GEORGIAN MEDICAL NEWS

ISSN 1512-0112

NO 5 (374) Май 2026

ТБИЛИСИ - NEW YORK

ЕЖЕМЕСЯЧНЫЙ НАУЧНЫЙ ЖУРНАЛ

Медицинские новости Грузии

საქართველოს სამედიცინო სიახლენი

GEORGIAN MEDICAL NEWS

Monthly Georgia-US joint scientific journal published both in electronic and paper formats of the Agency of Medical Information of the Georgian Association of Business Press. Published since 1994. Distributed in NIS, EU and USA.

GMN: Georgian Medical News is peer-reviewed, published monthly journal committed to promoting the science and art of medicine and the betterment of public health, published by the GMN Editorial Board since 1994. GMN carries original scientific articles on medicine, biology and pharmacy, which are of experimental, theoretical and practical character; publishes original research, reviews, commentaries, editorials, essays, medical news, and correspondence in English and Russian.

GMN is indexed in MEDLINE, SCOPUS, PubMed and VINITI Russian Academy of Sciences. The full text content is available through EBSCO databases.

GMN: Медицинские новости Грузии - ежемесячный рецензируемый научный журнал, издаётся Редакционной коллегией с 1994 года на русском и английском языках в целях поддержки медицинской науки и улучшения здравоохранения. В журнале публикуются оригинальные научные статьи в области медицины, биологии и фармации, статьи обзорного характера, научные сообщения, новости медицины и здравоохранения. Журнал индексируется в MEDLINE, отражён в базе данных SCOPUS, PubMed и ВИНИТИ РАН. Полнотекстовые статьи журнала доступны через БД EBSCO.

GMN: Georgian Medical News – საქართველოს სამედიცინო სიახლენი – არის ყოველთვიური სამეცნიერო სამედიცინო რეცენზირებადი ჟურნალი, გამოიცემა 1994 წლიდან, წარმოადგენს სარედაქციო კოლეგიისა და აშშ-ის მეცნიერების, განათლების, ინდუსტრიის, ხელოვნებისა და ბუნებისმეტყველების საერთაშორისო აკადემიის ერთობლივ გამოცემას. GMN-ში რუსულ და ინგლისურ ენებზე ქვეყნდება ექსპერიმენტული, თეორიული და პრაქტიკული ხასიათის ორიგინალური სამეცნიერო სტატიები მედიცინის, ბიოლოგიისა და ფარმაციის სფეროში, მიმოხილვითი ხასიათის სტატიები.

ჟურნალი ინდექსირებულია MEDLINE-ის საერთაშორისო სისტემაში, ასახულია SCOPUS-ის, PubMed-ის და ВИНИТИ РАН-ის მონაცემთა ბაზებში. სტატიების სრული ტექსტი ხელმისაწვდომია EBSCO-ს მონაცემთა ბაზებიდან.

WEBSITE

www.geomednews.com

К СВЕДЕНИЮ АВТОРОВ!

При направлении статьи в редакцию необходимо соблюдать следующие правила:

1. Статья должна быть представлена в двух экземплярах, на русском или английском языках, напечатанная через **полтора интервала на одной стороне стандартного листа с шириной левого поля в три сантиметра**. Используемый компьютерный шрифт для текста на русском и английском языках - **Times New Roman** (**Кириллица**), для текста на грузинском языке следует использовать **AcadNusx.** Размер шрифта - **12**. К рукописи, напечатанной на компьютере, должен быть приложен CD со статьей.

2. Размер статьи должен быть не менее десяти и не более двадцати страниц машинописи, включая указатель литературы и резюме на английском, русском и грузинском языках.

3. В статье должны быть освещены актуальность данного материала, методы и результаты исследования и их обсуждение.

При представлении в печать научных экспериментальных работ авторы должны указывать вид и количество экспериментальных животных, применявшиеся методы обезболивания и усыпления (в ходе острых опытов).

4. К статье должны быть приложены краткое (на полстраницы) резюме на английском, русском и грузинском языках (включающее следующие разделы: цель исследования, материал и методы, результаты и заключение) и список ключевых слов (key words).

5. Таблицы необходимо представлять в печатной форме. Фотокопии не принимаются. **Все цифровые, итоговые и процентные данные в таблицах должны соответствовать таковым в тексте статьи**. Таблицы и графики должны быть озаглавлены.

6. Фотографии должны быть контрастными, фотокопии с рентгенограмм - в позитивном изображении. Рисунки, чертежи и диаграммы следует озаглавить, пронумеровать и вставить в соответствующее место текста **в tiff формате**.

В подписях к микрофотографиям следует указывать степень увеличения через окуляр или объектив и метод окраски или импрегнации срезов.

7. Фамилии отечественных авторов приводятся в оригинальной транскрипции.

8. При оформлении и направлении статей в журнал МНГ просим авторов соблюдать правила, изложенные в «Единых требованиях к рукописям, представляемым в биомедицинские журналы», принятых Международным комитетом редакторов медицинских журналов - http://www.spinesurgery.ru/files/publish.pdf и http://www.nlm.nih.gov/bsd/uniform_requirements.html
В конце каждой оригинальной статьи приводится библиографический список. В список литературы включаются все материалы, на которые имеются ссылки в тексте. Список составляется в алфавитном порядке и нумеруется. Литературный источник приводится на языке оригинала. В списке литературы сначала приводятся работы, написанные знаками грузинского алфавита, затем кириллицей и латиницей. Ссылки на цитируемые работы в тексте статьи даются в квадратных скобках в виде номера, соответствующего номеру данной работы в списке литературы. Большинство цитированных источников должны быть за последние 5-7 лет.

9. Для получения права на публикацию статья должна иметь от руководителя работы или учреждения визу и сопроводительное отношение, написанные или напечатанные на бланке и заверенные подписью и печатью.

10. В конце статьи должны быть подписи всех авторов, полностью приведены их фамилии, имена и отчества, указаны служебный и домашний номера телефонов и адреса или иные координаты. Количество авторов (соавторов) не должно превышать пяти человек.

11. Редакция оставляет за собой право сокращать и исправлять статьи. Корректура авторам не высылается, вся работа и сверка проводится по авторскому оригиналу.

12. Недопустимо направление в редакцию работ, представленных к печати в иных издательствах или опубликованных в других изданиях.

При нарушении указанных правил статьи не рассматриваются.

REQUIREMENTS

Please note, materials submitted to the Editorial Office Staff are supposed to meet the following requirements:

1. Articles must be provided with a double copy, in English or Russian languages and typed or compu-ter-printed on a single side of standard typing paper, with the left margin of **3** centimeters width, and **1.5** spacing between the lines, typeface - **Times New Roman** (**Cyrillic**), print size - **12** (referring to Georgian and Russian materials). With computer-printed texts please enclose a CD carrying the same file titled with Latin symbols.

2. Size of the article, including index and resume in English, Russian and Georgian languages must be at least 10 pages and not exceed the limit of 20 pages of typed or computer-printed text.

3. Submitted material must include a coverage of a topical subject, research methods, results, and review.

Authors of the scientific-research works must indicate the number of experimental biological species drawn in, list the employed methods of anesthetization and soporific means used during acute tests.

4. Articles must have a short (half page) abstract in English, Russian and Georgian (including the following sections: aim of study, material and methods, results and conclusions) and a list of key words.

5. Tables must be presented in an original typed or computer-printed form, instead of a photocopied version. **Numbers, totals, percentile data on the tables must coincide with those in the texts of the articles.** Tables and graphs must be headed.

6. Photographs are required to be contrasted and must be submitted with doubles. Please number each photograph with a pencil on its back, indicate author's name, title of the article (short version), and mark out its top and bottom parts. Drawings must be accurate, drafts and diagrams drawn in Indian ink (or black ink). Photocopies of the X-ray photographs must be presented in a positive image in **tiff format**.

Accurately numbered subtitles for each illustration must be listed on a separate sheet of paper. In the subtitles for the microphotographs please indicate the ocular and objective lens magnification power, method of coloring or impregnation of the microscopic sections (preparations).

7. Please indicate last names, first and middle initials of the native authors, present names and initials of the foreign authors in the transcription of the original language, enclose in parenthesis corresponding number under which the author is listed in the reference materials.

8. Please follow guidance offered to authors by The International Committee of Medical Journal Editors guidance in its Uniform Requirements for Manuscripts Submitted to Biomedical Journals publication available online at: http://www.nlm.nih.gov/bsd/uniform_requirements.html
http://www.icmje.org/urm_full.pdf
In GMN style for each work cited in the text, a bibliographic reference is given, and this is located at the end of the article under the title "References". All references cited in the text must be listed. The list of references should be arranged alphabetically and then numbered. References are numbered in the text [numbers in square brackets] and in the reference list and numbers are repeated throughout the text as needed. The bibliographic description is given in the language of publication (citations in Georgian script are followed by Cyrillic and Latin).

9. To obtain the rights of publication articles must be accompanied by a visa from the project instructor or the establishment, where the work has been performed, and a reference letter, both written or typed on a special signed form, certified by a stamp or a seal.

10. Articles must be signed by all of the authors at the end, and they must be provided with a list of full names, office and home phone numbers and addresses or other non-office locations where the authors could be reached. The number of the authors (co-authors) must not exceed the limit of 5 people.

11. Editorial Staff reserves the rights to cut down in size and correct the articles. Proof-sheets are not sent out to the authors. The entire editorial and collation work is performed according to the author's original text.

12. Sending in the works that have already been assigned to the press by other Editorial Staffs or have been printed by other publishers is not permissible.

Articles that Fail to Meet the Aforementioned Requirements are not Assigned to be Reviewed.

ავტორთა საყურადღებოდ!

რედაქციაში სტატიის წარმოდგენისას საჭიროა დავიცვათ შემდეგი წესები:

1. სტატია უნდა წარმოადგინოთ 2 ცალად, რუსულ ან ინგლისურ ენებზე,დაბეჭდილი სტანდარტული ფურცლის 1 გვერდზე, 3 სმ სიგანის მარცხენა ველისა და სტრიქონებს შორის 1,5 ინტერვალის დაცვით. გამოყენებული კომპიუტერული შრიფტი რუსულ და ინგლისურენოვან ტექსტებში - **Times New Roman (Кириллица)**, ხოლო ქართულენოვან ტექსტში საჭიროა გამოვიყენოთ **AcadNusx.** შრიფტის ზომა – 12. სტატიას თან უნდა ახლდეს CD სტატიით.

2. სტატიის მოცულობა არ უნდა შეადგენდეს 10 გვერდზე ნაკლებს და 20 გვერდზე მეტს ლიტერატურის სიის და რეზიუმეების (ინგლისურ, რუსულ და ქართულ ენებზე) ჩათვლით.

3. სტატიაში საჭიროა გაშუქდეს: საკითხის აქტუალობა; კვლევის მიზანი; საკვლევი მასალა და გამოყენებული მეთოდები; მიღებული შედეგები და მათი განსჯა. ექსპერიმენტული ხასიათის სტატიების წარმოდგენისას ავტორებმა უნდა მიუთითონ საექსპერიმენტო ცხოველების სახეობა და რაოდენობა; გაუტკივარებისა და დაძინების მეთოდები (მწვავე ცდების პირობებში).

4. სტატიას თან უნდა ახლდეს რეზიუმე ინგლისურ, რუსულ და ქართულ ენებზე არანაკლებ ნახევარი გვერდის მოცულობისა (სათაურის, ავტორების, დაწესებულების მითითებით და უნდა შეიცავდეს შემდეგ განყოფილებებს: მიზანი, მასალა და მეთოდები, შედეგები და დასკვნები; ტექსტუალური ნაწილი არ უნდა იყოს 15 სტრიქონზე ნაკლები) და საკვანძო სიტყვების ჩამონათვალი (key words).

5. ცხრილები საჭიროა წარმოადგინოთ ნაბეჭდი სახით. ყველა ციფრული, შემაჯამებელი და პროცენტული მონაცემები უნდა შეესაბამებოდეს ტექსტში მოყვანილს.

6. ფოტოსურათები უნდა იყოს კონტრასტული; სურათები, ნახაზები, დიაგრამები - დასათაურებული, დანომრილი და სათანადო ადგილას ჩასმული. რენტგენოგრამების ფოტოასლები წარმოადგინეთ პოზიტიური გამოსახულებით **tiff** ფორმატში. მიკროფოტოსურათების წარწერებში საჭიროა მიუთითოთ ოკულარის ან ობიექტივის საშუალებით გადიდების ხარისხი, ანათალების შეღებვის ან იმპრეგნაციის მეთოდი და აღნიშნოთ სურათის ზედა და ქვედა ნაწილები.

7. სამამულო ავტორების გვარები სტატიაში აღინიშნება ინიციალების თანდართვით, უცხოურისა – უცხოური ტრანსკრიპციით.

8. სტატიას თან უნდა ახლდეს ავტორის მიერ გამოყენებული სამამულო და უცხოური შრომების ბიბლიოგრაფიული სია (ბოლო 5-8 წლის სიღრმით). ანბანური წყობით წარმოდგენილ ბიბლიოგრაფიულ სიაში მიუთითეთ ჯერ სამამულო, შემდეგ უცხოელი ავტორები (გვარი, ინიციალები, სტატიის სათაური, ჟურნალის დასახელება, გამოცემის ადგილი, წელი, ჟურნალის №, პირველი და ბოლო გვერდები). მონოგრაფიის შემთხვევაში მიუთითეთ გამოცემის წელი, ადგილი და გვერდების საერთო რაოდენობა. ტექსტში კვადრატულ ფჩხილებში უნდა მიუთითოთ ავტორის შესაბამისი N ლიტერატურის სიის მიხედვით. მიზანშეწონილია, რომ ციტირებული წყაროების უმეტესი ნაწილი იყოს 5-6 წლის სიღრმის.

9. სტატიას თან უნდა ახლდეს: ა) დაწესებულების ან სამეცნიერო ხელმძღვანელის წარდგინება, დამოწმებული ხელმოწერითა და ბეჭდით; ბ) დარგის სპეციალისტის დამოწმებული რეცენზია, რომელშიც მითითებული იქნება საკითხის აქტუალობა, მასალის საკმაობა, მეთოდის სანდოობა, შედეგების სამეცნიერო-პრაქტიკული მნიშვნელობა.

10. სტატიის ბოლოს საჭიროა ყველა ავტორის ხელმოწერა, რომელთა რაოდენობა არ უნდა აღემატებოდეს 5-ს.

11. რედაქცია იტოვებს უფლებას შეასწოროს სტატია. ტექსტზე მუშაობა და შეჯერება ხდება საავტორო ორიგინალის მიხედვით.

12. დაუშვებელია რედაქციაში ისეთი სტატიის წარდგენა, რომელიც დასაბეჭდად წარდგენილი იყო სხვა რედაქციაში ან გამოქვეყნებული იყო სხვა გამოცემებში.

აღნიშნული წესების დარღვევის შემთხვევაში სტატიები არ განიხილება.

GEORGIAN MEDICAL NEWS
NO 5 (374) 2026

Содержание:

GEORGIAN MEDICAL NEWS
No 5 (374) 2026

PAN-CANCER ANALYSIS OF CHEMOKINE (C-C MOTIF) LIGAND 26 (CCL26) AS A PROMISING PROGNOSTIC BIOMARKER AND IMMUNOMODULATORY MEDIATOR

Sara Ali1*, Marwan Ismail[1], Praveen kumar[2], Salma Elnour Mohamed[1], Weam Alyoubi[3], Hiba Mohamed[4], Raghad Alamri[5], Fatima Mohamed Osman Yasin[6], Safa Mohamed Abdelrahman[7], Huda F. Alshaibi[7], Einas Awad Osman[8], Akhtamova Shahzoda Fozilovna[9], Matlyuba Badritdnova[10], Rihab Akasha[11], Mohamed Alfaki[12].

[1]Department of Medical Laboratory Sciences, College of Health Sciences, Gulf Medical University, Ajman, United Arab Emirates.

[2]Department of Physiotherapy, College of Health Sciences, Gulf Medical University, Ajman, United Arab Emirates.

[3]Department of Biological Science, Faculty of Science, King Abdulaziz University, Jeddah 21589, Saudi Arabia.

[4]Faculty of Medical Laboratory Sciences, University of Gezira, Sudan.

[5]Department of Medical Laboratory Sciences, Faculty of Applied Medical Sciences, King Abdulaziz University, Saudi Arabia.

[6]Faculty of Medical Laboratory Sciences, University of Gadarif, Gadarif, Sudan.

[7]King Abdul Aziz University – Applied College – Health Information Technology Department – Medical Sterilization.

[8]Collage of Applied and Health Sciences, A'Sharqiyah University, Ibra, Oman.

[9]Department of Medical Rehabilitation, Sports Medicine and Traditional Medicine Samarkand, Samarkand state medical university, Uzbekistan.

[10]Department of Internal Medicine, Bukhara State Medical Institute, Uzbekistan

[11]University of Hail,College of Applied Medical Sciences, Department of Clinival Laboratory Sciences, Saudi Arabia.

[12]Software Engineering Department, Faculty of Computer Science, Al-Neelain University, Sudan.

Abstract.

Background: Chemokine (C-C motif) ligand 26 (CCL26), also known as eotaxin-3, is an immune-regulatory chemokine involved in inflammatory responses and immune cell recruitment. Emerging evidence suggests its expression is dysregulated across multiple malignancies, yet comprehensive pan-cancer evaluations remain limited.

Objectives: To systematically analyze CCL26 expression, clinicopathological correlations, survival associations, and immune modulatory functions across 33 cancer types, and evaluate its potential as a prognostic biomarker and immunomodulatory therapeutic target.

Methods: CCL26 expression was analyzed using GEPIA, TIMER2.0, and UALCAN, three platforms that share the TCGA data source but employ distinct analytical pipelines; cross-platform concordance was therefore used as a measure of pipeline robustness, not validation. external validation was performed using four GEO microarray cohorts (GSE53757, GSE30219, GSE39582, and GSE33479)

Results: CCL26 was significantly upregulated in 11 cancer types (33.33%), particularly epithelial malignancies including lung squamous cell carcinoma (LUSC), colon adenocarcinoma (COAD), and esophageal carcinoma (ESCA), and downregulated in genitourinary cancers including kidney renal papillary cell carcinoma (KIRP) and prostate adenocarcinoma (PRAD). Cross-database validation achieved 96.97% concordance. Expression correlated with advancing cancer stage, older age (61–80 years), and racial/ethnic background. Survival analysis revealed opposing prognostic associations: high CCL26 predicted poor outcomes in LIHC, KIRC, and KIRP (HR 1.45–3.79, $p<0.036$), but favorable outcomes in STAD and ESCA (HR 0.35–0.71, $p<0.047$). Significant immune correlations were identified with dendritic cells (LUAD, $r=0.373$), macrophages (COAD, $r=0.318$), and CD4+ T cells (STAD, $r=0.296$). Promoter hypermethylation was identified in lung and breast cancers, with minimal genomic mutation frequency (1%).

Conclusions: CCL26 exhibits cancer-type-specific expression and differential prognostic significance across malignancies. Its strong immune infiltration correlations support potential utility in prognostic panels and as an immunomodulatory target. GEO-based validation supported selected expression findings, particularly in renal and lung cohorts, while also highlighting context-dependent variability. Collectively, these data support the biological relevance of CCL26 in cancer but indicate that its clinical and therapeutic value should be interpreted in a tumor-specific manner and confirmed in further mechanistic studies.

Key words. CCL26, pan-cancer analysis, biomarker, prognosis, immune infiltration, immunomodulation, bioinformatics, epigenetic regulation.

Introduction.

Chemokines represent critical mediators of immune cell migration and play dual roles in cancer biology: antimicrobial immunity and tumorigenic effects [1]. Recent advances in cancer genomics have identified numerous chemokine ligands as potential biomarkers with prognostic significance across hematologic and solid tumors [2]. The chemokine/chemokine receptor axis contributes to development of immunosuppressive tumor microenvironments and protects developing tumors from immune surveillance [3].

Chemokine (C-C motif) ligand 26 (CCL26), also known as eotaxin-3, is predominantly expressed in epithelial tissues and plays important roles in immune regulation and inflammatory responses [4]. Normally, CCL26 functions to recruit and activate immune cells at inflammatory sites. However, emerging evidence suggests that in malignant contexts, CCL26 may influence cancer progression through interactions with stem cell development pathways and immune response regulatory networks. This dual functionality makes CCL26 particularly intriguing as a prognostic biomarker.

Previous studies have identified dysregulated CCL26 expression in various malignancies, suggesting context-dependent roles in tumor biology [5]. However, comprehensive

pan-cancer evaluations remain limited. This study addresses this gap through systematic analysis of CCL26 expression patterns, clinicopathological correlations, survival associations, and immune modulatory functions across 33 cancer types.

Study Objectives:

- Evaluate CCL26 expression across multiple cancer types.
- Correlate expression with clinicopathological parameters and patient outcomes.
- Assess relationships between CCL26 and immune cell infiltration.
- Identify genetic and epigenetic mechanisms of CCL26 dysregulation.
- Determine potential clinical utility as a stratification and prognostic biomarker.

Materials and Methods.

Data Sources and Bioinformatics Tools:

CCL26 expression analysis employed four major publicly accessible bioinformatics platforms accessed January 2025:

GEPIA Database: (http://gepia.cancer-pku.cn/) [6]: Analyzes RNA-sequencing data from The Cancer Genome Atlas (TCGA) and Genotype-Tissue Expression (GTEx) projects. Enables comparison of tumor versus normal tissue expression across 33 cancer types.

TIMER2.0 Database: (http://timer.cistrome.org/ and https://cistrome.shinyapps.io/timer/) [7]: Provides comprehensive tumor-infiltrating immune cell estimates alongside gene expression data, enabling correlation analyses.

UALCAN Database: (https://ualcan.path.uab.edu/) [8,9]: Facilitates clinicopathological stratification analysis including cancer stage, patient age, gender, and race/ethnicity.

Kaplan-Meier Plotter: (https://kmplot.com/analysis) [10]: Enables survival analysis with hazard ratio calculation and log-rank test statistics.

Expression Analysis:

Differential expression of CCL26 between tumor and normal tissues was investigated across 33 cancer types. Expression categorization employed the following criteria: significant upregulation ($p < 0.05$ with fold-change >1.5), significant downregulation ($p < 0.05$ with fold-change <0.67), and non-significant changes. P-values were determined using t-tests with multiple testing corrections where applicable.

Clinicopathological Correlation Analysis:

CCL26 expression was stratified by: (1) cancer stage (Stages I–IV), (2) patient age (21–40, 41–60, 61–80, 81–100 years), and (3) race/ethnicity (Caucasian, African American, Asian). Statistical significance was assessed using ANOVA with post-hoc testing where appropriate.

Survival Analysis:

Overall survival (OS) and disease-free survival (DFS) were evaluated using Kaplan-Meier curves stratified by CCL26 expression (high vs. low, median expression cutoff). Hazard ratios (HR) with 95% confidence intervals were calculated using log-rank tests (10). P-values <0.05 were considered statistically significant.

Immune Cell Infiltration Analysis:

Correlations between CCL26 expression and tumor-infiltrating immune populations (CD8+ T cells, CD4+ T cells, macrophages, dendritic cells, B cells, and neutrophils) were quantified using Spearman's correlation coefficient (7). Correlation strength was classified as: strong ($r > 0.5$), moderate ($0.3 < r \leq 0.5$), or weak ($r \leq 0.3$).

Genetic Alteration and Methylation Analysis:

cBioPortal (https://www.cbioportal.org/) [11] was used to assess mutation frequency and copy number variations affecting CCL26. Promoter methylation analysis examined DNA methylation status across cancer types, comparing tumor versus normal tissue methylation patterns (hypermethylation vs. hypomethylation) [12,13].

Gene Interaction Network Analysis:

GeneMANIA (http://www.genemania.org/) generated co-expression networks and identified functional relationships, categorizing interactions by type: co-expression, physical protein-protein interactions, predicted interactions, shared protein domains, pathway involvement, and genetic interactions.

Cross-Platform Concordance Analysis:

To enhance analytical robustness and reduce potential platform-specific bias, results from the three TCGA-based analytical platforms (GEPIA, TIMER2.0, and UALCAN) were systematically compared. Concordance was defined as agreement in the direction of expression change (upregulation or downregulation) and statistical significance ($p < 0.05$) across the platforms. Since these tools primarily utilize overlapping TCGA-derived datasets, the observed agreement should be interpreted as cross-platform analytical concordance rather than biological validation. external validation was subsequently performed using separate GEO cohorts derived from non-TCGA patient populations [6-8].

Statistical Methods and Reproducibility:

Expression Classification: For survival analyses, CCL26 expression was stratified using median expression value as cutoff within each cancer cohort to define "high" (≥median) versus "low" (<median) expression groups. This approach maintains balanced group sizes and is standard for biomarker stratification studies [10].

Multiple Testing Correction: All p-values reported from bioinformatics database tools reflect corrections applied internally by GEPIA, TIMER2.0, and UALCAN platforms. TIMER and UALCAN apply FDR (False Discovery Rate) correction with Benjamini-Hochberg adjustment [7,9]; GEPIA applies Benjamini-Hochberg adjustment [6].

Cutoff Definition for Significance: Statistical significance threshold set at $p < 0.05$ for all analyses. For survival analysis, log-rank test p-values reported [10]; for correlation analysis, Spearman correlation p-values reported [7]; for expression comparisons, t-test or Mann-Whitney U test p-values reported as determined by each platform.

Batch Correction: All three primary databases (GEPIA, TIMER, UALCAN) employ standardized data preprocessing including normalization and batch effect correction using established TCGA pipelines [6-8]. No additional batch correction was performed by authors.

Reproducibility: Results are fully reproducible by accessing the same databases using the same cancer type definitions and default parameters of each platform.

External GEO-based validation: To assess the reproducibility of selected discovery-phase expression findings, external validation was performed using Gene Expression Omnibus (GEO) cohorts with available processed microarray expression data. Three cohorts were suitable for tumor-versus-normal differential expression analysis: GSE53757 for clear cell renal cell carcinoma (KIRC), GSE30219 for lung tumors, and GSE39582 for colon adenocarcinoma (COAD). In addition, GSE33479 was used as a progression-oriented cohort to examine CCL26 expression across the histologic stages of squamous carcinogenesis. For hepatocellular carcinoma, uploaded GSE14520 platform-specific matrices were screened, but a usable CCL26 probe was not present in the available uploaded files; therefore, this cohort was not included in the external validation analysis.

GEO datasets were identified through manual searches of the Gene Expression Omnibus (GEO) database using combinations of the terms "CCL26," "cancer," "tumor," "carcinoma," "expression," and individual cancer-type names identified during the TCGA-based discovery phase (e.g., renal cell carcinoma, lung cancer, colon adenocarcinoma). Dataset inclusion criteria were predefined and included: (1) availability of publicly accessible processed expression matrices or series matrix files; (2) inclusion of tumor and non-tumoral/control tissues or progression-stage samples; (3) adequate sample size to support statistical comparison; (4) compatible probe annotation for CCL26 expression analysis; and (5) human clinical tissue samples rather than cell-line-only datasets. Preference was given to cohorts with matched tumor-normal designs, larger sample numbers, and clear clinicopathological annotation. Datasets lacking usable CCL26 probe annotation, incomplete processed data, or insufficient control samples were excluded from validation analyses.

Series matrix files were used as the source of processed expression values. Sample groups were defined according to GEO sample annotations. For GSE53757, tumor samples were compared with matched normal kidney tissues. For GSE30219, lung tumor samples were compared with non-tumoral lung tissues. For GSE39582, colon adenocarcinoma samples were compared with non-tumoral colorectal mucosa. In GSE33479, normal bronchial epithelium, premalignant lesions, and squamous cell carcinoma samples were evaluated as a progression spectrum rather than only a binary tumor-versus-normal contrast.

Genome-wide differential expression analysis was performed for each tumor-versus-normal cohort by calculating log2 fold change as the difference in mean expression between tumor and normal groups. Two-sided Welch's t-tests were used to compare expression values between groups for each probe, and p-values were adjusted for multiple testing using the Benjamini–Hochberg false discovery rate (FDR) method. Volcano plots were generated for each cohort, with the x-axis representing log2 fold change and the y-axis representing −log10(p-value). CCL26 was labeled using probe 223710_at in GSE53757, GSE30219, and GSE39582, and probe A_23_P215484 in GSE33479.

For the progression-oriented GSE33479 cohort, CCL26 expression was compared between normal and squamous cell carcinoma samples, and the relationship between CCL26 expression and histologic progression stage was additionally assessed using Spearman rank correlation. Statistical significance was defined as $p < 0.05$.

Results.

CCL26 Expression Distribution Across Cancer Types.

GEPIA Database Analysis:

Significant CCL26 upregulation was identified in 2 cancer types (6.06%): lung squamous cell carcinoma (LUSC) and pancreatic adenocarcinoma (PAAD). Testicular germ cell tumors (TGCT) showed elevated expression at borderline significance. Conversely, significant downregulation occurred in 3 cancers (9.09%): acute myeloid leukemia (LAML), ovarian serous cystadenocarcinoma (OV), and uterine corpus endometrial carcinoma (UCEC). Non-significant upregulation appeared in 13 additional cancers (39.39%), while 12 cancers (36.36%) demonstrated downregulation (Figure 1).

TIMER2.0 Database Analysis:

Significantly elevated CCL26 expression characterized 9 cancer types (27.27%): cholangiocarcinoma (CHOL), colon adenocarcinoma (COAD), oesophageal carcinoma (ESCA), head and neck squamous cell carcinoma (HNSC), liver hepatocellular carcinoma (LIHC), lung adenocarcinoma (LUAD), lung squamous cell carcinoma (LUSC), gastric adenocarcinoma (STAD), and thyroid carcinoma (THCA) (all $p < 0.001$). Significant downregulation affected 5 cancer types (15.15%): kidney renal clear cell carcinoma (KIRC, $p < 0.05$), bladder urothelial carcinoma (BLCA, $p < 0.01$), prostate adenocarcinoma (PRAD, $p < 0.01$), kidney renal papillary cell carcinoma (KIRP, $p < 0.001$), and UCEC ($p < 0.001$) (Figure 2).

UALCAN Database Analysis:

Significant upregulation was confirmed in 10 cancer types (30.30%): CHOL, COAD, ESCA, HNSC, LIHC, LUAD, LUSC, STAD, THCA, and TGCT. Significant downregulation occurred in 4 cancer types (12.12%): KIRP, PRAD, TGCT, and UCEC. Data were unavailable for 8 cancer types (24.24%) (Figure 3) S1.

CCL26 expression patterns across three bioinformatics platforms—GEPIA, TIMER2.0, and UALCAN. Several epithelial malignancies, including lung squamous cell carcinoma (LUSC), cholangiocarcinoma (CHOL), colon adenocarcinoma (COAD), esophageal carcinoma (ESCA), head and neck squamous cell carcinoma (HNSC), liver hepatocellular carcinoma (LIHC), lung adenocarcinoma (LUAD), gastric adenocarcinoma (STAD), thyroid carcinoma (THCA), and testicular germ cell tumors (TGCT), consistently exhibited CCL26 upregulation, with statistical significance observed in at least two platforms.

In contrast, uterine corpus endometrial carcinoma (UCEC) showed consistent CCL26 downregulation across all three Platform. For pancreatic adenocarcinoma (PAAD), CCL26

upregulation was observed in GEPIA and TIMER, while UALCAN data were unavailable. Overall, this cross- Platform agreement highlights a subset of tumor types with robust and reproducible CCL26 expression patterns, supporting the reliability of the observed dysregulation and justifying their prioritization for downstream prognostic and immunological analyses (Figure 4).

Cross-Platform Concordance of Expression Patterns:

Analysis of all three databases revealed concordant significant upregulation in LUSC across all platforms. Both TIMER and UALCAN concordantly identified significant upregulation in CHOL, COAD, ESCA, HNSC, LIHC, LUAD, STAD, and THCA (83.3% concordance in high-expressing tumors). Concordant downregulation was observed in KIRP and PRAD (83.4% concordance), and UCEC across all three Platform (100% concordance). Overall cross-platform concordance rate: 96.97% (32/33 cancers show concordant patterns across at least 2 databases) (Figure 5).

CCL26 Expression Correlates with Cancer Stage:

Stage-specific analysis revealed progressive CCL26 elevation across tumor stages in multiple epithelial cancers. Lung squamous cell carcinoma demonstrated significant upregulation across all stages (Stage I: $p = 3.31 \times 10^{-12}$, Stage II: $p = 5.05 \times 10^{-7}$, Stage III: $p = 5.94 \times 10^{-6}$, Stage IV: $p = 2.36 \times 10^{-2}$). Similar patterns appeared in LUAD, COAD, and THCA across all four stages. Cholangiocarcinoma showed significant upregulation in Stages I, II, and IV (p values: 9.04×10^{-3}, 5.86×10^{-3}, 3.59×10^{-2} respectively). Esophageal carcinoma exhibited significant upregulation in Stages II and III ($p = 2.95 \times 10^{-9}$ and 3.71×10^{-5}) (Figure 6) S2.

Conversely, downregulated tumors showed inverse patterns. Kidney renal papillary cell carcinoma demonstrated significantly higher expression in normal tissue compared to Stage 1 ($p = 1.15 \times 10^{-2}$) and Stage 2 ($p = 4.80 \times 10^{-2}$). Similarly, UCEC showed normal-tissue predominance at Stage 2 ($p = 2.93 \times 10^{-2}$) and Stage 3 ($p = 2.90 \times 10^{-2}$) (Figure 7).

CCL26 Expression Correlates with Patient Age:

Stage- age-related expression variations were observed across multiple cancer types. Cholangiocarcinoma, head and neck squamous cell carcinoma, and liver hepatocellular carcinoma showed significantly elevated tumor expression (vs. normal) predominantly in the 41–60 and 61–80-year age groups (p values ranging from 1.91×10^{-5} to 9.24×10^{-3}). Lung adenocarcinoma and lung squamous cell carcinoma demonstrated consistent upregulation across all age groups (41–80+ years), with maximal significance in the 61–80-year cohort (LUSC: $p < 1 \times 10^{-13}$). Thyroid carcinoma exhibited remarkably consistent upregulation across all age groups (p values 3.40×10^{-14} to 9.78×10^{-3}). Colon adenocarcinoma demonstrated age-progressive upregulation from 21–40 years through 61–80 years (p values 1.85×10^{-2} to 5.86×10^{-8}) (Figure 8).

In downregulated tumors, normal tissue expression exceeded tumor expression particularly in older age groups. Uterine corpus endometrial carcinoma showed normal > tumor at ages 41–60 years ($p = 3.66 \times 10^{-2}$) and 61–80 years ($p = 2.74 \times 10^{-2}$). Kidney renal papillary cell carcinoma demonstrated normal-tissue predominance across multiple age groups (Figure 9).

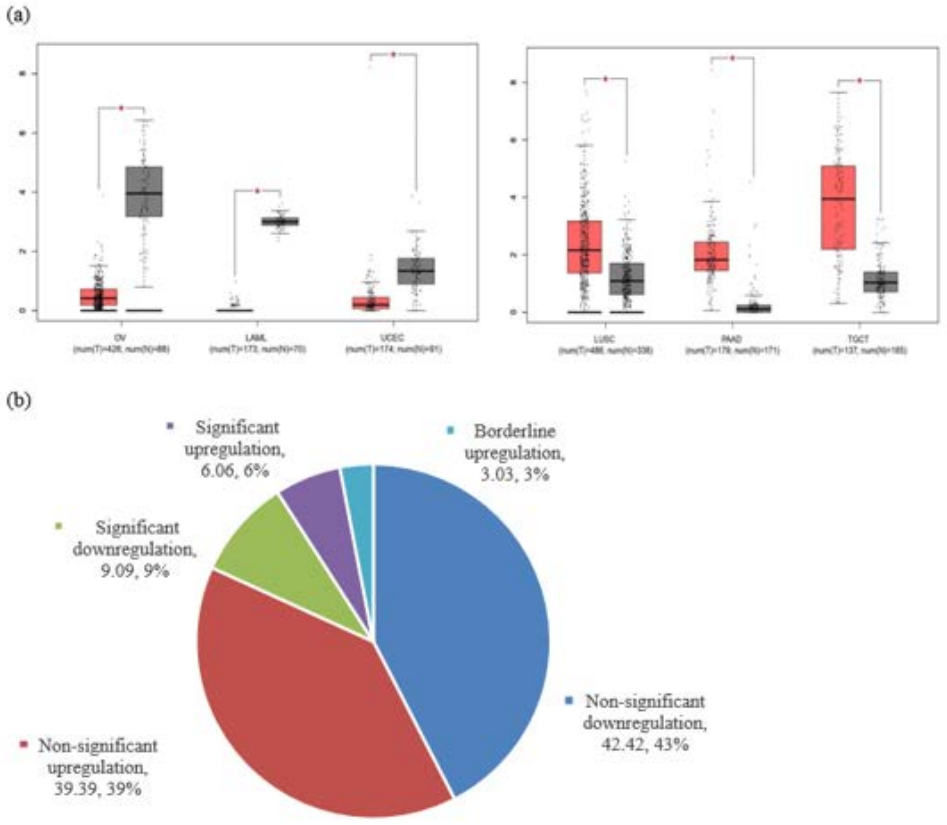


Figure 1. *(a) CCL26 expression analysis in various tumours using the GEPIA database. (b) Distribution of CCL26 Expression Patterns Across Cancer Types (GEPIA Analysis).*

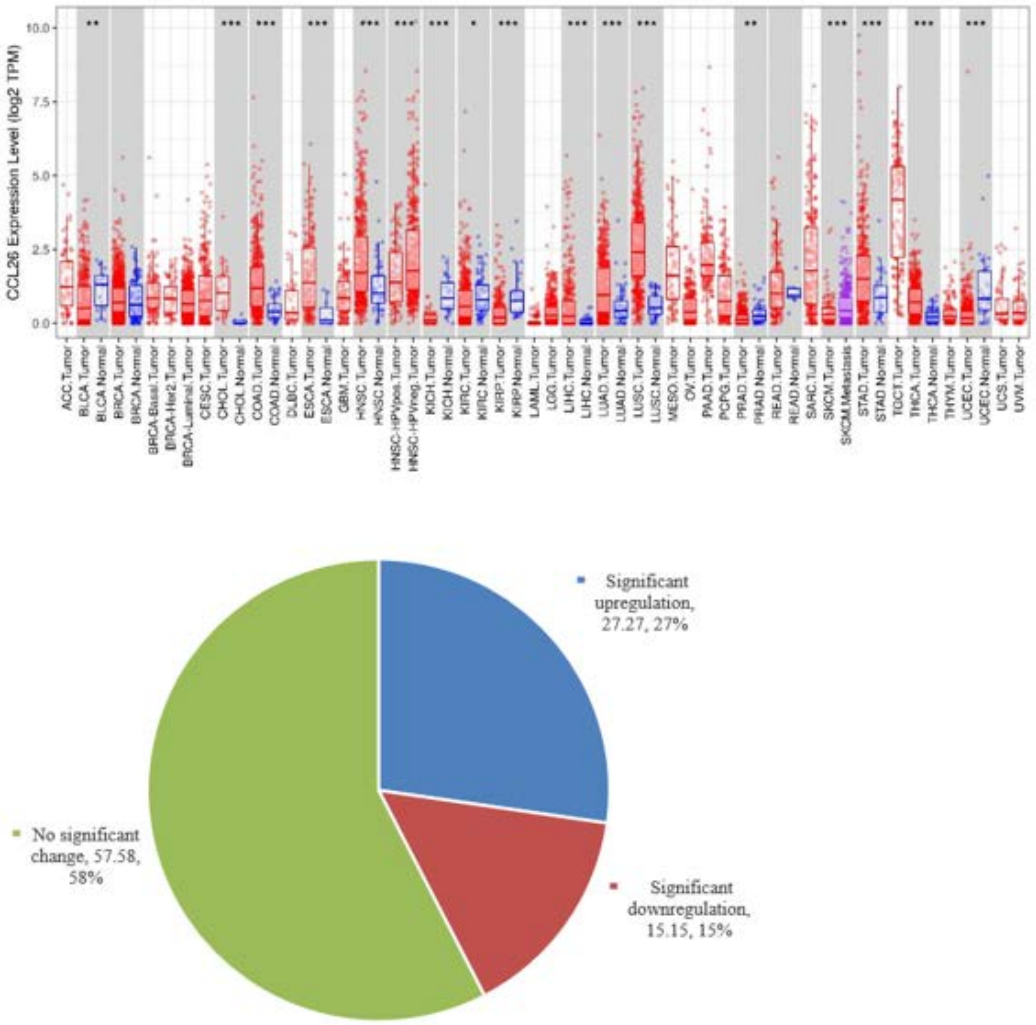


Figure 2. Differential expression between tumour and normal tissues for CCL26 gene across all TCGA tumours.
TIMER2.0 database (a) The box plot shows the expression of the CCL26 gene across various tumour (red) and normal (blue) tissue samples. Significant expression differences between tumour and normal tissues are indicated by stars (: p-value < 0.05: p-value <0.01; *: p-value <0.001), (b) Distribution of CCL26 Expression Patterns Across Cancer Types (TIMER2.0).*

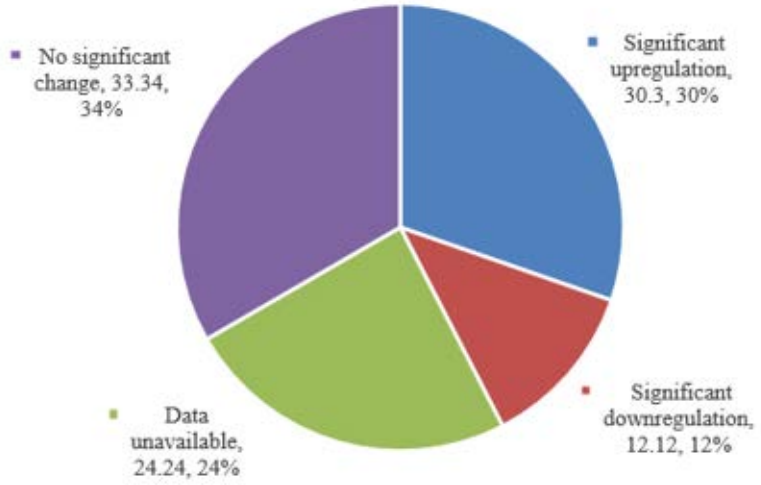


Figure 3. *CCL26 expression analysis in various tumours using the UALCAN database Distribution of CCL26 Expression Patterns Across Cancer Types (UALCAN Analysis).*

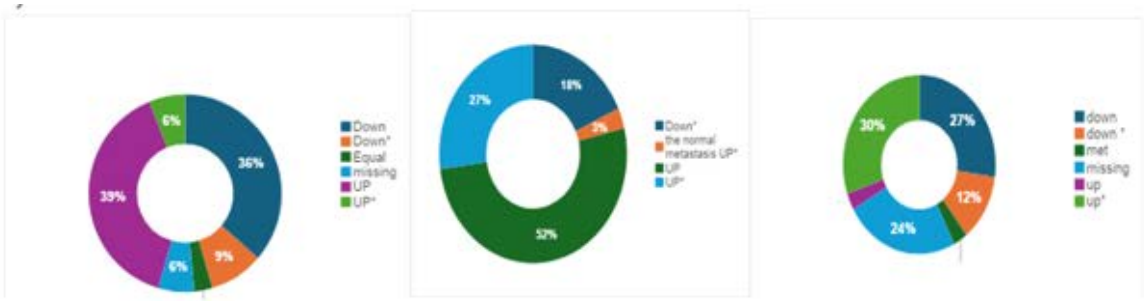


Figure 4. *The distribution of CCL26 expressions among tumour and normal samples: GEPIA, TIMER2.0 and UACLAN database.*

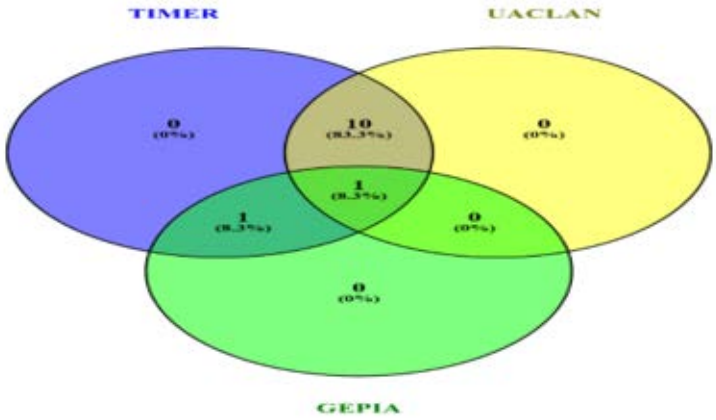


Figure 5. *Cross expression of CCL26 in Cancer: TIMER, GEPIA and UALCAN database.*

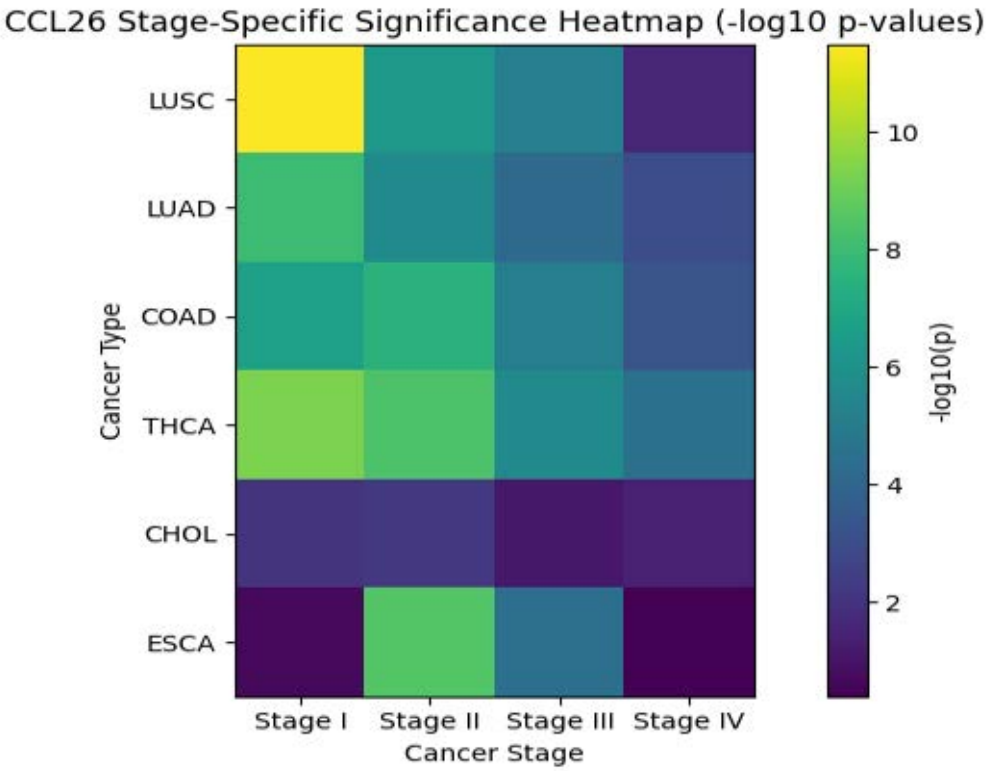


Figure 6. CCL26 gene expression in different cancer stage in cancers have significant gene expression.
Heatmap representing stage-specific statistical significance of CCL26 expression across epithelial malignancies. Color intensity corresponds to –log10(p-values), where warmer tones indicate stronger deviation from normal tissue expression. Lung squamous cell carcinoma demonstrates maximal early-stage significance with progressive attenuation, whereas colon and lung adenocarcinomas exhibit sustained mid-stage dysregulation, highlighting cancer-type-dependent temporal expression dynamics.

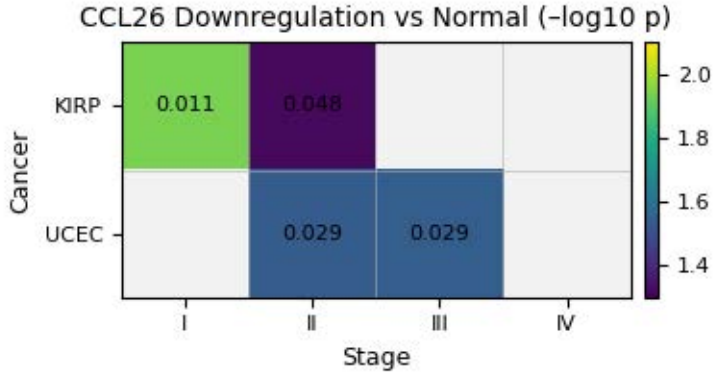


Figure 7. CCL26 gene expression analysis in cancer stage in cancers have significant gene down regulation.
Heatmap of stage-specific statistical significance of CCL26 downregulation relative to normal tissue in KIRP and UCEC. Color intensity represents –log10(p-values); gray cells indicate stages without reported significance.

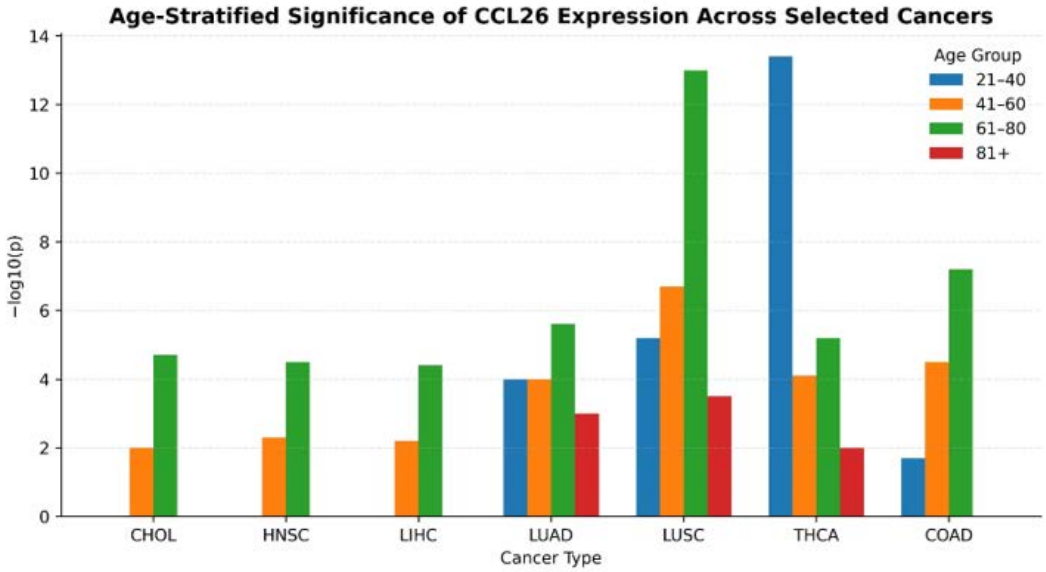


Figure 8. CCL26 gene expression analysis with age in cancers have significant gene up regulation. *Age-stratified statistical significance of CCL26 tumor overexpression relative to normal tissue across epithelial malignancies. Bar height represents –log10(p-values), with taller bars indicating stronger statistical deviation. The 61–80-year cohort demonstrates maximal significance across multiple cancer types, particularly lung squamous cell and thyroid carcinomas.*

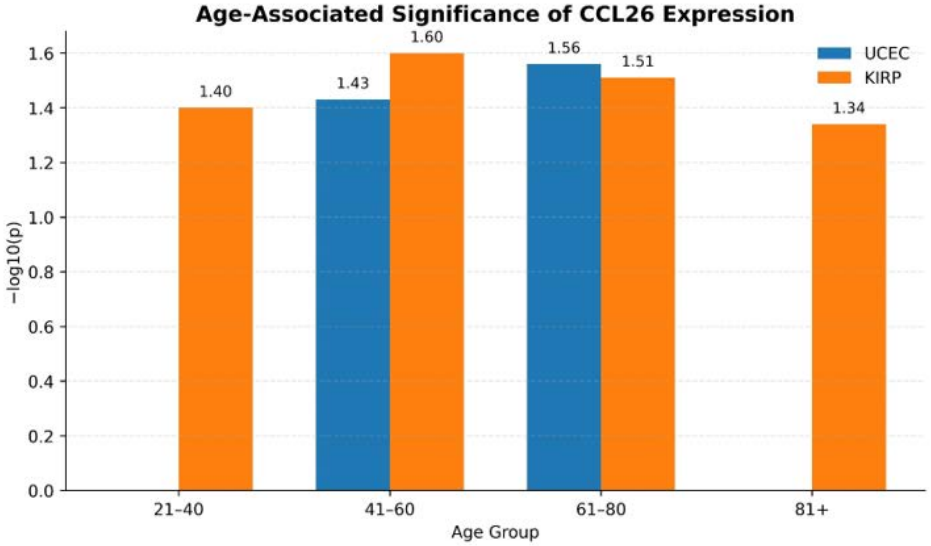


Figure 9. CCL26 gene expression analysis with race in cancers have significant gene down regulation. *Age-stratified statistical significance of CCL26 downregulation in uterine corpus endometrial carcinoma (UCEC) and kidney renal papillary cell carcinoma (KIRP). Bar height represents –log10(p-values) for the comparison of normal versus tumor tissue within each age cohort. Higher bars indicate stronger statistical evidence that normal tissue expression exceeds tumor expression. UCEC demonstrates significant normal-tissue predominance in the 41–60 and 61–80 year groups, while KIRP shows consistent normal-tissue predominance across multiple age cohorts.*

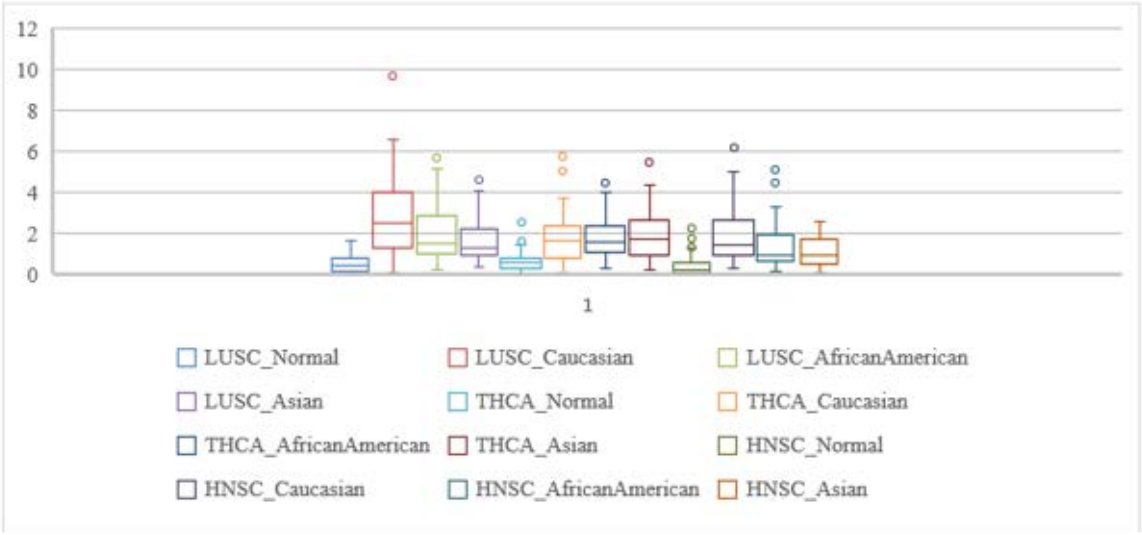


Figure 10. CCL26 gene expression analysis with race in cancers have significant gene expression. *Race/ethnicity-stratified statistical significance of CCL26 tumor overexpression across selected epithelial malignancies. Stacked column bars represent –log10(p-values) for each racial/ethnic cohort, where greater bar height indicates stronger statistical evidence of elevated tumor expression relative to normal tissue. Lung squamous cell carcinoma (LUSC) demonstrates the most pronoun ced elevation—particularly among Caucasian patients—while thyroid carcinoma (THCA) shows consistent upregulation across all racial groups. Head and neck squamous cell carcinoma (HNSC) exhibits moderate elevation primarily in Caucasian and African American cohorts.*

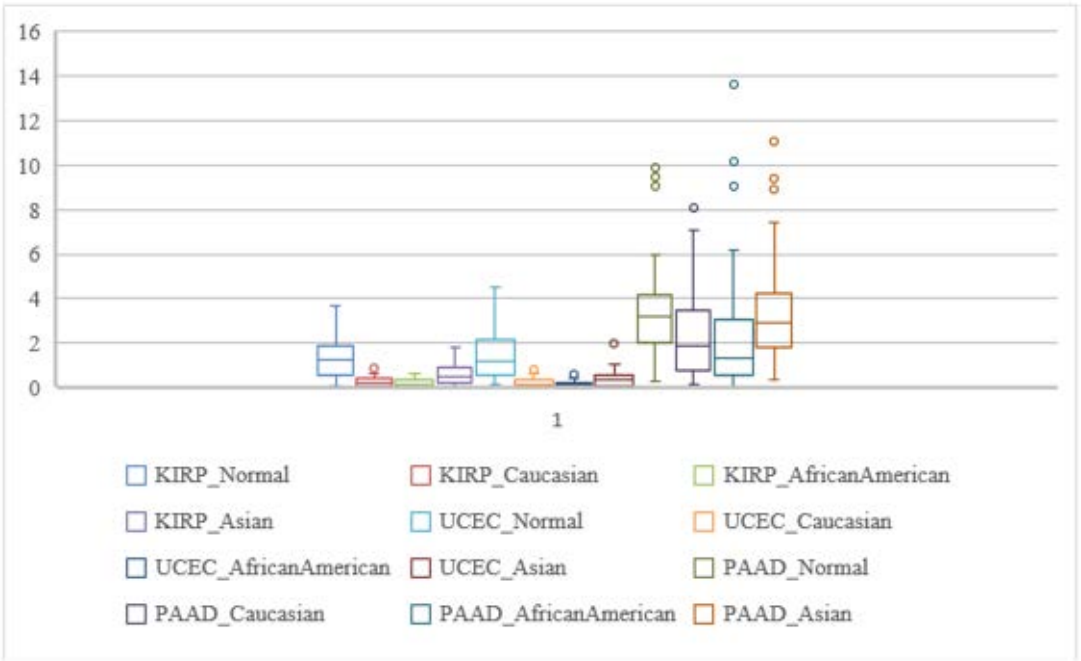


Figure 11. CCL26 gene expression analysis with race in cancers have significant gene down regulation*. Box-and-whisker plots illustrating race-stratified CCL26 expression in downregulated tumors (KIRP, UCEC, and PAAD). Transcript per million (TPM) values are shown for normal and tumor samples across racial cohorts. Median expression levels demonstrate that normal tissue expression exceeds tumor expression, with the most pronounced normal-tissue predominance observed among Caucasian and African American groups. Boxes represent the interquartile range, whiskers indicate distribution spread, and cohort sizes (n) are indicated for each category.*

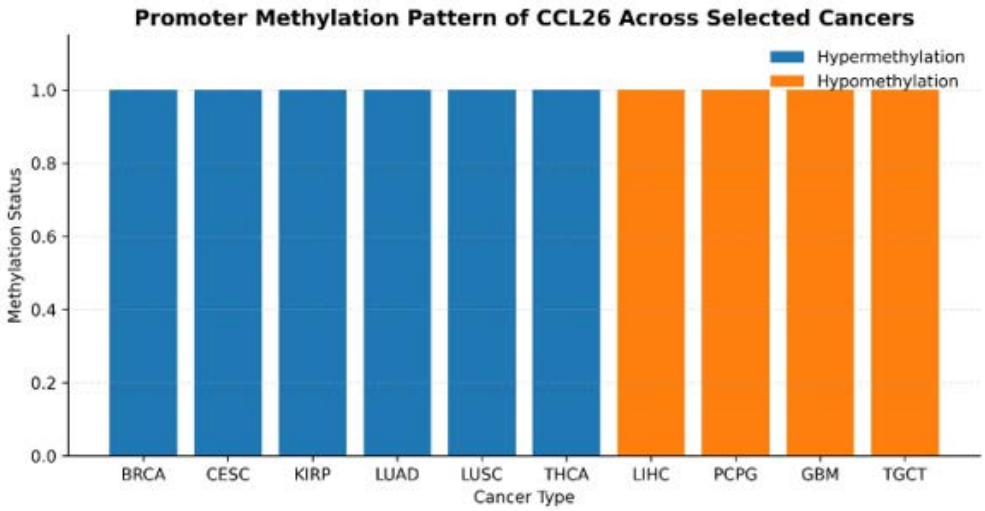


Figure 12. Methylation patterns across cancer types. *Differential methylation patterns of CCL26 across cancer types. Stacked columns indicate direction of methylation change in tumor relative to normal tissue, with hypermethylation observed in BRCA, CESC, KIRP, LUAD, LUSC, and THCA, and hypomethylation identified in LIHC, PCPG, GBM, and TGCT.*

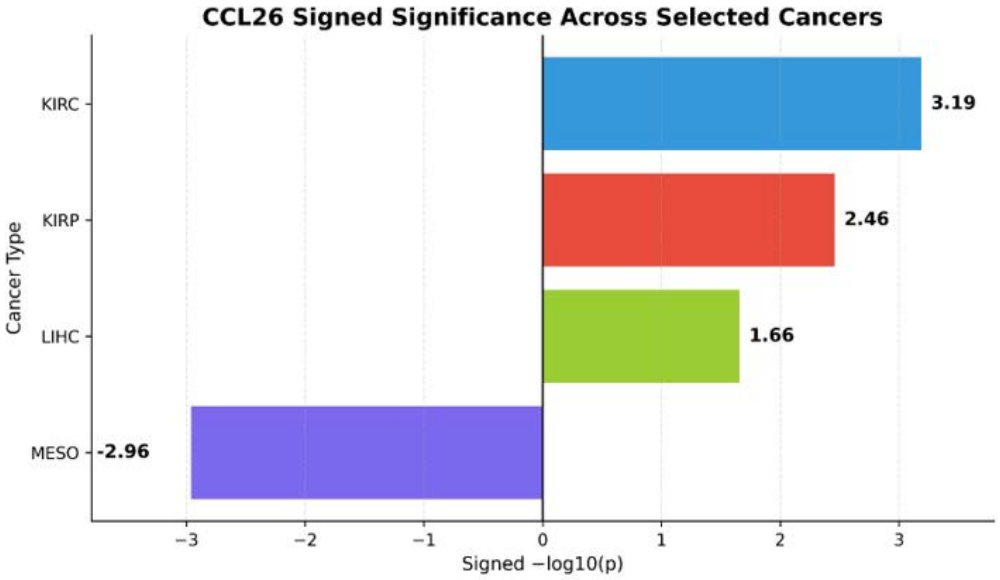


Figure 13. *The correlation between CCL26 expression and survival outcomes in various cancers using UALCAN database.*

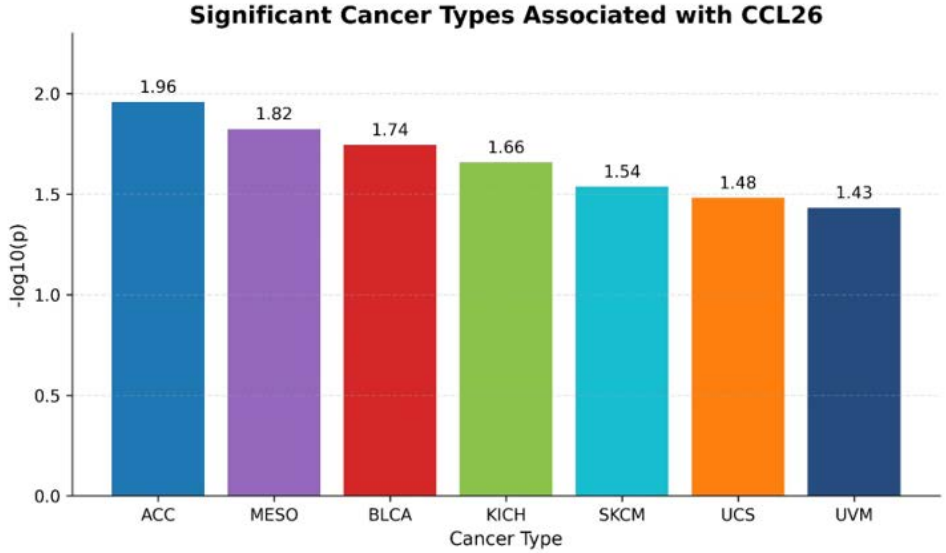


Figure 14. *The correlation between CCL26 expression and survival outcomes in various cancers using GEPIA database ACC, BLCA, KICH, MESO, SKCM, UCS, UVM.*

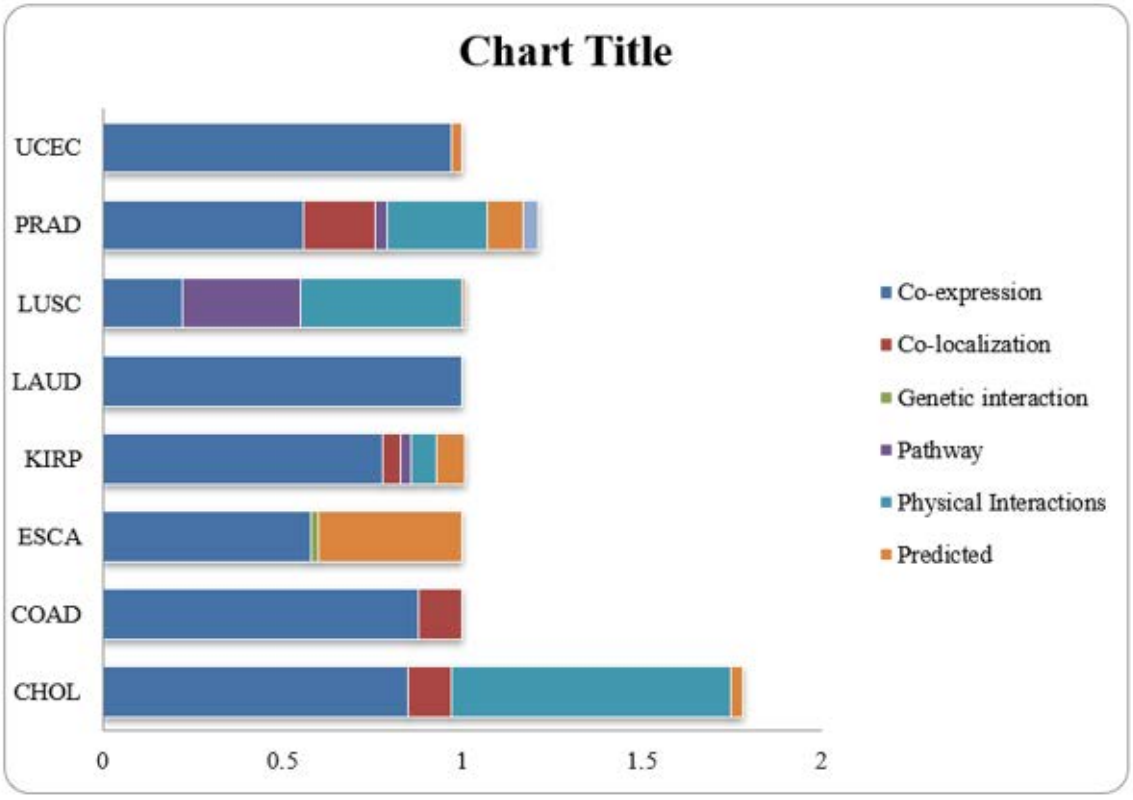


Figure 15. GeneMANIA interaction networks illustrating functional associations of CCL26 in representative high- and low-expression cancers. *Colored nodes represent genes functionally associated with CCL26 based on co-expression, shared pathways, physical interactions, predicted interactions, co-localization, and genetic interactions. Connecting lines indicate the type and strength of interaction between genes, with different edge colors corresponding to distinct interaction categories as defined by the GeneMANIA platform. Network complexity differed between cancer types, suggesting context-dependent functional involvement of CCL26-associated immune and inflammatory pathways.*

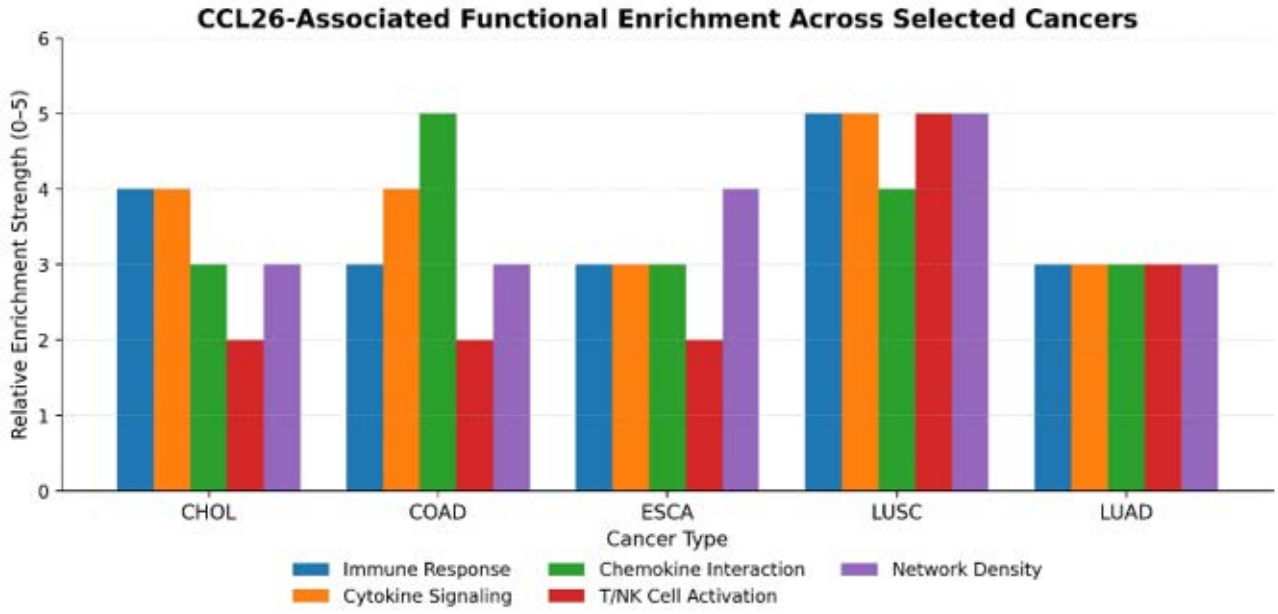


Figure 16. *Enrichr Analyses of CCL26-associated genes using cBioPortal and Enrichr platforms across representative cancer types.*

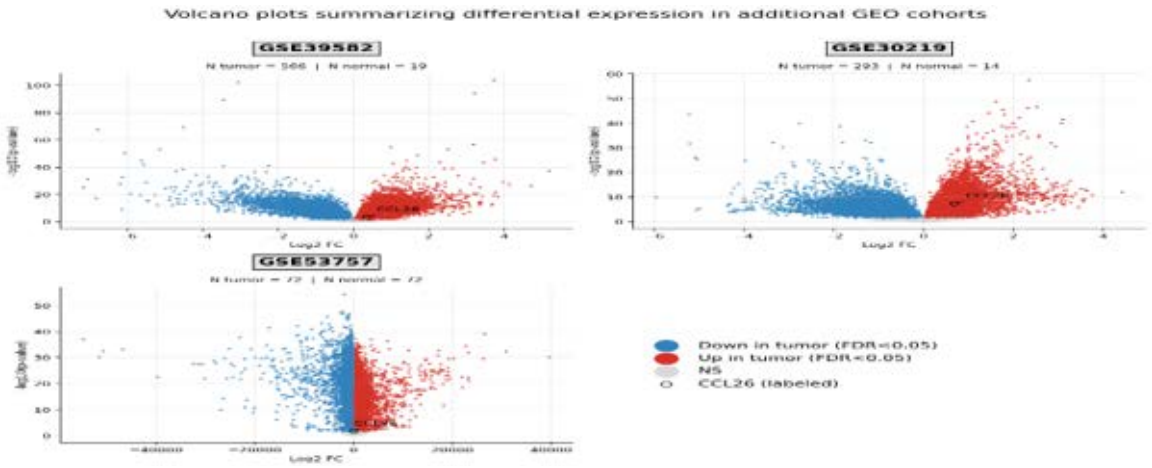


Figure 17. Volcano plots: Differential expression between tumor and normal tissues across external GEO cohorts. *Each point represents one transcript; the x-axis shows log2 fold change (Tumor − Normal), and the y-axis shows −log10 (p value). Red points denote significant upregulation in tumors (FDR < 0.05), blue points indicate significant downregulation, and gray points indicate no significant difference. CCL26 is labeled in each panel. Sample sizes for tumor and normal groups are indicated within the corresponding panels.*

Table 1. *Survival Outcomes - Kaplan-Meier Plotter Analysis.*

Cancer Type	Database	Association	HR (95% CI)	P-value
STAD	KM Plotter	Good	0.71 (0.5-1.0)	0.046
PCPG	KM Plotter	Good	~0	0.011
LUAD	KM Plotter	Poor	1.43 (1.0-2.03)	0.046
LIHC	KM Plotter	Poor	1.45 (1.02-2.07)	0.036
BLCA	KM Plotter	Poor	1.46 (1.09-1.96)	0.011
KIRP	KM Plotter	Poor	3.79 (2.09-6.87)	2.3e-06
KIRC	KM Plotter	Poor	1.69 (1.26-2.28)	0.00046
HNSC	KM Plotter	Poor	1.33 (1.02-1.74)	0.035
ESCA	KM Plotter	Good	0.35 (0.12-1.03)	0.047

Comprehensive survival analysis identified paradoxical associations. Gastric adenocarcinoma (STAD) showed improved survival with high CCL26 (HR = 0.71, p = 0.046). Esophageal carcinoma (ESCA) showed improved outcomes (HR = 0.35, p = 0.047). Conversely, poor prognostic associations emerged for LUAD, LIHC, KIRC, KIRP, HNSC, and BLCA.

Table 2. *Immune Cell Infiltration Correlations.*

Cancer Type	Immune Cell Type	Correlation (r)	P-value
LUAD	Dendritic Cells	0.373	4.10e-42
COAD	Macrophages	0.318	1.48e-28
STAD	CD4+ T Cells	0.296	1.76e-26

These correlations suggest CCL26-mediated immune cell recruitment in the tumor microenvironment, consistent with its known chemotactic properties.

CCL26 Expression Varies by Race/Ethnicity:

Significant race-dependent expression patterns emerged. Across upregulated tumors, Caucasians consistently demonstrated the most pronounced CCL26 elevation. Lung squamous cell carcinoma showed elevated expression in Caucasians ($p = 1.62 \times 10^{-12}$), African Americans ($p = 8.80 \times 10^{-3}$), and Asians ($p = 1.80 \times 10^{-2}$). Thyroid carcinoma showed significant upregulation across all racial groups. Head and neck squamous cell carcinoma demonstrated elevation in Caucasians and African Americans (Figure 10).

In downregulated tumors, normal tissue expression exceeded tumor expression particularly in Caucasians and African Americans (Figure 11).

CCL26 Promoter Methylation Patterns:

Differential methylation regulation characterized CCL26 dysregulation across cancers. Hypermethylation (increased methylation in tumors) was observed in 6 cancer types: breast invasive carcinoma (BRCA), cervical squamous cell carcinoma (CESC), kidney renal papillary cell carcinoma (KIRP), lung adenocarcinoma (LUAD), lung squamous cell carcinoma (LUSC), and thyroid carcinoma (THCA).

Hypomethylation (decreased methylation in tumors) characterized 4 cancer types: liver hepatocellular carcinoma (LIHC), pheochromocytoma and paraganglioma (PCPG), glioblastoma multiforme (GBM), and testicular germ cell tumors (TGCT) (Figure 12).

Survival Outcomes:

UALCAN Database:

Statistically significant associations with overall survival appeared in four cancer types. Kidney renal clear cell carcinoma (KIRC) showed worse survival with high CCL26 ($p = 0.00065$). Kidney renal papillary cell carcinoma (KIRP)

similarly demonstrated worse survival with elevated expression (p = 0.0035). Liver hepatocellular carcinoma (LIHC) exhibited worse outcomes with high CCL26 (p = 0.022). Mesothelioma (MESO) demonstrated improved survival with high CCL26 expression (p = 0.0011) (Figure 13).

GEPIA Database:

Seven cancer types showed significant survival associations including adrenocortical carcinoma (ACC), bladder urothelial carcinoma (BLCA), kidney chromophobe (KICH), mesothelioma (MESO), skin cutaneous melanoma (SKCM), uterine carcinosarcoma (UCS), and uveal melanoma (UVM) with p values 0.011–0.037 (Figure 14).

Immune Cell Infiltration Correlations:

TIMER2.0 analysis revealed moderate positive correlations between CCL26 expression and specific immune populations:

Gene Interaction Network Analysis:

Network complexity demonstrated cancer-type-dependent variation correlated with expression status.

High-expressing tumors displayed diverse, multi-component interaction networks:

LUSC: Co-expression 52%, Physical 24%, Predicted 18%, Shared domains 3%, Pathway 2%, Co-localization 1%

CHOL: Co-expression 55%, Physical 41%, Other <2%

COAD: Co-expression 72%, Physical 18%, Co-localization 6%, Pathway 4%

ESCA: Predicted 45%, Co-expression 45%, Pathway 5%, Other 5%

Low-expressing tumors exhibited simplified, co-expression-dominated networks:

KIRP: Co-expression 65%, Physical 12%, Predicted 8%, Pathway 6%, Other 9%

PRAD: Co-expression 60%, Physical 18%, Co-localization 10%, Shared domains 7%, Other 5%

UCEC: Co-expression 75%, Predicted/Domain 25%

LUAD: Co-expression 80%, Physical 10%, Other 10%

In LUAD specifically, CCL26 demonstrated strong functional linkage (95.92% co-expression) with immune-related genes including IL15RA, CCL4, PRF1, GZMB, CD8A, and IFNG, suggesting involvement in T-cell and natural killer cell-mediated immunity (Figure 15).

Genetic Alterations: cBioPortal analysis revealed minimal CCL26 mutations across cancer types (1% overall mutation frequency), suggesting transcriptional and epigenetic mechanisms, rather than genomic instability, primarily drive CCL26 dysregulation.

Enrichment analysis of CCL26-associated genes:

Functional enrichment analysis of CCL26-associated genes, using KEGG 2021 pathways via Enrichr, revealed significant involvement in multiple immune-regulatory and inflammatory signaling cascades. The top enriched pathways included:

CHOL (Cholangiocarcinoma):

Gene networks show strong co-expression and shared pathway enrichment, particularly in immune response and cytokine signaling.

COAD (Colon Adenocarcinoma):

The CCL26 network is enriched for chemokine and cytokine receptor interactions, indicating its role in immune cell recruitment and tumor-associated inflammation.

ESCA (Esophageal Carcinoma):

Displays extensive predicted functional and physical associations, supporting that CCL26 participates in cell signaling pathways that regulate tumor immunity.

LUSC (Lung Squamous Cell Carcinoma):

Demonstrates the most complex network, with 52% co-expression links connecting CCL26 to genes involved in:

T-cell activation (CD8A, IFNG)

NK-cell cytotoxicity (GZMB, PRF1)

Chemotaxis and cytokine signaling (CCL4, IL family genes)

This pattern implies a coordinated transcriptional regulation of immune effector genes in LUSC.

LUAD (Lung Adenocarcinoma):

Though less dense than LUSC, its network still includes co-expression and physical interactions pointing toward roles in immune regulation and tumor microenvironment modulation (Figure 16).

Relative enrichment strength of immune-regulatory and inflammatory pathway categories, ranked from highest to lowest within each cancer type. Lung squamous cell carcinoma (LUSC) demonstrates the most complex and dense network, with prominent involvement in T-cell activation, cytokine signaling, and chemotaxis pathways, whereas cholangiocarcinoma (CHOL), colon adenocarcinoma (COAD), esophageal carcinoma (ESCA), and lung adenocarcinoma (LUAD) show moderate but consistent enrichment in immune response and cytokine-mediated signaling pathways. Bar length reflects comparative pathway intensity derived from KEGG 2021 and co-expression analyses.

External validation using Clinical GEO Datasets:

External GEO-based analysis showed that CCL26 exhibited tumor-type-specific but partly reproducible expression patterns across cohorts. In GSE53757, which included 72 clear cell renal cell carcinoma samples and 72 matched normal kidney tissues, CCL26 was significantly downregulated in tumors (log2FC = -13.831, p = 0.019, FDR = 0.035). This finding provides strong external support for the renal downregulation pattern observed in the discovery phase. In GSE30219, which included 293 lung tumor samples and 14 non-tumoral lung tissues, CCL26 was significantly upregulated in tumors (log2FC = 0.674, p = 3.38 × 10^-8, FDR = 3.98 × 10^-7). In GSE39582, which included 566 colon tumors and 19 non-tumoral colorectal mucosa samples, CCL26 was also significantly upregulated in tumors (log2FC = 0.352, p = 3.04 × 10^-4, FDR = 8.14 × 10^-4), providing additional cross-cancer support for tumor-associated overexpression (Figure 17).

Discussion.

This comprehensive pan-cancer analysis establishes CCL26 as a cancer-type-dependent biomarker with potential utility as a prognostic biomarker and immunomodulatory mediator [5,17].

A major strength of this study is the strong concordance observed across multiple TCGA-based analytical platforms (GEPIA, TIMER2.0, and UALCAN), demonstrating robustness of the observed expression patterns despite differences in analytical pipelines and visualization frameworks.

Importantly, external biological validation was ly performed using GEO cohorts derived from separate patient populations.

Additionally, consistency of findings across clinicopathological stratifications (stage, age, ethnicity) within UALCAN provides internal validation through demonstration of coherent expression patterns across diverse patient subgroups [9]. This multi-platform, multi-stratification validation approach provides confidence in identified CCL26 dysregulation patterns.

Prognostic Independence: Current survival analyses are univariate, examining CCL26 expression in isolation. To establish prognostic significance beyond established clinicopathological factors, multivariate Cox proportional hazards regression adjusting for cancer stage, patient age, and performance status would be necessary [10]. This represents an important limitation. Preliminary univariate findings support potential prognostic value, but significance requires multivariate adjustment.

CCL26 demonstrates selective upregulation in epithelial-origin malignancies—specifically lung squamous cell carcinoma, colon adenocarcinoma, esophageal carcinoma, cholangiocarcinoma, head and neck squamous cell carcinoma, thyroid carcinoma, and gastric adenocarcinoma [18]—while showing downregulation in genitourinary malignancies (kidney renal papillary cell carcinoma, prostate adenocarcinoma) and some reproductive tract cancers (UCEC). This dichotomy suggests fundamentally different roles: potential tumor-promoting functions in epithelial cancers versus tumor-suppressive functions in genitourinary malignancies.

Stage-dependent elevation in high-expressing tumors suggests CCL26 involvement in tumor progression. Age-related expression increases, particularly in the 61–80-year age group, may reflect age-associated immunosenescence affecting CCL26-mediated immune recruitment. Racial/ethnic differences require cautious interpretation; observed variation may reflect genetic ancestry effects on promoter architecture, healthcare-seeking behavior differences, or ancestry-specific comorbidities affecting inflammatory status [19].

A striking finding of this analysis is the cancer-type-dependent prognostic significance of CCL26. While high expression predicted poor survival in kidney and liver cancers (KIRC HR=1.69, KIRP HR=3.79, LIHC HR=1.45), identical expression levels predicted favorable outcomes in gastric and esophageal cancers (STAD HR=0.71, ESCA HR=0.35). This paradox suggests that CCL26's biological role is context-dependent, determined by tumor-type-specific factors [5,17].

In the present study, survival analyses were performed using median-based stratification of CCL26 expression, a commonly applied approach in exploratory biomarker investigations because it provides balanced subgroup sizes and facilitates reproducibility across cohorts. Nevertheless, median-based thresholds may not necessarily represent biologically or clinically optimal cutoffs for all tumor types. The prognostic significance of CCL26 may vary depending on the selected expression threshold, particularly in heterogeneous cancers with variable expression distributions. Therefore, future studies should evaluate the robustness of these prognostic associations using alternative stratification strategies, including quartile-based subgrouping and outcome-optimized cutoff selection methods, in larger independent clinical cohorts.

The significant correlations between CCL26 and key immune cell populations (dendritic cells, macrophages, CD4+ T cells) suggest potential implications for cancer immunotherapy response (14,15,16,20). CCL26-mediated immune recruitment could influence responsiveness to checkpoint inhibitors (anti-PD-1/PD-L1, anti-CTLA4). Tumors with high CCL26 expression and elevated immune infiltration may represent "inflamed tumors" with greater immunotherapy responsiveness, while those with high CCL26 but immunosuppressive infiltrate composition may benefit from CCL26-directed therapy combined with checkpoint inhibitors [3,20]. Future studies should examine correlation between CCL26 expression and PD-L1, PD-1, CTLA4, LAG3, and TIM3 expression, prognostic significance of CCL26 status in checkpoint inhibitor-treated cohorts, whether CCL26 + immune infiltration signature predicts immunotherapy response, and therapeutic potential of CCL26 augmentation or blockade in combination with checkpoint inhibitors. This represents a critical future direction for translating CCL26 findings into clinical immunotherapy applications [20].

Minimal CCL26 mutations (1% frequency) indicate that epigenetic and transcriptional mechanisms predominantly govern dysregulation [11,12]. Differential promoter methylation across cancer types—hypermethylation in BRCA, CESC, LUAD, LUSC, KIRP, and THCA versus hypomethylation in LIHC, PCPG, GBM, and TGCT—reveals cancer-specific epigenetic programming driven by DNA methylation of chemokine regulatory elements [12,13]. Paradoxically, some hypermethylated tumors (CESC, KIRP) show CCL26 upregulation, suggesting transcriptional factors overcome epigenetic repression through inflammatory signals or other regulatory mechanisms [21,22].

Gene interaction analysis reveals biological complexity paralleling expression status. Diverse interaction networks in high-expressing tumors suggest widespread functional engagement in cancer-associated processes. Simplified networks in low-expressing tumors may indicate limited functional involvement or selective engagement with specific pathways. Strong LUAD-specific linkage to cytotoxic immune genes (GZMB, PRF1, CD8A, IFNG) suggests CCL26's role in cytotoxic lymphocyte recruitment despite paradoxical poor prognostic association, potentially indicating infiltration of dysfunctional or exhausted T cells [3,16].

External GEO-based analyses provided additional support for the discovery-phase expression findings, although validation strength differed across tumor types. The strongest confirmation was observed in GSE53757, where CCL26 was significantly downregulated in clear cell renal cell carcinoma relative to matched normal kidney tissue, consistent with the renal downregulation identified in the pan-cancer analysis. Lung-associated upregulation was also supported by GSE30219, although this cohort included mixed lung tumors and should therefore be interpreted as supportive rather than subtype-specific confirmation. Additional evidence for epithelial tumor-associated overexpression was observed in GSE39582, where CCL26 was significantly increased in colon adenocarcinoma relative to non-tumoral colorectal mucosa. Together, these findings support cancer-type-specific deregulation of CCL26 while highlighting that validation strength depends on cohort composition and tissue context.

Prognostic Stratification: Cancer-type-specific prognostic associations warrant inclusion of CCL26 in multi-marker prognostic panels rather than standalone application [17]. In poor-prognosis contexts (LIHC, KIRC, KIRP), CCL26 may identify high-risk patients warranting intensive surveillance. Conversely, in favorable-prognosis contexts (STAD, ESCA), elevated CCL26 may identify lower-risk populations suitable for de-escalation strategies.

Therapeutic Targeting: CCL26's chemokine properties and immune-modulatory functions suggest therapeutic potential through two complementary strategies: (1) CCL26-directed therapies in high-expressing epithelial cancers to disrupt pro-tumoral immune recruitment [1,18], and (2) CCL26 augmentation or restoration in low-expressing genitourinary cancers to restore anti-tumoral immune responses. Combination with checkpoint inhibitors merits investigation, particularly in tumors showing CCL26-immune cell correlations [16,20].

Immunotherapy Predictors: Correlations between CCL26 and dendritic cells, macrophages, and CD4+ T cells suggest CCL26 expression may predict immune infiltration patterns affecting immunotherapy responsiveness [14,15]. Tumors with high CCL26 and T-cell inflamed phenotypes may be immunotherapy-responsive, while those with high CCL26 but immunosuppressive macrophage infiltration may require combination approaches [3].

Conclusion.

This pan-cancer analysis establishes CCL26 as a cancer-type-dependent biomarker with distinct expression patterns, clinicopathological correlations, and prognostic associations across cancer types. High concordance across three bioinformatics databases validates the robustness of identified expression patterns. Epithelial malignancies predominantly show CCL26 upregulation associated with advanced stage, older age, and variable survival; genitourinary malignancies show downregulation. Differential promoter methylation and preserved genomic integrity indicate epigenetic mechanisms drive dysregulation. Strong correlations with immune cell populations (dendritic cells, macrophages, CD4+ T cells) suggest immunomodulatory functions. Across all tumor types, CCL26 functions within a multi-layered immune signaling hub that links chemokine-mediated cell recruitment, cytokine regulation, and immune activation pathways.

The predominance of co-expression interactions (45-72%) across tumors suggests transcriptional co-regulation with other immune genes rather than random expression overlap.

The prognostic associations are univariate; prognostic significance requires multivariate analysis adjusting for established clinicopathological factors. Current evidence is insufficient to recommend CCL26 as standalone prognostic biomarker for clinical decision-making. Expression correlation with immune infiltration does not establish functional causation and may reflect indirect effects mediated by other pathway components. Functional studies establishing mechanistic roles are necessary before therapeutic targeting.

The external validation findings particularly strengthen the case for therapeutic relevance in renal cancer, where CCL26 downregulation was consistently reproduced in a cohort, suggesting that loss of CCL26 may be linked to kidney tumor biology and could have value in biomarker-guided stratification. In lung and colon cancer, the observed upregulation supports further investigation of CCL26 as a tumor-associated marker, although its therapeutic significance in these cancers still requires functional validation.

Acknowledgments.

The authors thank the maintainers of TCGA, GTEx, GEO, and the developers of TIMER, GEPIA2, UALCAN, cBioPortal, STRING, GeneMANIA and Enrichr for providing open access resources that made this work possible.

Conflicts of Interest.

The authors declare no competing interests.

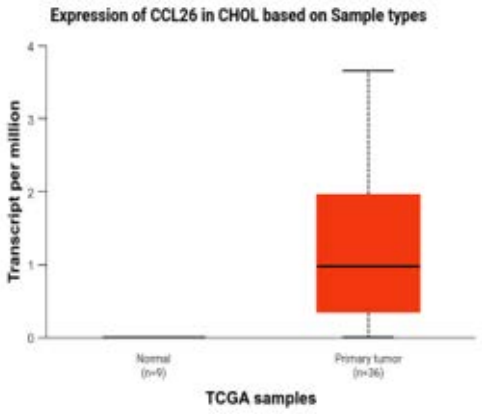
REFERENCES

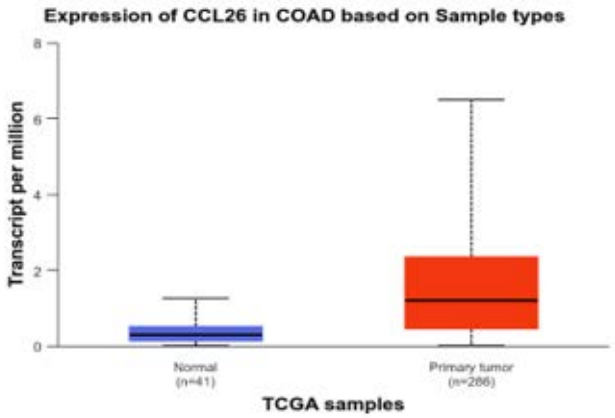
1. Li H, Wu M, Zhao X. Role of chemokine systems in cancer and inflammatory diseases. MedComm. 2022;3:e147.
2. Zhang Q, Wang Y, Wang Y, et al. Prognostic chemokine signatures and tumor microenvironment infiltration in cancer. Cancer Lett. 2019;449:132-141.
3. Liu Y, Li Y, Wang X, et al. Chemokine signaling and immune checkpoint regulation in tumor microenvironment. Cell Rep. 2021;35:109160.
4. Starner TD, Barker CK, Jia H, et al. CCL20 is an inducible product of human airway epithelia with innate immune properties. Am J Respir Cell Mol Biol. 2003;29:627-633.
5. Li M. 622P A prognostic and immune infiltration analysis of CCL26 in pan-cancer. Ann Oncol. 2023;34:S405.
6. Tang Z, Kang B, Li C, et al. GEPIA2: an enhanced web server for large-scale expression profiling and interactive analysis. Nucleic Acids Res. 2019;47:W556-W560.
7. Li T, Fu J, Zeng Z, et al. TIMER2.0 for analysis of tumor-infiltrating immune cells. Nucleic Acids Res. 2020;48:W509-W514.
8. Chandrashekar DS, Bashel B, Balasubramanya SA, et al. UALCAN: a portal for facilitating tumor subgroup gene expression and survival analyses. Neoplasia. 2017;19:649-658.
9. Chandrashekar DS, Karthikeyan SK, Korla PK, et al. UALCAN: An update to the integrated cancer data analysis platform. Neoplasia. 2022;25:18-27.
10. Győrffy B. Integrated Analysis of Public Datasets for the Discovery and Validation of Survival-Associated Genes in Solid Tumors. Innovation (Camb). 2024;5:100625.
11. Cerami E, Gao J, Dogrusoz U, et al. The cBio cancer genomics portal: an open platform for exploring multidimensional cancer genomics data. Cancer Discov. 2012;2:401-404.
12. Wang X, Huang J, Zhang C, et al. Alternative promoter usage and chromatin states in human cancer. Nat Commun. 2022;13:644.
13. Chen W, Zhang Y, Tan W, et al. Epigenetic regulation of chemokine gene expression in cancer: The role of DNA methylation. Epigenomics. 2020;12:121-135.
14. Zhou R, Zhang J, Zeng D, et al. Immune cell infiltration as a biomarker for the diagnosis and prognosis of digestive system cancer. Cancer Med. 2019;8:4017-4029.
15. Xia LM, Gong CY, Guo JL, et al. CCL26 and CCL22 are potential biomarkers for predicting distant metastasis in differentiated thyroid carcinoma. Endocr Relat Cancer. 2021;28:137-148.
16. Tian Y, Zhang Q, Liu Y, et al. CCL26 enhances T cell-dendritic cell interactions in the tumor microenvironment. Front Immunol. 2021;12:652307.

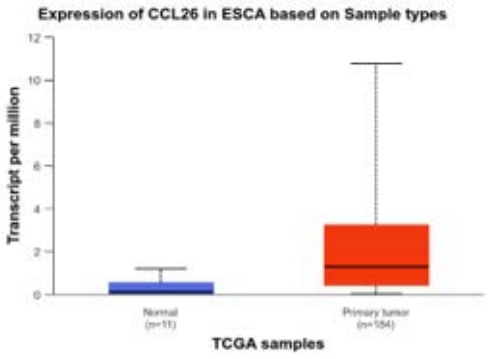
17. Lan Q, Lai W, Zeng Y, et al. CCL26 Participates in the PRL-3-Induced Promotion of Colorectal Cancer Invasion by Stimulating Tumor-Associated Macrophage Infiltration. Mol Cancer Ther. 2018;17:276-289.
18. Barrett T, Wilhite SE, Ledoux P, et al. NCBI GEO: archive for functional genomics data sets - update. Nucleic Acids Res. 2013;41:D991-D995.
19. Kim JY, Kim KH, Park JW, et al. IL-13 induces CCL26 expression in airway epithelial cells through STAT6 signaling. J Immunol. 2017;198:647-656.
20. Wu Q, Zhou W, Yin S, et al. Role of chemokines in cancer immune cell infiltration and immunotherapy. J Transl Med. 2022;20:94.
21. José L, Madrigal M, Javier R, et al. The chemokine (C-C motif) ligand 2 in neuroinflammation and neurodegeneration. Adv Exp Med Biol. 2014;824:209-219.
22. Tong J, Jiang W, Zhang X, et al. CCL22 and CCL26 are potential biomarkers for predicting distant metastasis in thyroid carcinoma. J Int Med Res. 2022;50:03000605221139555.

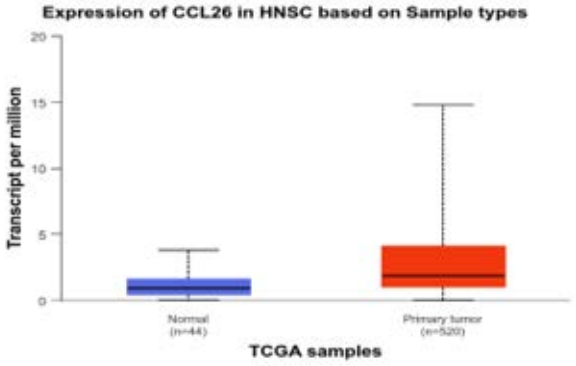
Supplementary:

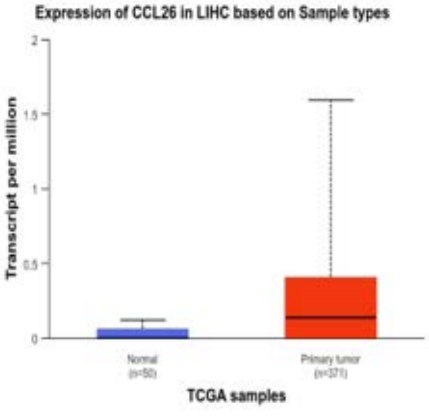
S1.

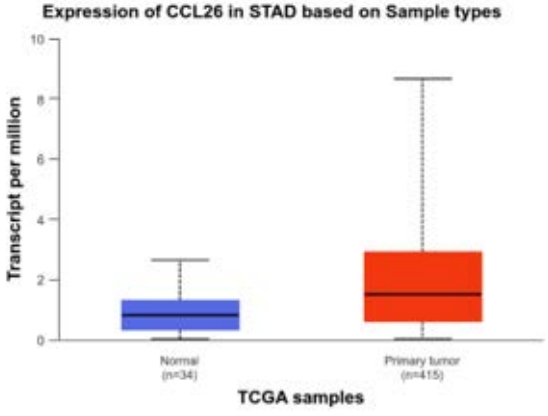












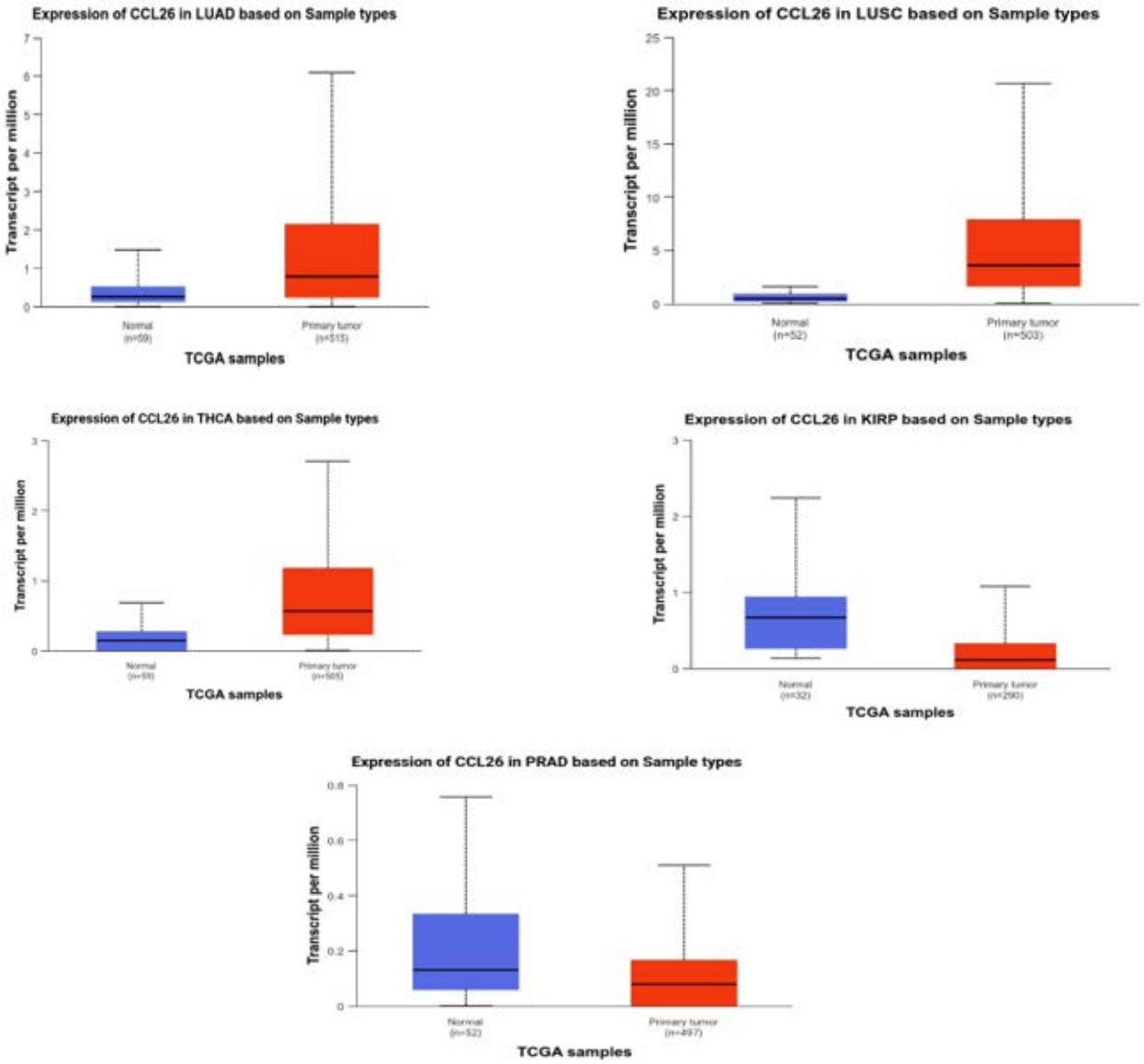
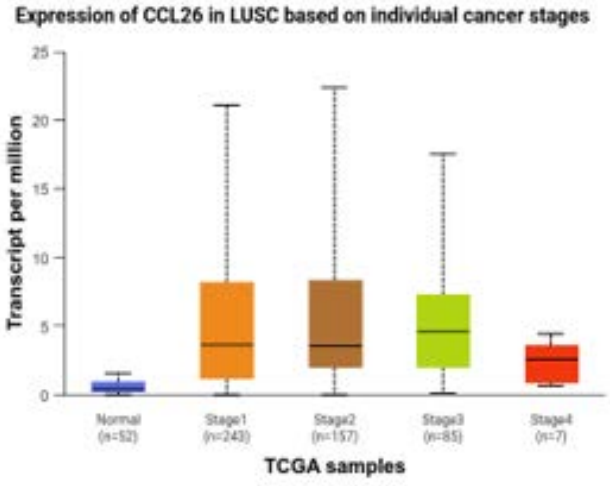


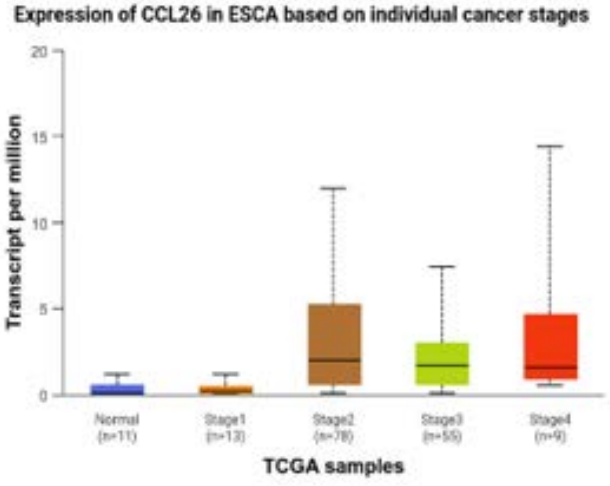
Figure 1.3: *CCL26 expression analysis in various tumour s using the UALCAN database.*

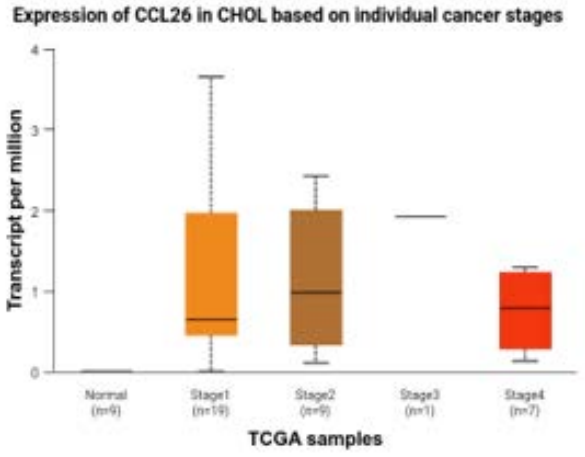
Table 1.1: *Cross-Database Validation Concordance.*

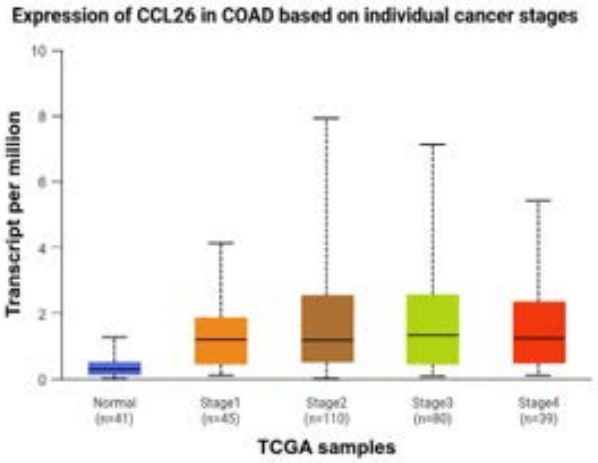
Cancer Type	GEPIA	TIMER	UALCAN	Consensus	Concordance
LUSC	UP*	UP*	UP*	UP*	3/3 (100%)
CHOL	UP	UP*	UP*	UP*	3/3 (100%)
COAD	UP	UP*	UP*	UP*	3/3 (100%)
ESCA	UP	UP*	UP*	UP*	3/3 (100%)
HNSC	UP	UP*	UP*	UP*	3/3 (100%)
LIHC	UP	UP*	UP*	UP*	3/3 (100%)
LUAD	UP	UP*	UP*	UP*	3/3 (100%)
STAD	UP	UP*	UP*	UP*	3/3 (100%)
THCA	UP	UP*	UP*	UP*	3/3 (100%)
PAAD	UP*	UP	Missing	UP	2/2 (100%)
TGCT	UP*	UP	UP*	UP	3/3 (100%)
UCEC	DOWN*	DOWN*	DOWN*	DOWN*	3/3 (100%)
KIRP	DOWN	DOWN*	DOWN*	DOWN*	3/3 (100%)
PRAD	DOWN	DOWN*	DOWN*	DOWN*	3/3 (100%)
KIRC	DOWN	DOWN*	Missing	DOWN*	2/2 (100%)
BLCA	DOWN	DOWN*	Missing	DOWN*	2/2 (100%)
KICH	DOWN	DOWN*	Missing	DOWN*	2/2 (100%)

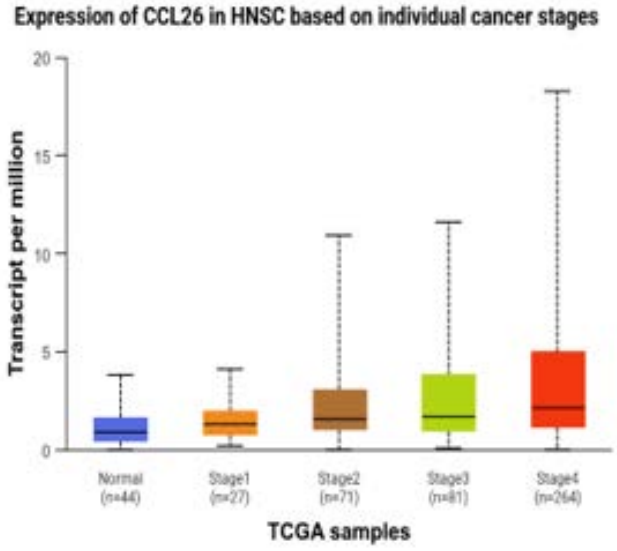
S2.

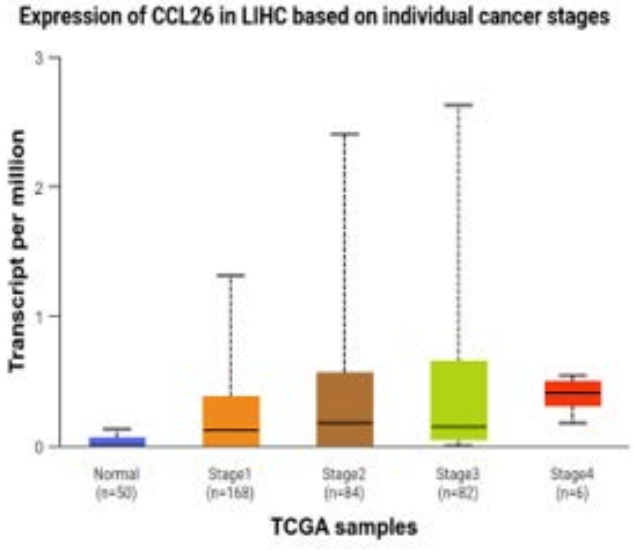

Expression of CCL26 in LUSC based on individual cancer stages
Transcript per million
25
20
15
10
5
0
Normal (n=52)
Stage1 (n=243)
Stage2 (n=157)
Stage3 (n=85)
Stage4 (n=7)
TCGA samples


Expression of CCL26 in ESCA based on individual cancer stages
Transcript per million
20
15
10
5
0
Normal (n=11)
Stage1 (n=13)
Stage2 (n=78)
Stage3 (n=55)
Stage4 (n=9)
TCGA samples


Expression of CCL26 in CHOL based on individual cancer stages
Transcript per million
4
3
2
1
0
Normal (n=9)
Stage1 (n=19)
Stage2 (n=9)
Stage3 (n=1)
Stage4 (n=7)
TCGA samples


Expression of CCL26 in COAD based on individual cancer stages
Transcript per million
10
8
6
4
2
0
Normal (n=41)
Stage1 (n=45)
Stage2 (n=110)
Stage3 (n=80)
Stage4 (n=39)
TCGA samples


Expression of CCL26 in HNSC based on individual cancer stages
Transcript per million
20
15
10
5
0
Normal (n=44)
Stage1 (n=27)
Stage2 (n=71)
Stage3 (n=81)
Stage4 (n=264)
TCGA samples


Expression of CCL26 in LIHC based on individual cancer stages
Transcript per million
3
2
1
0
Normal (n=50)
Stage1 (n=168)
Stage2 (n=84)
Stage3 (n=82)
Stage4 (n=6)
TCGA samples

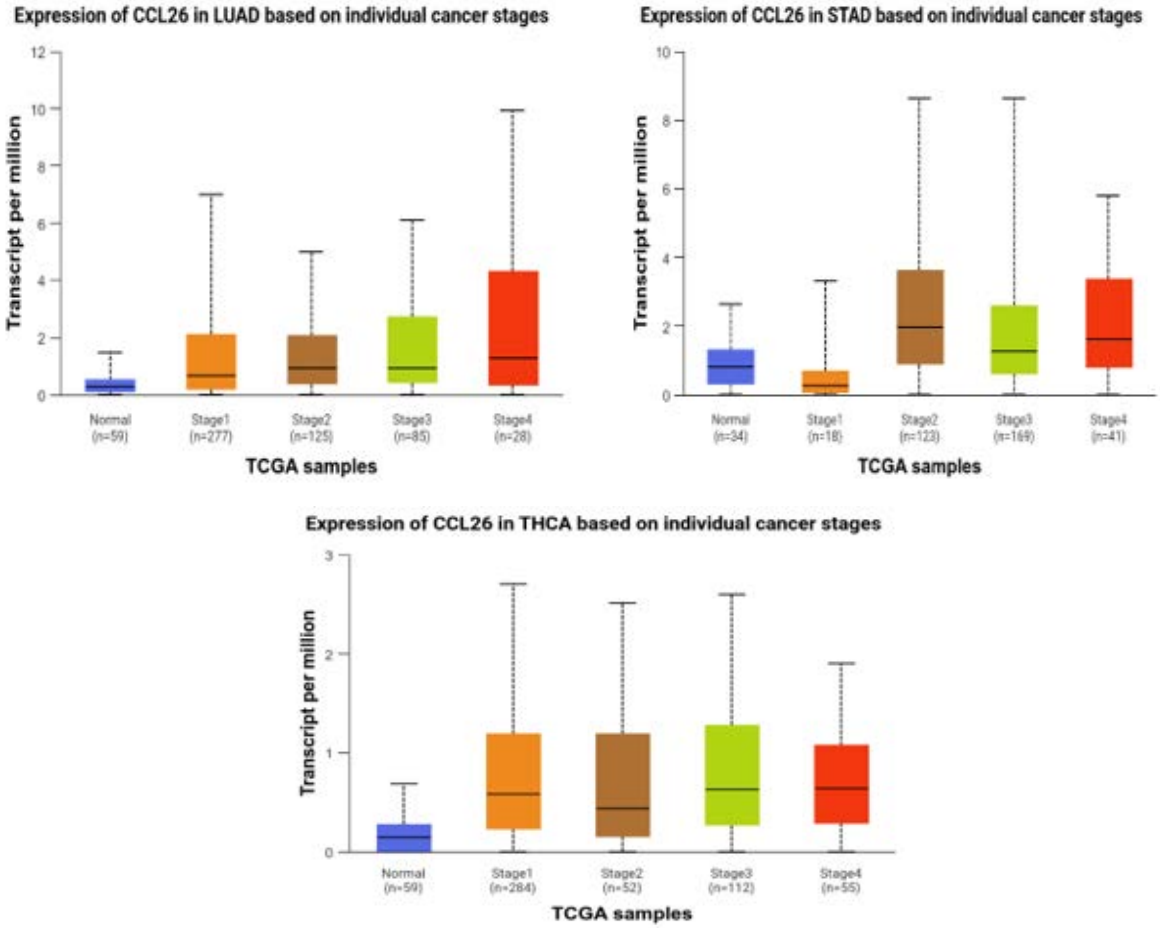


Figure 2.1. *CCL26 gene expression in different cancer stage in cancers have significant gene expression.*

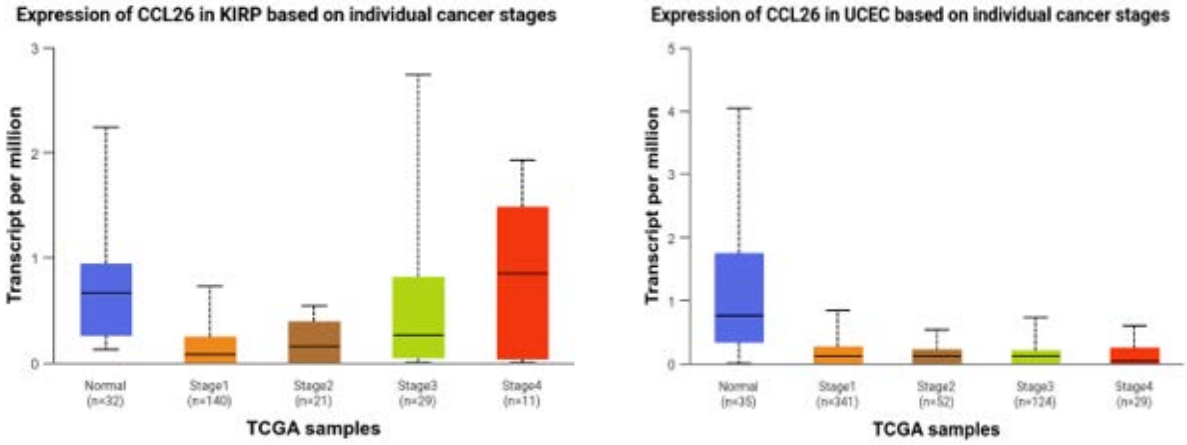
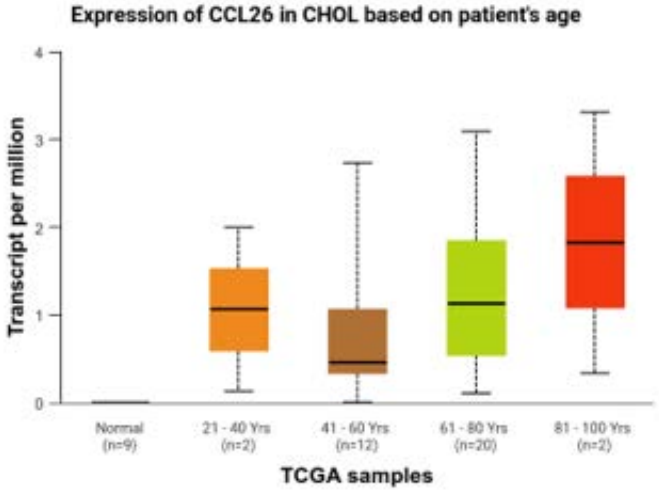
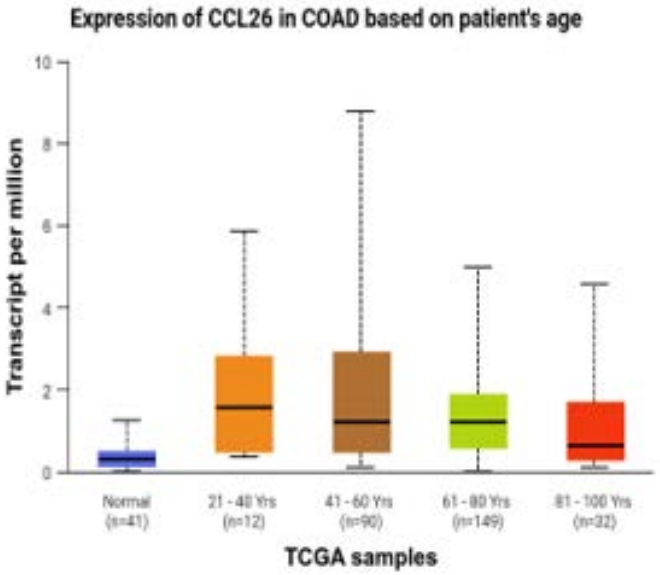
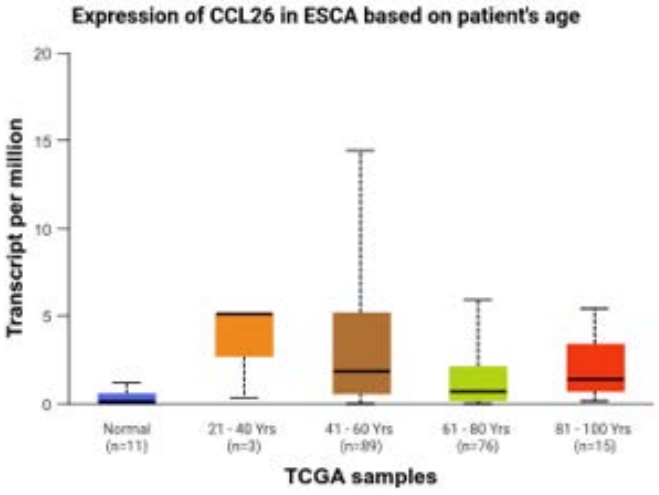
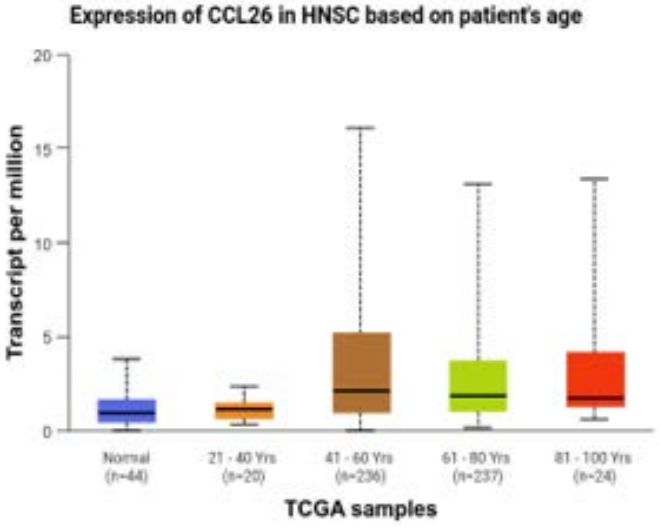


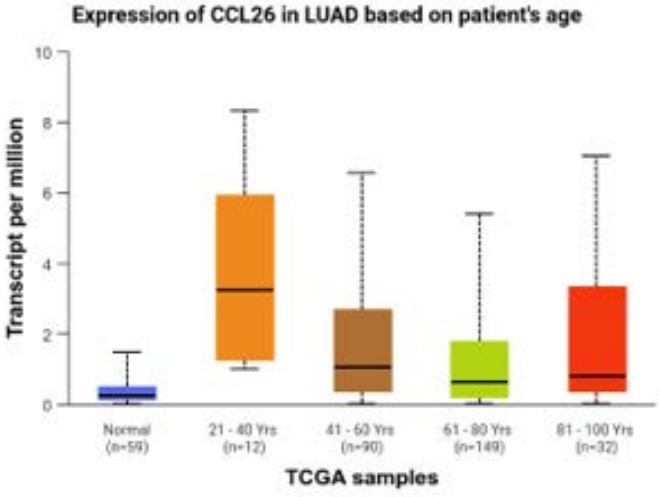
Figure 2.2. *CCL26 gene expression analysis in cancer stage in cancers have significant gene down regulation.*

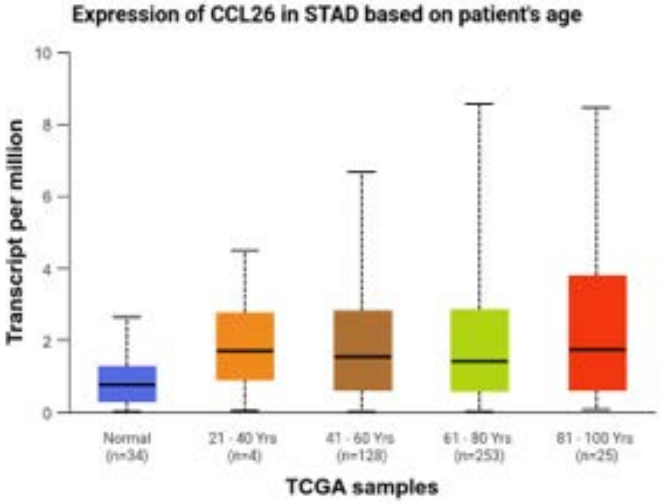

Expression of CCL26 in CHOL based on patient's age
Transcript per million
0
1
2
3
4
Normal (n=9)
21 - 40 Yrs (n=2)
41 - 60 Yrs (n=12)
61 - 80 Yrs (n=20)
81 - 100 Yrs (n=2)
TCGA samples


Expression of CCL26 in COAD based on patient's age
Transcript per million
0
2
4
6
8
10
Normal (n=41)
21 - 40 Yrs (n=12)
41 - 60 Yrs (n=90)
61 - 80 Yrs (n=149)
81 - 100 Yrs (n=32)
TCGA samples


Expression of CCL26 in ESCA based on patient's age
Transcript per million
0
5
10
15
20
Normal (n=11)
21 - 40 Yrs (n=3)
41 - 60 Yrs (n=89)
61 - 80 Yrs (n=76)
81 - 100 Yrs (n=15)
TCGA samples


Expression of CCL26 in HNSC based on patient's age
Transcript per million
0
5
10
15
20
Normal (n=44)
21 - 40 Yrs (n=20)
41 - 60 Yrs (n=236)
61 - 80 Yrs (n=237)
81 - 100 Yrs (n=24)
TCGA samples


Expression of CCL26 in LUAD based on patient's age
Transcript per million
0
2
4
6
8
10
Normal (n=59)
21 - 40 Yrs (n=12)
41 - 60 Yrs (n=90)
61 - 80 Yrs (n=149)
81 - 100 Yrs (n=32)
TCGA samples


Expression of CCL26 in STAD based on patient's age
Transcript per million
0
2
4
6
8
10
Normal (n=34)
21 - 40 Yrs (n=4)
41 - 60 Yrs (n=128)
61 - 80 Yrs (n=253)
81 - 100 Yrs (n=25)
TCGA samples

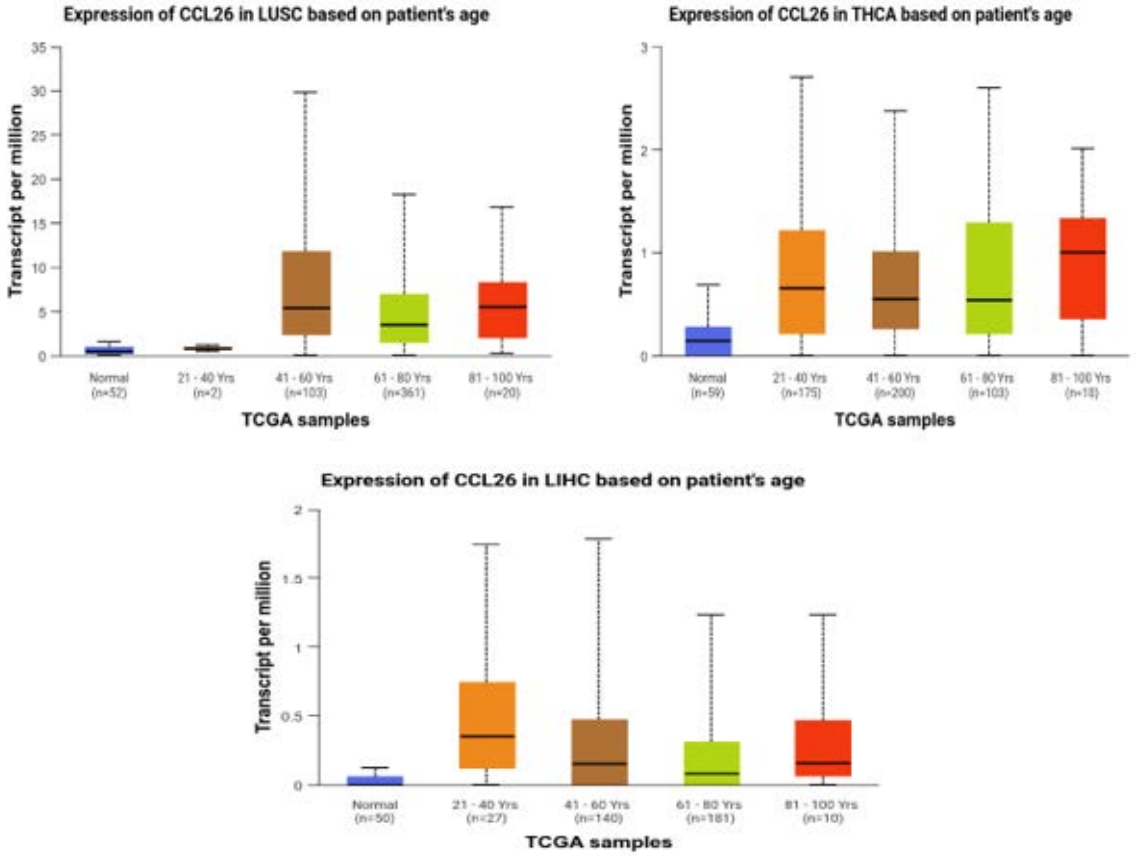


Figure 2.3. *CCL26 gene expression analysis with age in cancers have significant gene up regulation.*

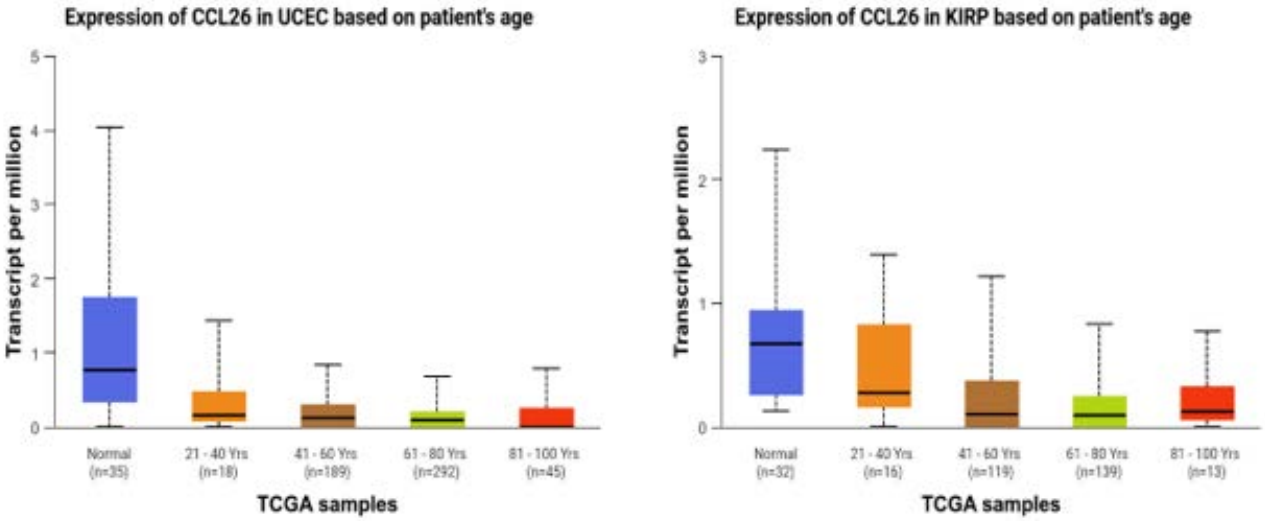
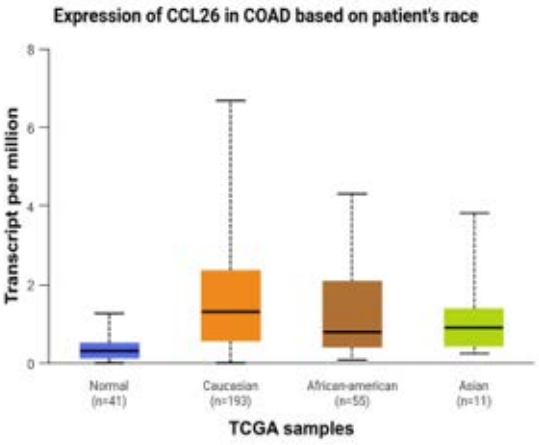
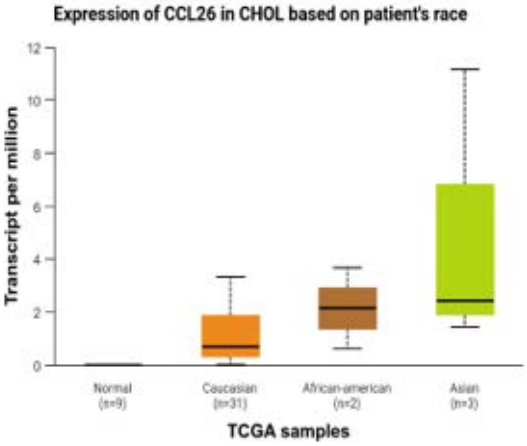
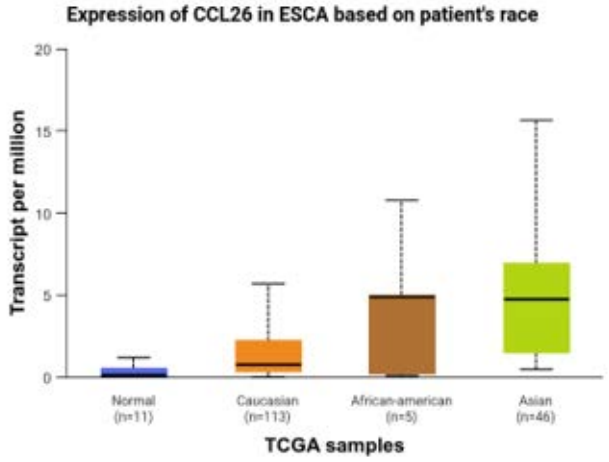
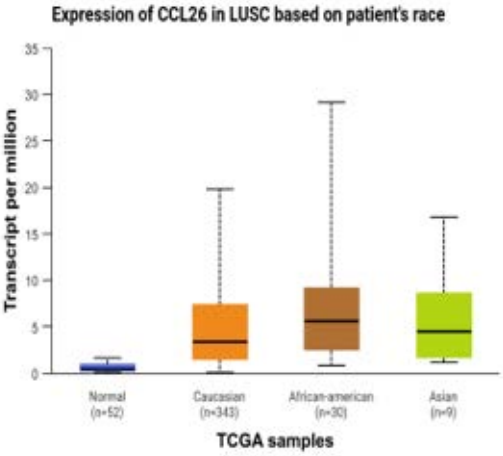


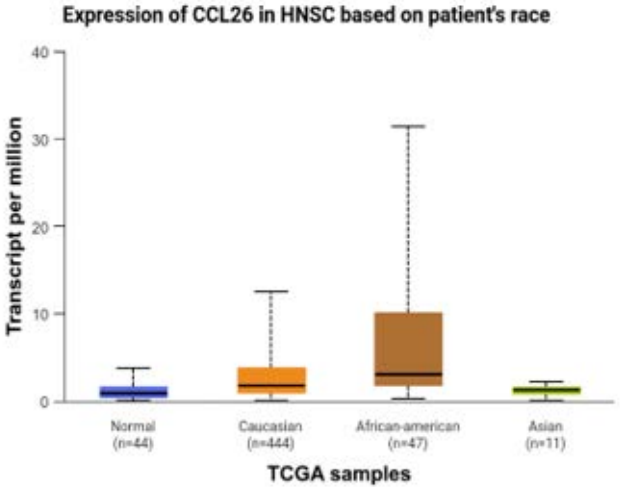
Figure 2.4. *CCL26 gene expression analysis with race in cancers have significant gene down regulation.*

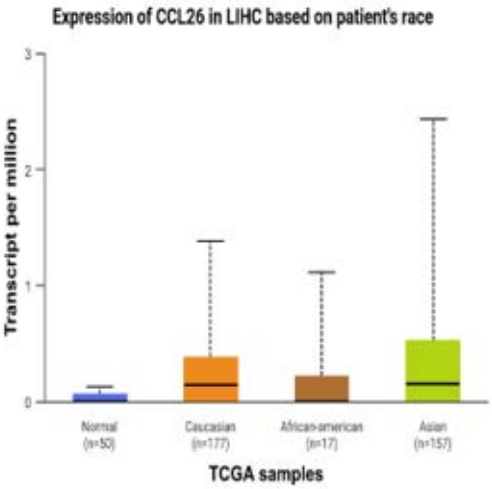

Expression of CCL26 in COAD based on patient's race
Transcript per million
Normal (n=41)
Caucasian (n=193)
African-american (n=55)
Asian (n=11)
TCGA samples


Expression of CCL26 in CHOL based on patient's race
Transcript per million
Normal (n=9)
Caucasian (n=31)
African-american (n=2)
Asian (n=3)
TCGA samples


Expression of CCL26 in ESCA based on patient's race
Transcript per million
Normal (n=11)
Caucasian (n=113)
African-american (n=5)
Asian (n=46)
TCGA samples


Expression of CCL26 in LUSC based on patient's race
Transcript per million
Normal (n=52)
Caucasian (n=343)
African-american (n=30)
Asian (n=9)
TCGA samples


Expression of CCL26 in HNSC based on patient's race
Transcript per million
Normal (n=44)
Caucasian (n=444)
African-american (n=47)
Asian (n=11)
TCGA samples


Expression of CCL26 in LIHC based on patient's race
Transcript per million
Normal (n=50)
Caucasian (n=177)
African-american (n=17)
Asian (n=157)
TCGA samples

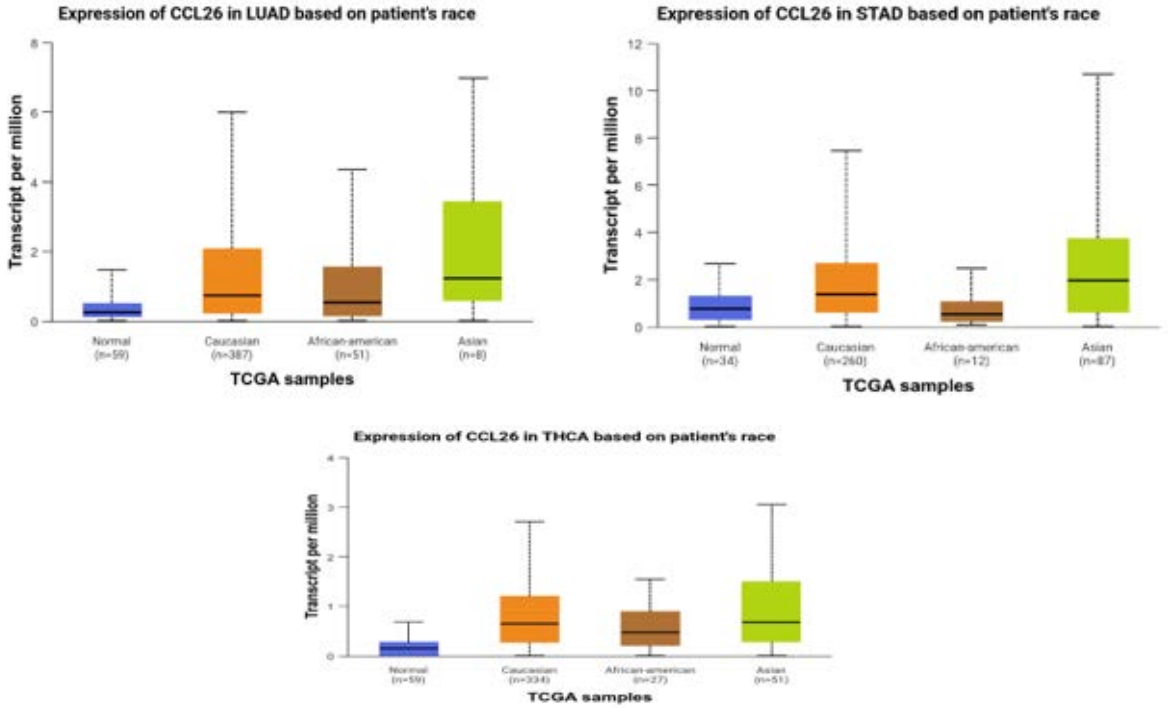


Figure 2.5. *CCL26 gene expression analysis with race in cancers have significant gene expression.*

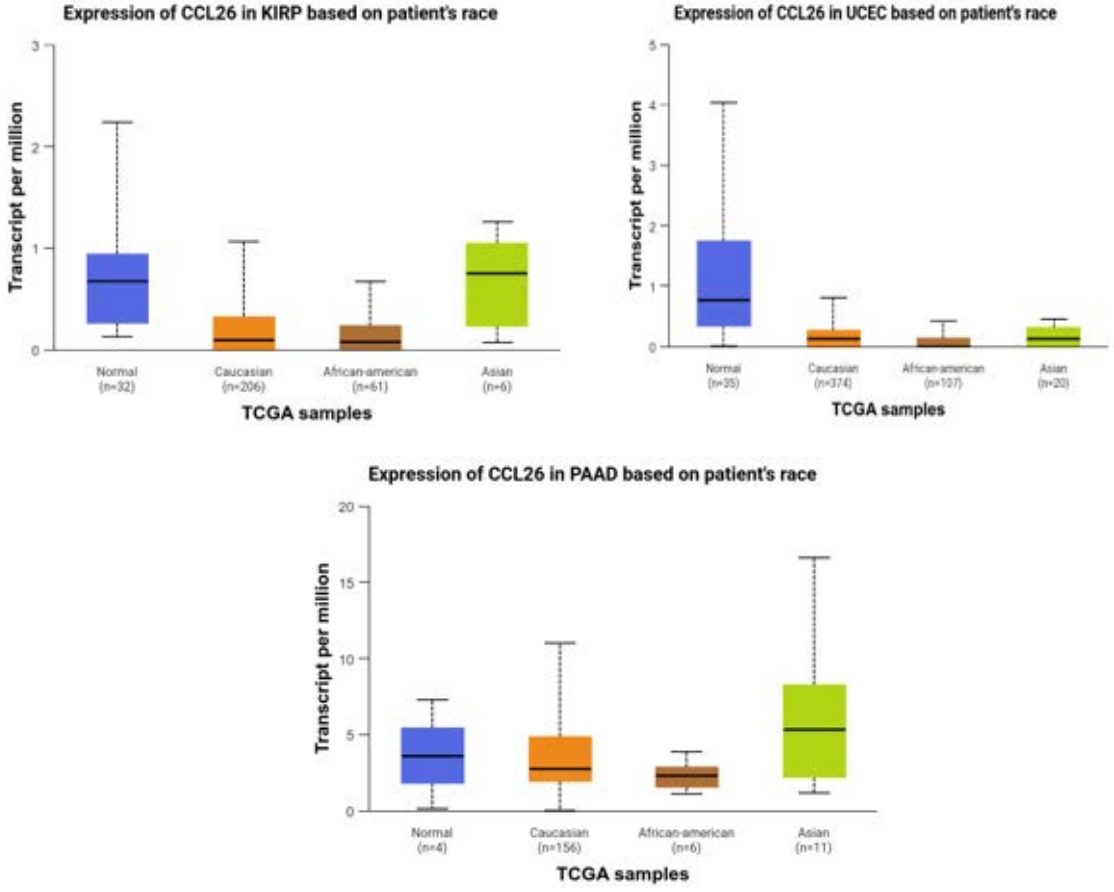
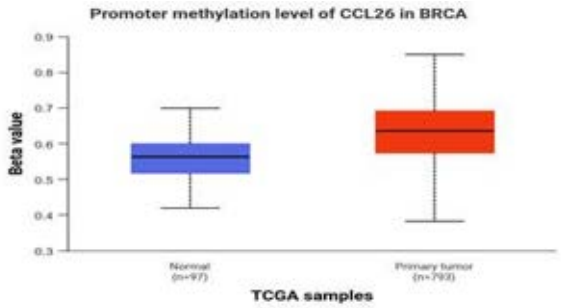
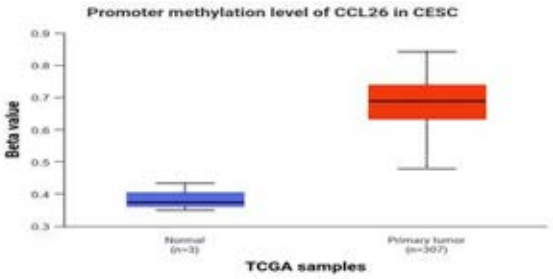
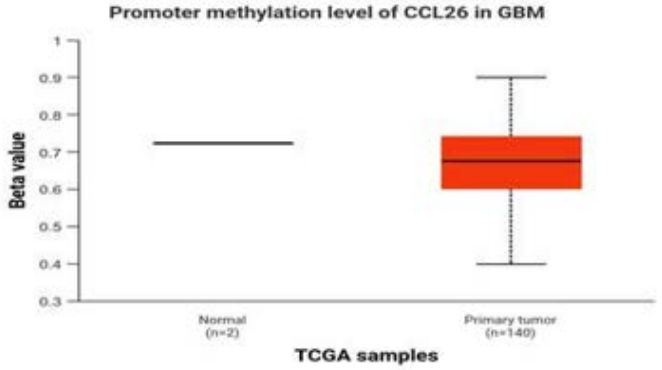


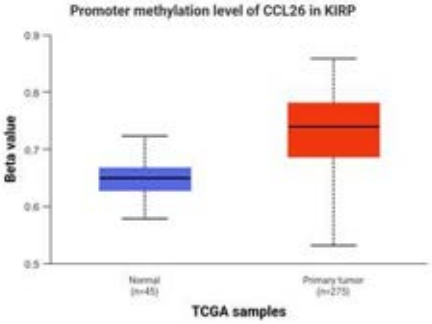
Figure 2.6. *ccl26 gene expression analysis with race in cancers have significant gene down regulation.*

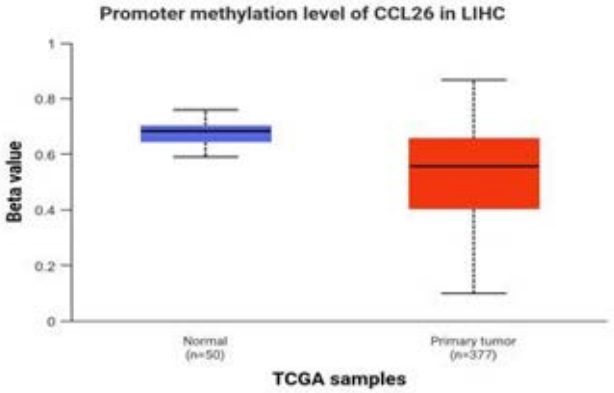
S3.

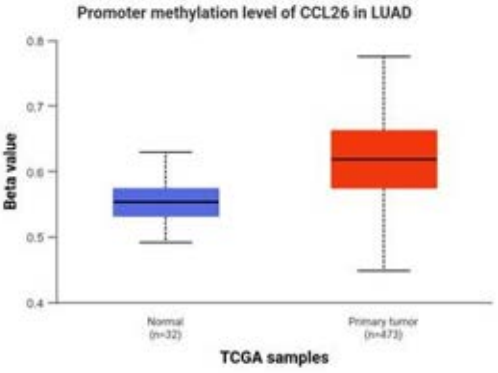

Promoter methylation level of CCL26 in BRCA
Beta value
Normal (n=97)
Primary tumor (n=793)
TCGA samples

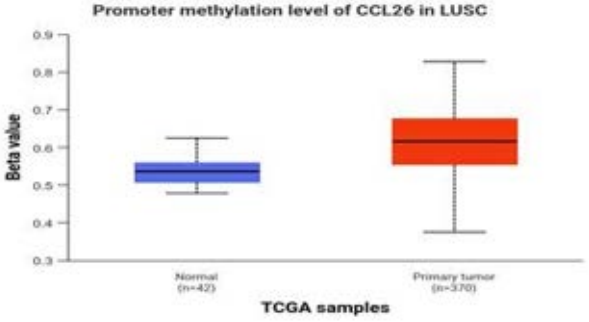

Promoter methylation level of CCL26 in CESC
Beta value
Normal (n=3)
Primary tumor (n=307)
TCGA samples

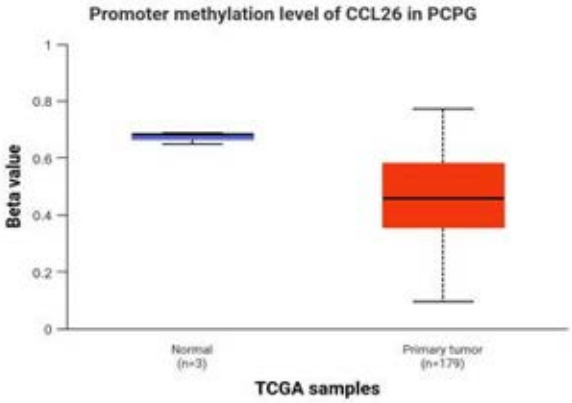

Promoter methylation level of CCL26 in GBM
Beta value
Normal (n=2)
Primary tumor (n=140)
TCGA samples


Promoter methylation level of CCL26 in KIRP
Beta value
Normal (n=45)
Primary tumor (n=275)
TCGA samples


Promoter methylation level of CCL26 in LIHC
Beta value
Normal (n=50)
Primary tumor (n=377)
TCGA samples


Promoter methylation level of CCL26 in LUAD
Beta value
Normal (n=32)
Primary tumor (n=473)
TCGA samples


Promoter methylation level of CCL26 in LUSC
Beta value
Normal (n=42)
Primary tumor (n=370)
TCGA samples


Promoter methylation level of CCL26 in PCPG
Beta value
Normal (n=3)
Primary tumor (n=179)
TCGA samples

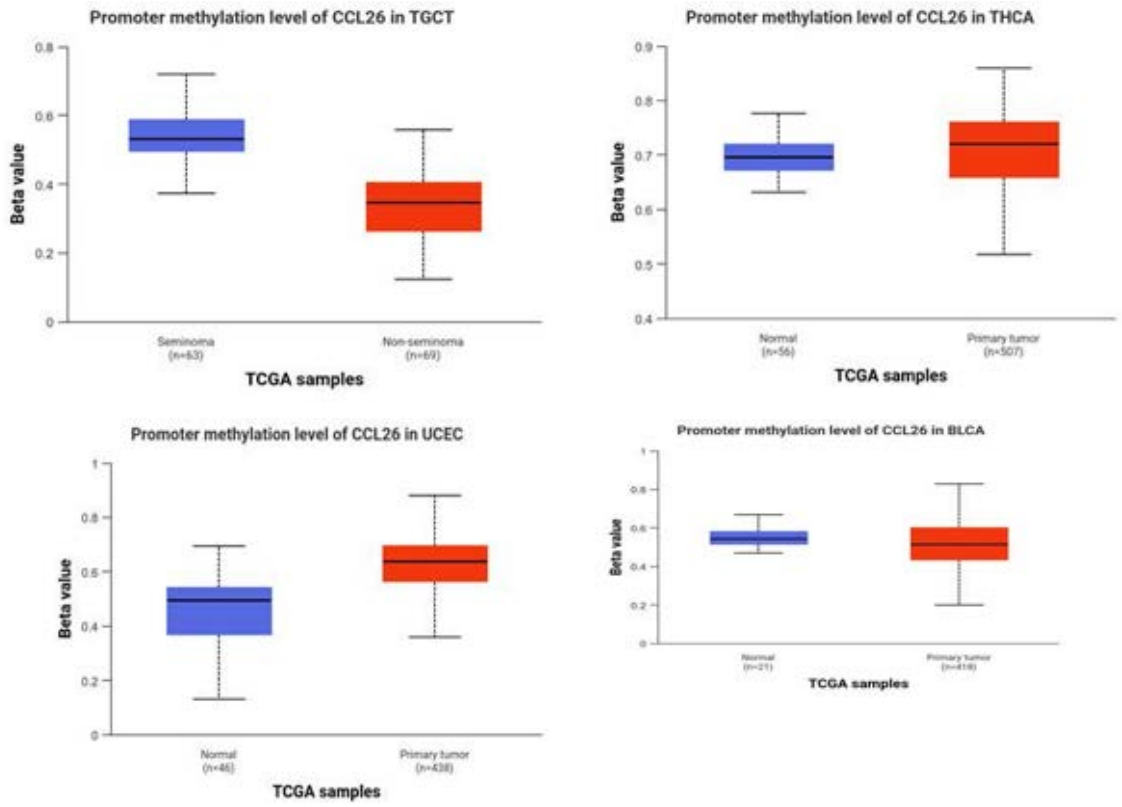
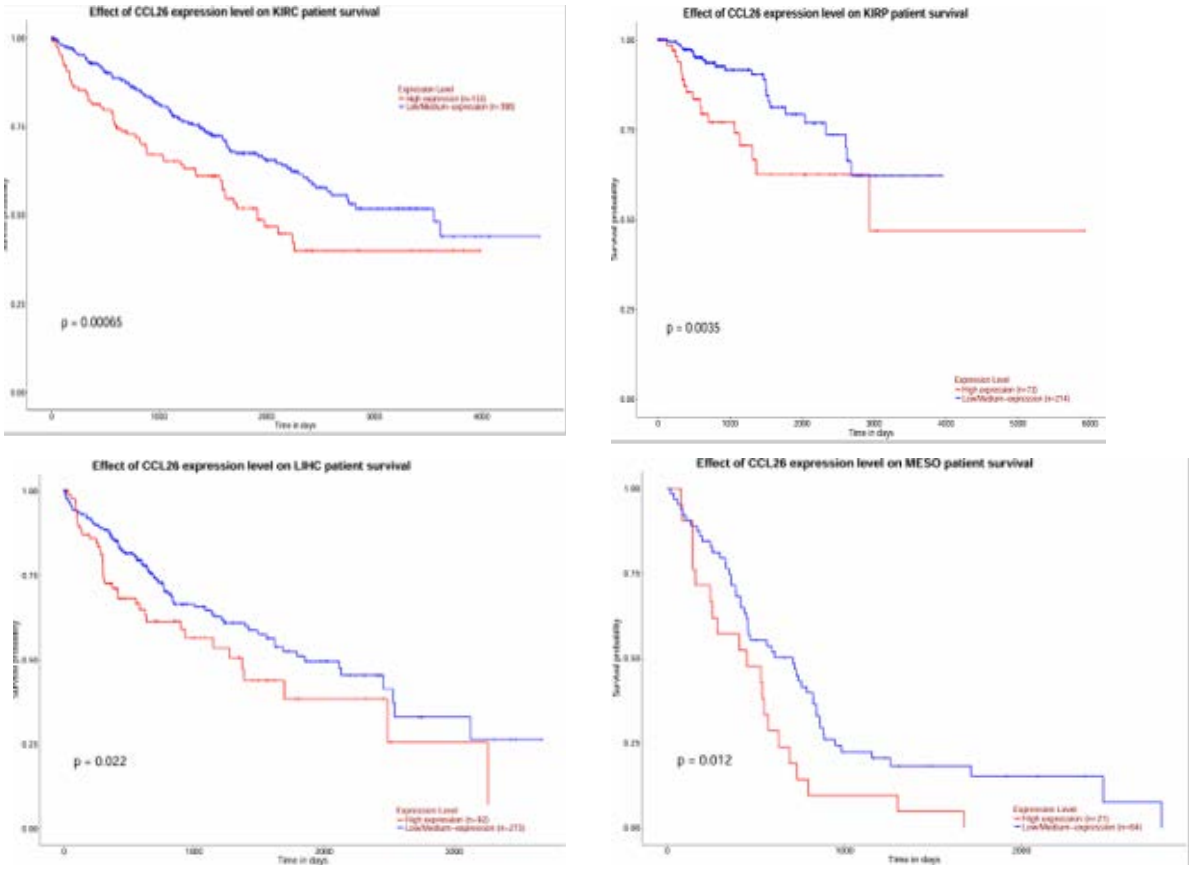


Figure 2.7. *Methylation patterns across cancer types.*
Hypermethylated cancers (n=6): BRCA, CESC, KIRP, LUAD, LUSC, THCA
Hypomethylated cancers (n=4): LIHC, PCPG, GBM, TGCT

S3.

Figure 3.1.



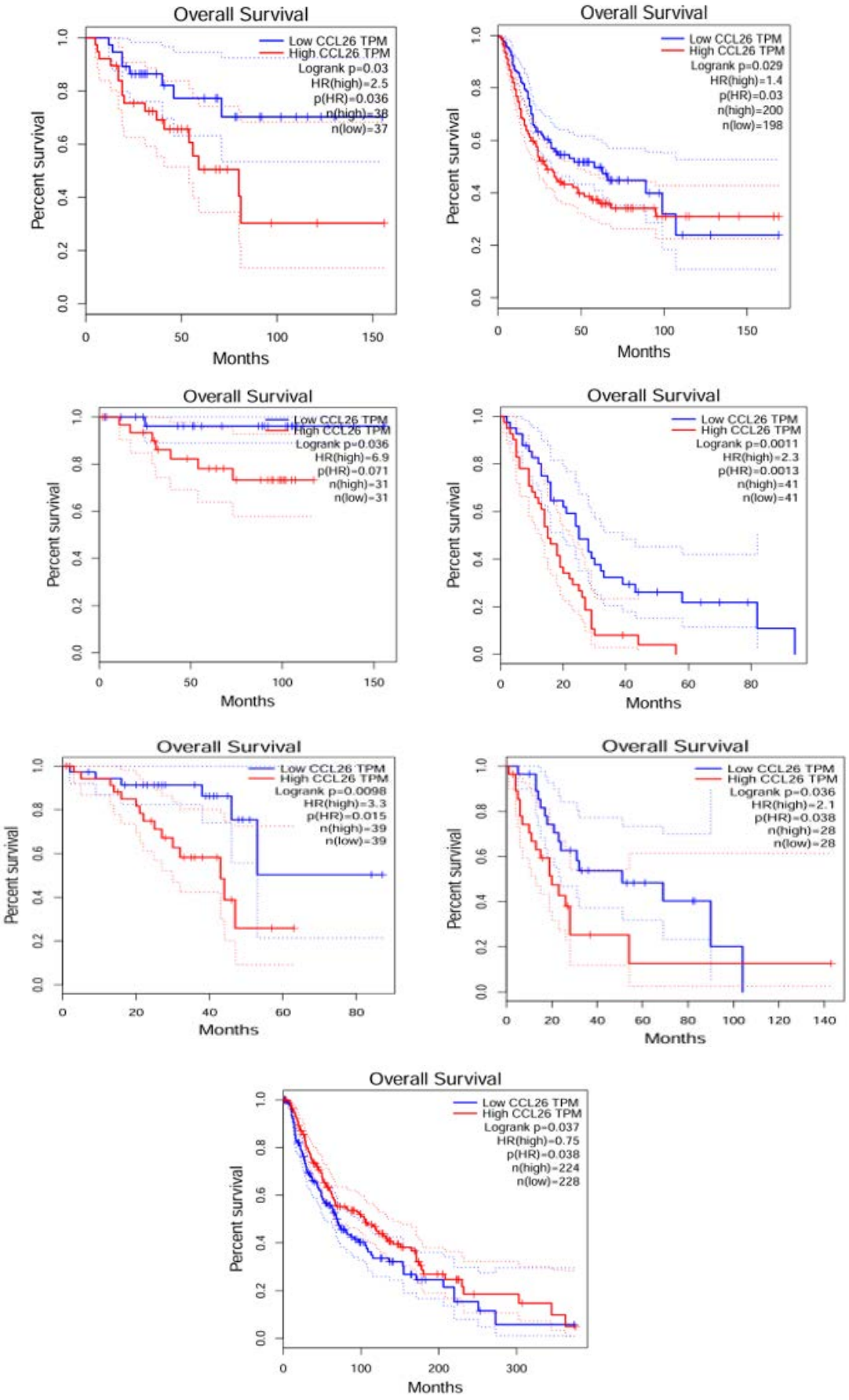


Figure 3.2. *The correlation between CCL26 expression and survival outcomes in various cancers using GEPIA database ACC, BLCA, KICH, MESO, SKCM, UCS, UVM.*

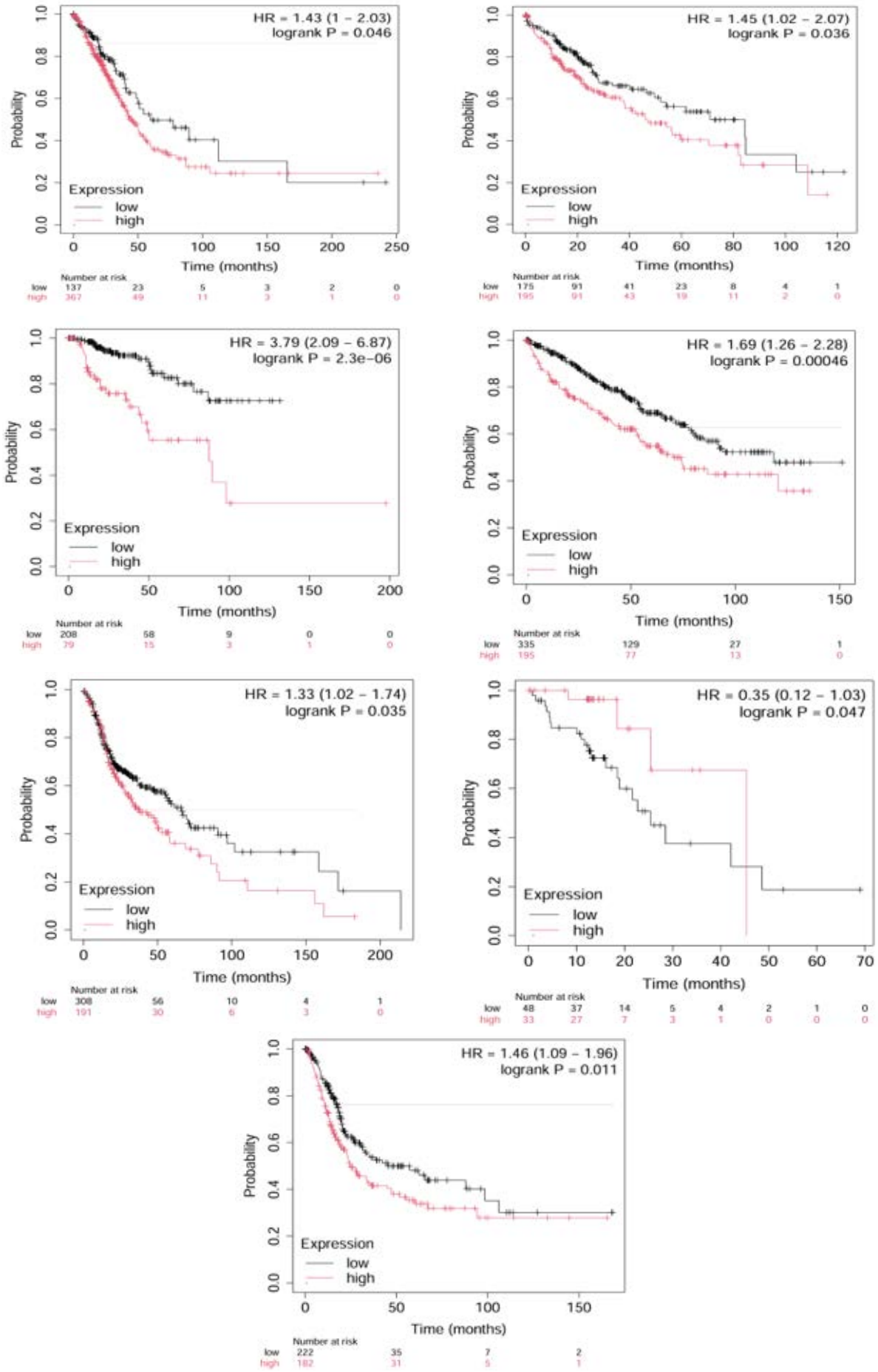


Figure 3.3. *The correlation between CCL26 expression and survival outcomes in various cancers using Kaplan-Meier plotter database.*

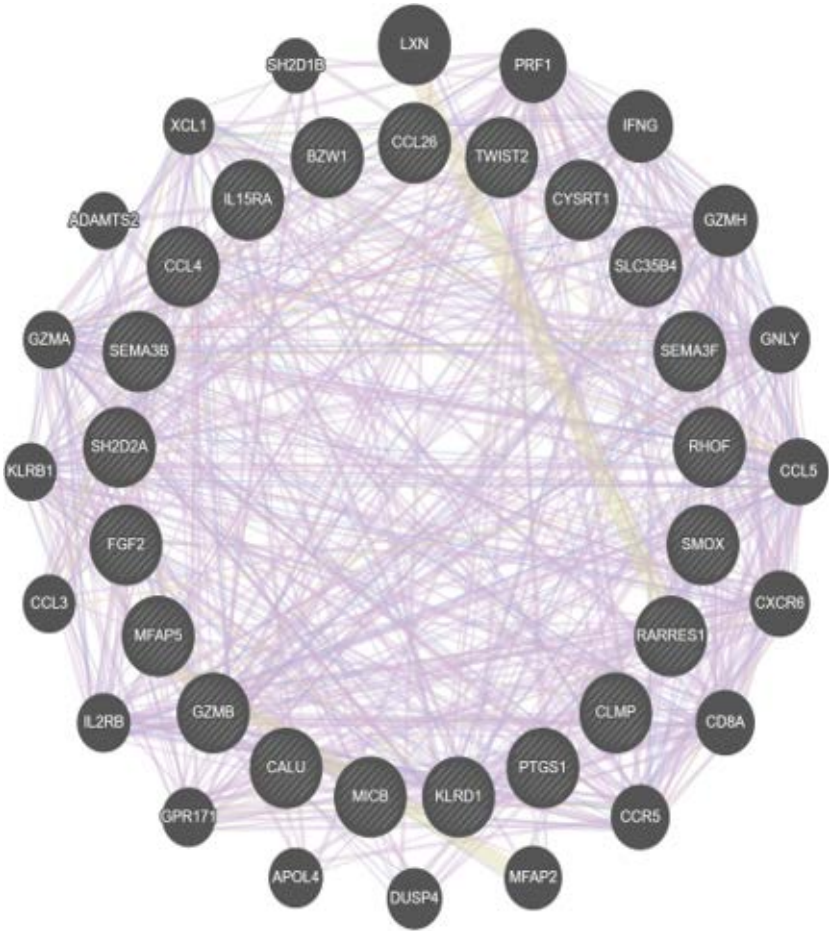
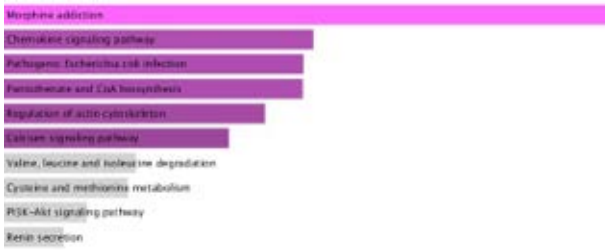
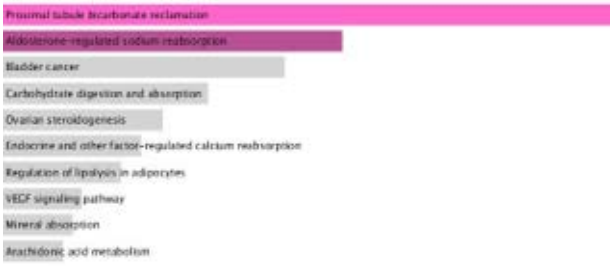


Figure 4.1. *HERE: Gene networks for high-expressing cancers.*

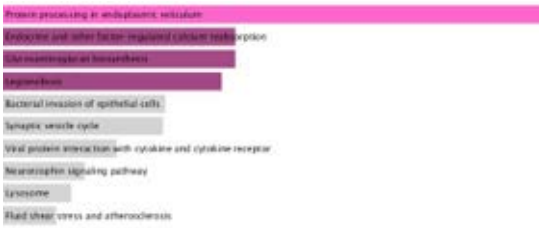
https://genemania.org/search/homo-sapiens/ccl26



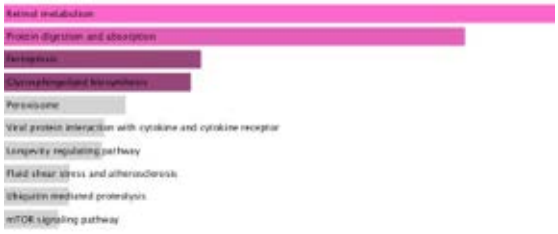
COAD

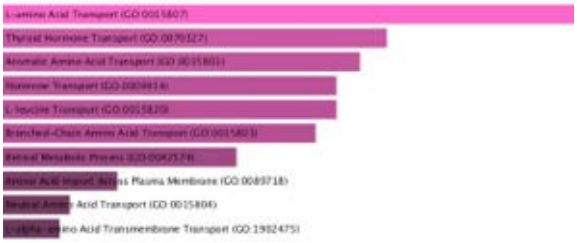


ESCA



LUSC





LAUD