence L, Dignat-George F, Dubois C, Panicot-Dubois L: Thrombosis risk associated with head and neck cancer: a review. Int J Mol Sci 20(11): 2838, 2019. DOI: 10.3390/ijms20112838
20 Isand K, Feelders R, Brue T, Toth M, Deutschbein T, Reincke M, Kršek M, Santos A, Demtröder F, Chabre O, Strasburger CJ, Aulinas Maso A, Volke V, Pereira AM, Lohmann R, Gich Saladich I, Ercusyn Study Group, Webb SM, Wass J, Valassi E: High prevalence of venous thrombotic events in Cushing's

syndrome: data from ERCUSYN and details in relation to surgery. Eur J Endocrinol 190(1): 75-85, 2024. DOI: 10.1093/ejendo/lvad176

21 Einarsdottir MJ, Kibiwott Kirui B, Li H, Olsson D, Johannsson G, Nyberg F, Ragnarsson O: Impact of chronic oral glucocorticoid treatment on mortality in patients with COVID-19: analysis of a population-based cohort. BMJ Open 14(3): e080640, 2024. DOI: 10.1136/bmjopen-2023-080640

22 Majoor CJ, Sneeboer MMS, de Kievit A, Meijers JCM, van der Poll T, Lutter R, Bel EH, Kamphuisen PW: The influence of corticosteroids on hemostasis in healthy subjects. J Thromb Haemost 14(4): 716-723, 2016. DOI: 10.1111/jth.13265

23 Kubala MH, DeClerck YA: The plasminogen activator inhibitor-1 paradox in cancer: a mechanistic understanding. Cancer Metastasis Rev 38(3): 483-492, 2019. DOI: 10.1007/s10555-019-09806-4

24 Morrow GB, Mutch NJ: Past, present, and future perspectives of plasminogen activator inhibitor 1 (PAI-1). Semin Thromb Hemost 49(03): 305-313, 2023. DOI: 10.1055/s-0042-1758791

25 Tran HCM, Mbemba E, Mourot N, Faltas B, Rousseau A, Lefkou E, Sabbah M, van Dreden P, Gerotziafas G: The procoagulant signature of cancer cells drives fibrin network formation in tumor microenvironment and impacts its quality. Implications in cancer cell migration and the resistance to anticancer agents. Thromb Res 238: 172-183, 2024. DOI: 10.1016/j.thromres.2024.04.015

26 Kubala MH, Punj V, Placencio-Hickok VR, Fang H, Fernandez GE, Sposto R, DeClerck YA: Plasminogen activator inhibitor-1 promotes the recruitment and polarization of macrophages in cancer. Cell Rep 25(8): 2177-2191.e7, 2018. DOI: 10.1016/j.celrep.2018.10.082

27 Cain DW, Cidlowski JA: Immune regulation by glucocorticoids. Nat Rev Immunol 17(4): 233-247, 2017. DOI: 10.1038/nri.2017.1

Received June 17, 2024
Revised June 24, 2024
Accepted June 26, 2024