

Global Research Status of Tertiary Lymphoid Structures: A Bibliometric Analysis

Can Yang, Haodong Guo, Xiaopeng Ma

Shanghai Research Institute of Acupuncture and Meridian, Shanghai University of Traditional Chinese Medicine, Shanghai 201203, China

Corresponding author: Xiaopeng Ma (e-mail: yc23232303@163.com)

ABSTRACT Tertiary lymphoid structures are organized lymphocyte aggregates ectopically formed at sites of chronic inflammation or tumorigenesis and are closely associated with pathological processes including autoimmune diseases, organ transplantation, tumors, and aging. In recent years, academic output on tertiary lymphoid structures has grown rapidly, but a systematic global perspective remains limited. This study conducts a bibliometric analysis of tertiary lymphoid structure-related literature retrieved from the Web of Science Core Collection. Using VOSviewer, CiteSpace, Rbibliometrix, and quantitative visualization tools, the study systematically analyzes publication trends, national and institutional distribution, author and journal contributions, highly cited literature, citation bursts, and keyword evolution. A total of 2,496 publications are included, consisting of 1,827 articles and 669 reviews. Results show that immunology and oncology are the dominant disciplines, the United States leads in publication output, and France demonstrates strong citation influence through institutional and author collaboration networks. Citation and keyword evolution analyses indicate that immunotherapy has become the core direction of current and future research. The findings provide an objective basis for understanding scientific development dynamics and identifying emerging research strategies in this field.

INDEX TERMS tertiary lymphoid structures, bibliometric analysis, immunotherapy, immune response, visualization analysis

I. INTRODUCTION

TERTIARY lymphoid structures (TLSs) are non-encapsulated lymphocyte aggregates ectopically formed at sites of chronic inflammation or tumorigenesis [1-3]. Their structural composition is highly similar to that of secondary lymphoid organs, mainly including B-cell zones, T-cell zones, follicular dendritic cells, resident dendritic cells, and peripheral lymph node addressin-positive high endothelial venules [4-6]. TLSs drive a variety of effector responses of B cells and T cells, including antibody formation, effector function induction, class switch recombination, and clonal proliferation, and play a key role in the sustainment and regulation of immune responses in the body [7].

Existing studies show that the formation of TLSs is closely related to various pathological processes such as autoimmune diseases [8,9], organ transplantation [10], tumors [11], and aging [12,13]. Notably, the function of TLSs is significantly pathologically context-dependent, exhibiting dual characteristics of protective and detrimental effects. In situations requiring enhanced immunity such as tumors and infections, the presence and maturation of TLSs usually predict a favorable prognosis [14-16]; whereas in situations requiring immunosuppression such as rheumatoid arthritis

and aging-related diseases, TLSs may promote the activation of autoreactive lymphocytes and autoantibody production, indicating a poor prognosis [17-19]. This dual attribute makes TLSs an important target with both biomarker value and therapeutic intervention potential. With the rapid accumulation of TLSs-related literature, a systematic method is urgently needed to objectively evaluate the overall landscape of this field. As a scientific tool for quantitative analysis of academic research output and impact using statistical methods, bibliometrics can reveal the historical trajectory, knowledge structure, cooperation network, and frontier trends of disciplines, providing an important reference for research policy formulation and academic evaluation [20]. At present, there is still a gap in systematic bibliometric analysis targeting the TLSs field, while previous bibliometric studies have explored broader themes in tumor immunology or autoimmune diseases, few have focused specifically on TLSs as an independent topic. This study builds upon established bibliometric methodologies used in adjacent clinical fields to provide a more granular perspective on TLSs development trajectory. Therefore, bibliometric methods were adopted, and visual tools such as VOS Viewer, CiteSpace, and Rbibliometrix were comprehensively used to conduct a com-

prehensive analysis of TLSs-related literature in the Web of Science database, so as to provide a scientific reference for future research in this field.

II. MATERIALS AND METHODS

A. DATA SOURCES

Web of Science Core Collection (WoSCC) is recognized as an authoritative data source for bibliometric research due to its wide coverage, high data quality, and complete citation information [20]. Compared with databases such as PubMed and Scopus, WoSCC has significant advantages in literature coverage and citation tracking in interdisciplinary fields, and can more completely present the interdisciplinary knowledge diffusion path of TLSs research. The retrieval time node was set to October 1, 2024, to ensure the timeliness and integrity of the data. All raw data were exported in plain text format (.txt) and then imported into Microsoft Excel 2021 for structured sorting, covering fields such as publication year, document type, country/region, institutional affiliation, author name, source journal, keywords, and cited literature, providing a standardized data basis for subsequent multi-dimensional bibliometric analysis.

B. RETRIEVAL STRATEGY

Retrieval terms were formulated based on the Medical Subject Headings (MeSH) and representative literature in this field. A retrieval formula was constructed with the topic field (TS), including a series of synonymous terms such as “Tertiary Lymphoid Structure”, “Tertiary Lymphoid Organ”, “Ectopic Lymphoid-Like Structures”, “Ectopic Lymphoid Tissue”, “Ectopic Lymphoid Follicle”, and supplementary retrieval was conducted combined with the subject term “TLSs”. The time range was set from January 1, 1900 to October 1, 2024. Document types were limited to articles and reviews, and languages were limited to English. The purpose of adopting the dual retrieval strategy was to reconcile standardized medical terminology with the diverse nomenclature used in existing literature, thereby enhancing the retrieval’s comprehensiveness and ensuring the inclusion of core research in the field.

C. DATA COLLECTION AND SCREENING CRITERIA

Inclusion criteria consist of three aspects: the research topic clearly involves tertiary lymphoid structures (TLSs); document types are articles and reviews; and the language is English. Exclusion criteria include nonacademic document types such as news reports, conference abstracts, errata, conference proceedings, letters, editorials, book chapters, and commentaries. According to the above criteria, a total of 3044 publications were initially retrieved. Following the removal of duplicates ($n=112$) and the exclusion of non-academic document types such as news, letters, and meeting abstracts ($n=436$), and 2496 were finally included after step-by-step screening, including 1827 articles and 669 reviews, involving 16,812 authors and 721 source journals. Data extraction was independently performed by two researchers,

and divergent entries were resolved through negotiation and discussion to ensure the accuracy and consistency of data extraction, providing a reliable literature basis for subsequent analysis.

D. BIBLIOMETRIC AND VISUAL ANALYSIS METHODS

Bibliometric analysis comprehensively used three tools: VOS Viewer (v1.6.13), CiteSpace (Drexel University), and R-bibliometrix, each performing its own duties and complementing each other. VOS Viewer was used to construct co-occurrence networks and co-citation networks of countries/regions, institutions, authors, journals, and keywords, intuitively presenting the output scale and collaboration intensity of each element through node size and connection weight [21,22]. CiteSpace focuses on capturing dynamic knowledge evolution, identifying conceptual shifts and emerging research trends in this field through journal dual-map overlay, keyword timeline visualization, and citation burst detection [23,24]. R-bibliometrix undertakes the quantitative description function of keyword evolution, supplementing the temporal variation pattern of research hotspots. Quantitative statistical analysis was completed using GraphPad Prism 9.0 to draw trend curves of annual publication volume and citation volume, so as to achieve a multi-level and multi-dimensional systematic evaluation of the overall development trend of the TLSs field (Fig. 1). The detailed network visualizations, including co-authorship networks, co-citation clusters, and keyword evolution maps generated by VOSviewer and CiteSpace, are integrated into the following sections to substantiate the claims regarding research collaboration and thematic shifts.

III. RESULTS

A. ANALYSIS OF GLOBAL PUBLICATION TRENDS

The global academic output of TLS-related research was sporadically documented as early as 1935 in the Web of Science database; however, these early records often represent incidental terminology usage rather than focused study of the structures. Systematic research into the formation and function of TLSs truly began to emerge in the 1990s, with a qualitative leap occurring around 2018. Annual publications and citations entered a phase of rapid growth, which is highly consistent with the intensive publication of several landmark studies in Nature series journals in the same year. This suggests that high-impact breakthroughs exert a significant positive stimulus on research enthusiasm in this field. After strict application of inclusion and exclusion criteria, a total of 2496 publications were finally included, comprising 1827 articles and 669 reviews, involving 16,812 authors and 722 journals. Disciplinary analysis revealed that immunology (886 publications, 35.50%) and oncology (579 publications, 23.20%) are the two dominant disciplines, while cell biology (197 publications, 7.89%) and medical experimental research (193 publications, 7.73%) serve as important complements, with an increasingly prominent interdisciplinary tendency (164 publications, 6.57%). Such

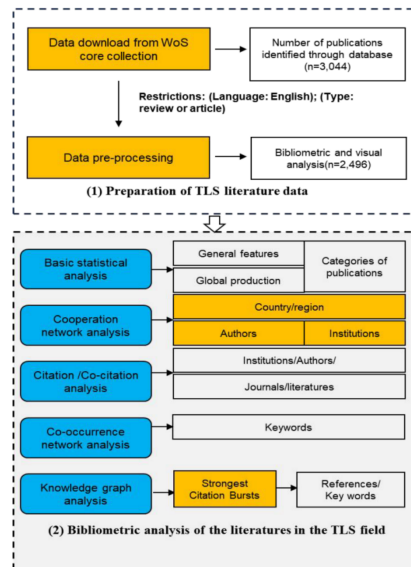


FIGURE 1. Flowchart of the study process

a disciplinary distribution indicates that TLS research is not confined to immunology alone; its deep integration with tumor biology has formed a stable interdisciplinary knowledge domain, implying that cross-disciplinary cooperation will continue to define the developmental trajectory of this field (Table 1, Fig. 2).

TABLE 1. Overview of literature in the TLSs field

Index	Value
Total included literature	2496
Articles	1827 (73.20%)
Reviews	669 (26.80%)
Total number of authors involved	16812
Number of source journals	721
Data retrieval deadline	October 1, 2024
Data source	Web of Science Core Collection

B. NATIONAL/REGIONAL DISTRIBUTION AND COOPERATION NETWORK

Seventy-five countries and regions have produced literature in the TLSs field, but the agglomeration effect of academic resources is very obvious. The United States ranks first with 804 articles (22.89%), followed by China (535 articles, 15.23%), and Germany (262 articles, 7.46%), France (229 articles, 6.52%), and the United Kingdom (228 articles, 6.49%) constitute the second echelon. However, measuring the influence of various countries solely by the number of publications has obvious limitations — in the total link strength of the cocitation network, the United States (17881) far exceeds other countries, when considering the average impact per paper, France and the United Kingdom exhibit high citation weights relative to their publication volumes, partly due to their early original contributions in the field which have had more time to accumulate citations, and

although China (12714) ranks second, the citation weights of France (10794) and the United Kingdom (7698) are disproportionately high relative to their publication volumes. This gap reflects the profound accumulation of original contributions of France and the United Kingdom in TLSs research. Co-authorship network analysis shows that the densest bilateral collaboration axis in the world has been formed between the United States and France. Quantitative analysis of international collaboration reveals that the co-authorship link strength between the US and France (link strength = 84) significantly exceeds that of US-China (62) and US-UK (58) collaborations, providing robust evidence for this primary axis. Many French institutions have long been deeply engaged in basic TLSs research, making them a pivotal hub node in the international cooperation network. In contrast, despite the large volume of publications, China has an obvious mismatch between the link depth in the international co-authorship network and its ranking, suggesting that Chinese scholars have great room for improvement in both improving research quality and expanding the breadth of international cooperation [21] (Table 2, Fig. 3).

TABLE 2. Top 5 countries/regions by publication volume and citation link strength in the TLSs field

Rank	Country/Region	Publication Volume	Proportion	Total Citation Link Strength
1	United States	804	22.89%	17881
2	China	535	15.23%	12714
3	Germany	262	7.46%	9214
4	France	229	6.52%	10794
5	United Kingdom	228	6.49%	7698

C. ANALYSIS OF INSTITUTIONAL PUBLICATION VOLUME AND CITATIONS

A total of 2986 academic institutions worldwide have contributed to the TLSs field, and the academic concentration

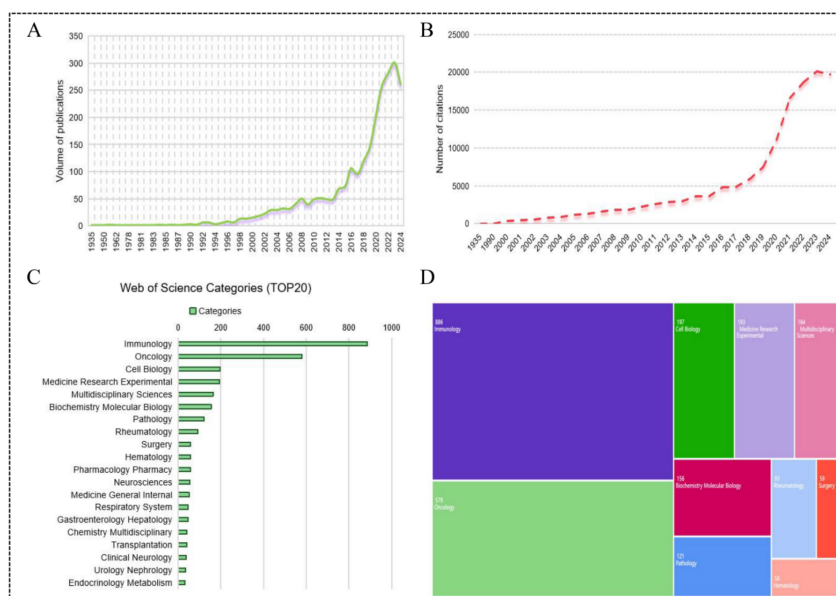


FIGURE 2. Global trends and disciplinary contributions in TLS research. (A) Annual publication volume on TLSs (1935-2024) (B) The annual citation count related to TLSs (C) The top 20 science categories related to TLSs (D) Publications quantity in the top 10 science categories

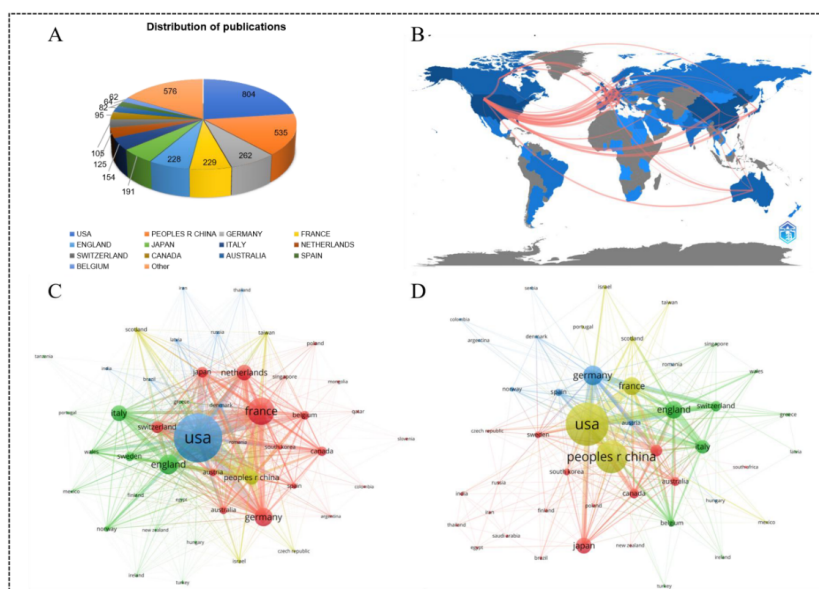


FIGURE 3. Countries/regions participating in the TLS research field. (A) The distribution of publications across countries/regions (B) Productions and collaboration map of countries/regions related to TLSs (C) Mapping of the countries/regions co-authorship analysis on TLSs field (D) Mapping of the countries/regions citation network analysis on TLSs field

at the institutional level is also prominent. In terms of publication volume, the French National Institute of Health and Medical Research (Inserm) ranks first globally with 60 articles (4.28%). Chinese institutions have performed brilliantly — Fudan University (52 articles), Sun Yat-sen University (51 articles), and Shanghai Jiao Tong University (42 articles) all rank among the top five, reflecting the rapid follow-up trend of domestic universities in this field. However, turning to the citation dimension, the pattern is fundamentally reversed: Inserm ranks first globally with 11,575 citations, followed by Paris Descartes University

(10,849 citations) and Sorbonne University (8,840 citations). French institutions also maintain an absolute advantage in the total link strength of the citation network (Inserm: 5635, Paris Descartes University: 4911). In the co-authorship co-operation network, Inserm (94), Paris Descartes University (80), Sorbonne University (64), Sorbonne Université (54), and Hôpital Européen Georges-Pompidou (53) constitute the core collaboration cluster for global TLSs research. This Paris-centered institutional ecosystem plays a decisive role in promoting cross-institutional knowledge sharing of TLSs, and also explains to a considerable extent why France can

still maintain a pivotal position in citation influence despite its publication volume being far lower than that of the United States and China [22] (Table 3, Fig. 4).

TABLE 3. Top 5 institutions by publication volume and citation status in the TLSs field

Rank	Institution	Publication Volume	Proportion	Citation Times	Total Co-authorship Link Strength
1	Inserm (France)	60	4.28%	11575	94
2	Fudan University (China)	52	3.71%	—	—
3	Sun Yat-sen University (China)	51	3.64%	—	—
4	Queen Mary University of London (UK)	50	3.56%	—	—
5	Shanghai Jiao Tong University (China)	42	2.99%	—	—
	Paris Descartes University (France)	—	—	10849	80

D. CORE AUTHOR CONTRIBUTIONS AND CO-AUTHORSHIP NETWORK

Analysis at the individual scholar level reveals a small group of highly active academic leaders in the TLSs field. Sautès-Fridman Catherine ranks first in publication volume with 42 articles (3.29%), and Pitzalis Costantino (38 articles, 2.98%), Bombardieri Michele (36 articles, 2.82%), and Dieu-Nosjean Marie-Caroline (34 articles, 2.66%) constitute the core first echelon. Co-authorship network analysis covers 147 authors meeting the minimum threshold of 5 articles. Sautès-Fridman Catherine (total link strength: 184) and Dieu-Nosjean Marie-Caroline (172) form a dual-core pattern in the network, and together with Damotte Diane (132), Pitzalis Costantino (128), and Germain Claire (119), they weave the closest academic collaboration network in this field. In terms of citation influence, the total citation link strength of Sautès-Fridman Catherine's papers reaches 6077, meaning that she is not only the main output body but also a key transmission node for knowledge diffusion. Co-citation analysis presents a different picture: Dieu-Nosjean Marie-Caroline (19350), Luther Sanjiv A (14187), and Lauss Martin (12398) rank top in co-citation strength, indicating that the work of these scholars has been widely cited repeatedly as the knowledge cornerstone of this field, forming a core reference group with high knowledge cohesion [22]. A comprehensive comparison of the two dimensions shows that there is partial overlap but not complete consistency between output leaders and citation core figures, and the nonlinear relationship between publication quantity and knowledge influence is also valid in the TLSs field (Table 4, Fig. 5).

TABLE 4. Comprehensive indicators of academic contributions of core authors in the TLSs field (top 5)

Rank	Author	Publication Volume	Proportion	Total Co-authorship Link Strength	Total Citation Link Strength
1	Sautès-Fridman C	42	3.29%	184	6077
2	Pitzalis C	38	2.98%	128	4962
3	Bombardieri M	36	2.82%	115	4320
4	Dieu-Nosjean MC	34	2.66%	172	5202
5	Barone F	19	1.49%	76	1895

E. JOURNAL PUBLICATION AND CO-CITATION ANALYSIS

The academic communication pattern in the TLSs field presents a typical characteristic of “head concentration,

long-tail dispersion”. *Frontiers in Immunology* (impact factor 5.7) leads the publication volume ranking with 238 articles (15.29%), followed by *Journal of Immunology* (83 articles), *Cancers* (59 articles), *Journal for Immunotherapy of Cancer* (51 articles), and *OncoImmunology* (38 articles). Notably, there is a structural difference between the ranking of publication volume and co-occurrence network link strength: although *Frontiers in Immunology* ranks first in both dimensions (link strength 4520), *Nature* (impact factor 50.5, link strength 1142) and *Immunity* (25.5, 803) maintain a powerful radiation force disproportionate to their publication volume by virtue of high single-paper impact, reflecting the multiplier effect of top journals in shaping the domain knowledge framework. In terms of co-citation analysis, *Journal of Immunology* (link strength 796450) and *Nature* (657163) rank top, indicating that TLSs research relies on a historical knowledge base centered on classic immunology journals in knowledge citation. The journal dual-map overlay generated by CiteSpace further reveals that the knowledge flow in this field mainly takes “molecular/biology/immunology” citing “molecular/biology/genetics” as the main path, and forms two-way knowledge exchange with “medicine/ clinic”, objectively presenting the disciplinary expansion trajectory of TLSs research from basic immunology to tumor clinical transformation [21-22] (Table 5, Fig. 6).

TABLE 5. Top 5 journals by publication volume and impact factor in the TLSs field

Rank	Journal	Impact Factor	Publication Volume	Proportion	Total Co-occurrence Link Strength
1	<i>Frontiers in Immunology</i>	5.7	238	15.29%	4520
2	<i>Journal of Immunology</i>	3.6	83	5.33%	1450
3	<i>Cancers</i>	—	59	3.79%	—
4	<i>Journal for Immunotherapy of Cancer</i>	—	51	3.28%	—
5	<i>OncoImmunology</i>	6.5	38	2.44%	956

F. HIGHLY CITED LITERATURE AND CITATION BURST ANALYSIS

The distribution structure of highly cited literature reveals the phasic migration of the knowledge focus in the TLSs field. The most frequently cited literature is the study by Binnewies M *et al.* published in *Nature Medicine* in 2018 (3397 citations), followed by the work by Becht E *et al.* published in *Genome Biology* in 2016 (2087 citations) and the achievement by Wei SC *et al.* published in *Cancer Discovery* in 2018 (2034 citations). In the reference co-citation network, the cluster with the highest link strength consists of three independent studies published in *Nature* in the same period of 2020 — Helmink BA *et al.* [14] (7320), Cabrita R *et al.* (7213), and Sautès-Fridman C *et al.* (7019). The high co-citation structure of these three papers indicates that they jointly laid the field consensus that “TLSs maturity is positively correlated with tumor immunotherapy response”, becoming a knowledge anchor driving the outbreak of subsequent research. In terms of citation burst analysis, the review by Pitzalis Costantino *et al.* [25] has the highest burst strength of 53.21 and lasts

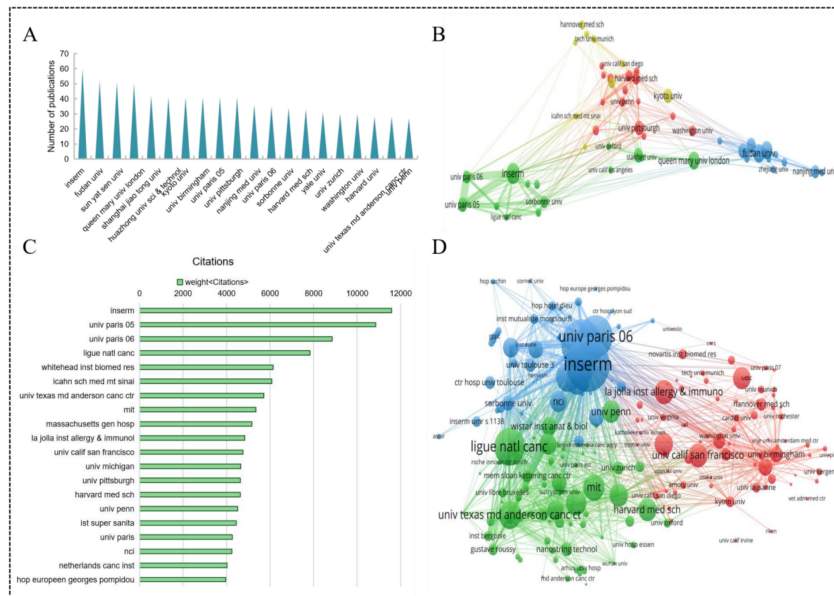


FIGURE 4. Academic institutions contributing to the field of TLSs. (A) The top 20 global academic institutions for TLSs-related publications (B) Analysis map of collaborative authorship relationships among 50 institutions in TLSs field. The size of the nodes reflects the publication counts. Edge weight (line thickness) is proportional to collaborative intensity between nodes (C) The top 20 global academic institutions for TLSs-related citations (D) Academic institutions citation network map. Nodes represent institutions, with size scaling proportionally to citation frequency

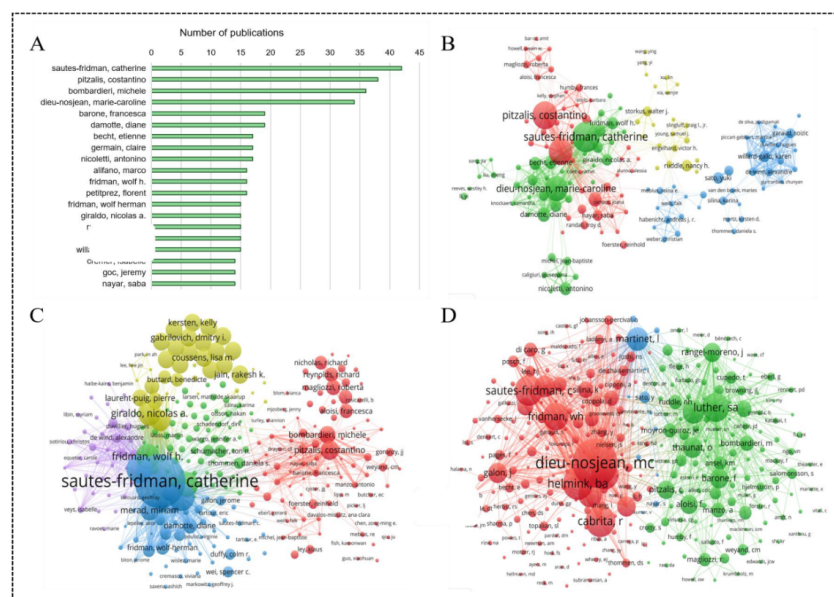


FIGURE 5. Author analysis of research in TLSs field. (A) The top 20 global academic authors for TLSs field (B) A co-authorship analysis map of 222 authors in TLSs field (C) Mapping of the cited authors in TLSs field (D) Mapping of the co-cited authors in TLSs field

for 5 years (2015-2019), systematically constructing the research paradigm of TLSs in human pathological contexts; the study by Silina et al. [26] still maintains an active burst of 25.25 until 2024, making it the literature that continues to attract the most attention currently. The mechanism revealed by this study — the role of CXCL13/CXCL12/PD-L1 positive epithelial cell niche in the maintenance of TLSs — provides a direct basis for targeted intervention. Citation cluster analysis identified 8 thematic clusters, with Cluster #0 (tertiary lymphoid structures, 314 articles) being the

largest. The earliest research direction is Cluster #2 (homing chemokines, average year 2000). The recent parallel activity of Cluster #0 and Cluster #5 (T cells, 2019) marks that this field has entered the in-depth stage of mechanistic analysis from phenotypic characterization [23-24] (Table 6, Figs. 7 a and 8).

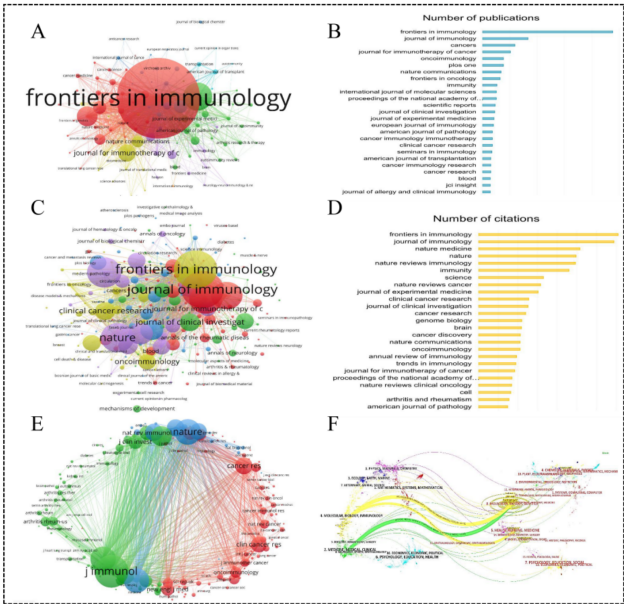


FIGURE 6. The influence of academic journals in the field of TLSs. (A) Journal bibliographic coupling analysis in TLSs research (B) The top 20 global academic journals for TLSs-related publications (C) The cited network of journals on TLSs research (D) The top 20 global academic journals for TLSs-related citations (E) Journal co-citation network visualization in TLSs research (F) Journal double-map overlay for knowledge flow analysis in TLSs Research

TABLE 6. Top 3 literatures by citation burst strength in the TLSs field

Rank	Corresponding/First Author	Journal	Burst Strength	Active Year Interval
1	Pitzalis C	Nat Rev Immunol	53.21	2015-2019
2	Silina K	Cancer Res	25.25	2019-2024
3	Mebius RE	Nat Rev Immunol	21.88	2004-2008

G. KEYWORD CO-OCCURRENCE AND EVOLUTION TREND

Keyword network and temporal evolution analysis present the historical reconstruction of the cognitive framework in the TLSs field from another dimension. In the co-occurrence network composed of 165 keywords with a frequency exceeding 25 times, tertiary lymphoid structures (total link strength 4033), b-cells (2914), and immunotherapy (2876) rank top three. The three core research themes — structure formation mechanism, B-cell immune regulation, and immunotherapy application — are thus clearly differentiated into interconnected but focused knowledge subdomains in the network topology [27]. The evolution trajectory presented by timeline analysis is more persuasive: phenotypic descriptive words represented by “expression”, “inflammation”, and “autoimmunity” dominated early research output, and then functional value-oriented words such as “prognosis”, “survival”, and “regulatory T cell” gradually took over, marking a paradigm shift in research concerns from “what TLSs are” to “what TLSs mean”. The temporal distribution of burst keywords fully records this transformation process: 18 words such as “germinal centers” burst intensively from 2001 to 2006, reflecting the characteristics of a wide exploration period; “tertiary lymphoid organs” has burst since 2016, reflecting researchers’ increasing recognition of the independent functional status of TLSs; “immunotherapy”

has continued to 2024 with the highest burst strength of 46.96 since 2021, not only far exceeding all other words in strength, but also extending to the retrieval end point in duration, strongly indicating that the cross-research of TLSs and immunotherapy has moved from germination to the mainstream, and is the frontier direction with the most explosive vitality and the most likely to produce significant clinical transformation value at present (Fig. 9).

IV. DISCUSSION
A. OVERALL RESEARCH SITUATION AND INTERNATIONAL PATTERN

The global academic output in the TLSs field saw its conceptual foundations laid in the 1990s (e.g., the landmark study by Kratz *et al.* in 1996), though the real research outbreak occurred around 2018. The direct incentive is the intensive publication of a number of landmark results in Nature series journals, and the emergence of high-impact research has a significant positive incentive effect on the overall research heat. From the perspective of national pattern, the United States firmly dominates the world with 804 articles and a total link strength of 17881; China ranks second with 535 articles, but the total citation link strength (12714) is significantly different from that of the United States, indicating that in addition to the rapid catch-up in quantity, Chinese scholars still have great room for improvement in research quality and international influence [21]. France is the most noteworthy “anomaly” in this pattern: with 229 articles ranking fourth, its total citation link strength (10794) surpasses Germany and approaches that of the United Kingdom. The Paris-centered collaboration cluster formed by Inserm and other institutions in the co-citation network profoundly reveals the multiplier effect of

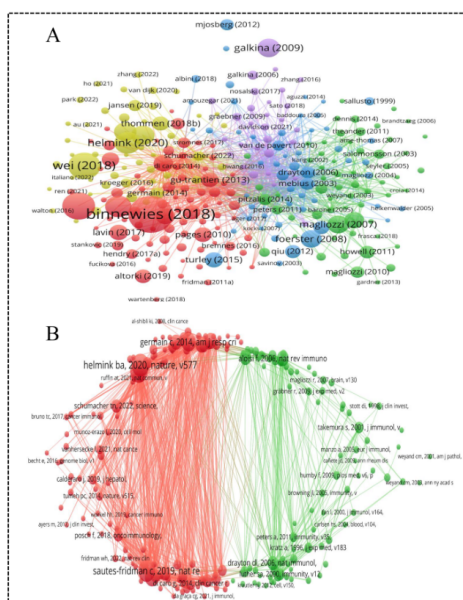


FIGURE 7. The analysis of relevant reference literature in the field. (A) Co-occurrence network of cited references in TLSs research (B) Mapping of the co-cited references related to this field

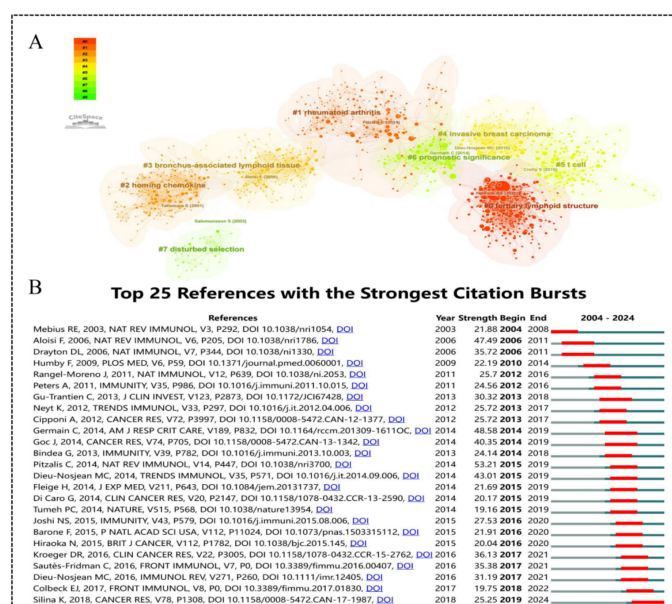


FIGURE 8. Cluster analysis and the outbreak citation analysis of references. (A) The evolution of research clusters in the TLSs field (B) The references that are cited in different periods of citation bursts. In the references, 'intensity' indicates the strength of the keyword's emergence, while 'year' corresponds to the point in time when the citation first appeared (as indicated by the dark green band). 'Start' and 'end' mark the beginning and end times of the outbreak cycle, with the red band highlighting the duration of the outbreak event

original basic research in knowledge dissemination [22]. This phenomenon suggests that the number of publications is not linearly related to academic influence, while total citation metrics are affected by the duration since publication, the sustained high co-citation strength of French and British research highlights the enduring impact of their foundational studies, and the accumulation of research quality is the real support for long-term international discourse power.

B. ANALYSIS OF RESEARCH HOTSPOTS IN THE TLSS FIELD

Keyword co-occurrence network and temporal evolution trajectory clearly present the historical reconstruction of the cognitive framework of TLSs research. Among the 165 keywords with a frequency exceeding 25 times, tertiary lymphoid structures (total link strength 4033), b-cells (2914), and immunotherapy (2876) rank top three, and the three themes form interconnected but focused knowledge subdomains in the network topology [27]. Timeline analysis shows a more persuasive paradigm shift: phenotypic descrip-

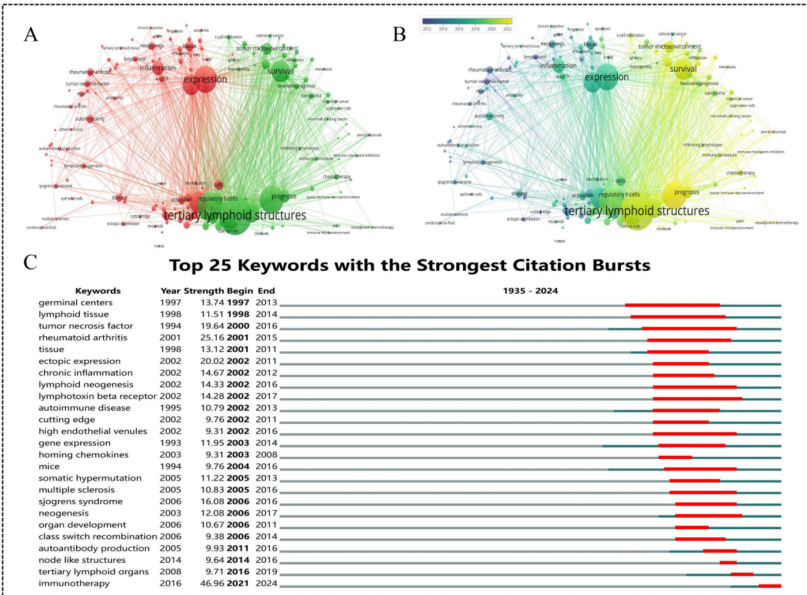


FIGURE 9. The analysis of keywords with a focus on TLSs. (A) The keywords co-occurrence network in the field of TLSs (B) A time series analysis of 165 high-frequency co-occurring keywords was conducted, where yellow indicates keywords that appeared later, and green represents those that appeared earlier (C) The top 25 burst-associated keywords in TLSs research. Red horizontal bars denote the duration of citation bursts, with bar length corresponding to the burst timespan

tive words dominated by “expression”, “inflammation”, and “autoimmunity” in the early stage are gradually replaced by functional value-oriented words such as “prognosis”, “survival”, and “regulatory T cell”, and research concerns shift from “what TLSs are” to “what TLSs mean”. This transformation is also reflected in the citation cluster structure: Cluster #2 (homing chemokines, average year 2000) is the earliest active research direction, and the recent parallel activity of Cluster #0 (tertiary lymphoid structures) and Cluster #5 (T cells, 2019) marks that this field has entered the in-depth stage of mechanistic analysis from phenotypic characterization [23,24]. The review by Pitzalis *et al.* [26] with the highest burst strength of 53.21 lasting for 5 years is exactly the key node to systematically establish the human pathological research paradigm of TLSs.

C. EVIDENCE FOR IMMUNOTHERAPY AS A FRONTIER DIRECTION

Multi-dimensional evidence points to a conclusion: cross-research between TLSs and immunotherapy has moved from germination to the mainstream. Keyword burst analysis shows that “immunotherapy” has continued to be active until the retrieval end point (2024) with the highest burst strength of 46.96 since 2021, its strength far exceeding all other words, and the duration also extends to the latest data node, making it the most explosive frontier direction at present [27]. Highly cited literature and citation burst analysis further provide specific knowledge anchors: three independent studies published in Nature in the same period of 2020 — Helmink *et al.* [14] (co-citation link strength 7320), Cabrita *et al.* (7213), and Sautès-Fridman *et al.* (7019) — jointly laid the field consensus that “TLSs maturity is positively correlated with tumor immunotherapy response”. The highly

co-cited structure of these three papers indicates that they have been repeatedly cited as the knowledge cornerstone. At the mechanistic level, the role of CXCL13/CXCL12/PD-L1 positive epithelial cell niche in the maintenance of TLSs revealed by Silina *et al.* [26] still maintains an active burst strength of 25.25 until 2024, providing a direct basis for targeted induction strategies of TLSs, and making it one of the most continuously attractive literatures currently.

D. RESEARCH LIMITATIONS

This study has several inherent limitations at the methodological level, which need to be carefully considered when interpreting the conclusions. First, the data source is limited to the Web of Science Core Collection. Although this database has recognized advantages in interdisciplinary coverage and citation tracking [20], there is still a certain risk of literature omission, especially non-English literature and some regional journal results are not included, leading to the possible underestimation of the actual contribution of some countries. Second, although the retrieval strategy is based on the MeSH vocabulary and includes multiple synonymous terms, the naming in the TLSs field is not yet fully unified, and different literatures use different terms such as “tertiary lymphoid structures” and “ectopic lymphoid-like structures”, making it difficult to completely eliminate the risk of missed detection. The inconsistency in terminology (e.g., ‘TLSs’, ‘ectopic lymphoid-like structures’, ‘tertiary lymphoid organs’) may affect the reproducibility of the search. Variations in indexing across different journals and databases could yield slightly different results, which is a fundamental limitation for any bibliometric study in an evolving field with non-standardized nomenclature. Third, the bibliometric method itself can only present the quan-

tative characteristics of research output and citation, and cannot capture the deep connotation of research quality; high citation strength is not necessarily equivalent to great academic contribution, and there are cases in highly cited achievements where methodologies are widely imitated but conclusions are controversial. The above limitations suggest that the conclusions of bibliometric analysis should be used as a reference for macro trend judgment, rather than the ultimate judgment of the domain pattern.

V. CONCLUSION

Bibliometric analysis systematically sorts out the global research evolution process in the field of tertiary lymphoid structures from 1935 to 2024. The results show that the publication volume and citation volume in this field show a continuous upward trend, especially the growth rate has accelerated significantly after 2018, reflecting that TLSs have become a core topic in immunopathological research. A total of 75 countries and regions around the world have participated in this field of research. The United States dominates the world in terms of publication volume and citation influence, and French institutions occupy an important hub position in the international cooperation network; China ranks second in the world in publication volume, but there is still room for improvement in citation quality and international cooperation depth. Keyword evolution analysis reveals a historical shift in research focus: early research focused on the phenotypic characteristics and formation mechanism of TLSs, and then gradually extended to its functional significance and prognostic value. The most explosive keyword at present is “immunotherapy”, indicating that cross-research between TLSs and immunotherapy has become the most active frontier direction in this field. Future research should, on the basis of deepening the analysis of TLSs formation mechanism, further explore the induction or inhibition strategies of TLSs as therapeutic targets in different disease contexts, and particularly strengthen cross-national and cross-institutional collaborative research to promote the in-depth development of TLSs basic research to clinical transformation and application.

AUTHOR CONTRIBUTIONS

Conceptualization – Can Yang, Haodong Guo and Xiaopeng Ma; methodology – Can Yang, Haodong Guo and Xiaopeng Ma; investigation – Can Yang, Haodong Guo and Xiaopeng Ma; writing-original draft preparation – Can Yang, Haodong Guo and Xiaopeng Ma. All authors have read and agreed to the published version of the manuscript.

CONFLICTS OF INTEREST

The authors declare no conflict of interest.

FUNDING

This research received no external funding.

ACKNOWLEDGEMENTS

Not applicable.

REFERENCES

- [1] A. Kratz, A. Campos-Neto, M. S. Hanson, and N. H. Ruddle, “Chronic inflammation caused by lymphotoxin is lymphoid neogenesis,” *J Exp Med*, vol. 183, no. 4, pp. 1461-1472, 1996, DOI: 10.1084/jem.183.4.1461.
- [2] C. Gago da Graça, L. G. M. van Baarsen, and R. E. Mebius, “Tertiary Lymphoid Structures: Diversity in Their Development, Composition, and Role,” *J Immunol*, vol. 206, no. 2, pp. 273-281, 2021, DOI: 10.4049/jimmunol.2000873.
- [3] Y. Sato, K. Silina, M. van den Broek, K. Hirahara, and M. Yanagita, “The roles of tertiary lymphoid structures in chronic diseases,” *Nat Rev Nephrol*, vol. 19, no. 8, pp. 525-537, 2023, DOI: 10.1038/s41581-023-00706-z.
- [4] H. M. Hsiao, W. Li, A. E. Gelman, A. S. Krupnick, and D. Kreisel, “The Role of Lymphoid Neogenesis in Allo-grafts,” *Am J Transplant*, vol. 16, no. 5, pp. 1079-1085, 2016, DOI: 10.1111/ajt.13645.
- [5] S. Salomonsson, M. V. Jonsson, K. Skarstein, K. A. Brokstad, P. Hjelmström, M. Wahren-Herlenius, and R. Jonsson, “Cellular basis of ectopic germinal center formation and autoantibody production in the target organ of patients with Sjögren’s syndrome,” *Arthritis Rheum*, vol. 48, no. 11, pp. 3187-3201, 2003, DOI: 10.1002/art.11311.
- [6] J. E. Moyron-Quiroz, J. Rangel-Moreno, K. Kusser, L. Hartson, F. Sprague, S. Goodrich, D. L. Woodland, F. E. Lund, and T. D. Randall, “Role of inducible bronchus associated lymphoid tissue (iBALT) in respiratory immunity,” *Nat Med*, vol. 10, no. 9, pp. 927-934, 2004, DOI: 10.1038/nm1091.
- [7] C. Yang, Y. X. Cai, Z. F. Wang, S. F. Tian, and Z. Q. Li, “Tertiary lymphoid structures in the central nervous system,” *Trends Mol Med*, vol. 31, no. 6, pp. 509-521, 2025, DOI: 10.1016/j.molmed.2024.10.014.
- [8] P. L. Kendall, G. Yu, E. J. Woodward, and J. W. Thomas, “Tertiary lymphoid structures in the pancreas promote selection of B lymphocytes in autoimmune diabetes,” *J Immunol*, vol. 178, no. 9, pp. 5643-5651, 2007, DOI: 10.4049/jimmunol.178.9.5643.
- [9] D. Lucchesi, C. Pitzalis, and M. Bombardieri, “EBV and other viruses as triggers of tertiary lymphoid structures in primary Sjögren’s syndrome,” *Expert Rev Clin Immunol*, vol. 10, no. 4, pp. 445-455, 2014, DOI: 10.1586/1744666x.2014.892417.
- [10] N. H. Ruddle, “Posttransplant Tertiary Lymphoid Organs,” *Transplantation*, vol. 108, no. 5, pp. 1090-1099, 2024, DOI: 10.1097/tp.0000000000004812.
- [11] M. Meylan, F. Petitprez, E. Becht, A. Bougouin, G. Pupier, A. Calvez, I. Giglioli, V. Verkarre, G. Lacroix, J. Verneau, C. M. Sun, P. Laurent-Puig, Y. A. Vano, R. Elaidi, A. Méjean, R. Sanchez-Salas, E. Barret, X. Cathelin-eau, S. Oudard, C. A. Reynaud, A. de Reyniès, C. Sautès-Fridman, and W. H. Fridman, “Tertiary lymphoid structures generate and propagate antitumor antibody-producing plasma cells in renal cell cancer,” *Immunity*, vol. 55, no. 3, pp. 527-541.e525, 2022, DOI: 10.1016/j.immuni.2022.02.001.
- [12] P. Singh, Z. Z. Coskun, C. Goode, A. Dean, L. Thompson-Snipes, and G. Darlington, “Lymphoid neogenesis and immune infiltration in aged liver,” *Hepatology*, vol. 47, no. 5, pp. 1680-1690, 2008, DOI: 10.1002/hep.22224.
- [13] J. G. Galletti, K. K. Scholand, C. M. Trujillo-Vargas, Z. Yu, O. Mauduit, V. Delcroix, H. P. Makarenkova, and C. S. de Paiva, “Ectopic lymphoid structures in the aged lacrimal glands,” *Clin Immunol*, vol. 248, p. 109251, 2023, DOI: 10.1016/j.clim.2023.109251.
- [14] B. A. Helmink, S. M. Reddy, J. Gao, S. Zhang, R. Basar, R. Thakur, et al., “B cells and tertiary lymphoid structures promote immunotherapy response,” *Nature*, vol. 577, no. 7791, pp. 549-555, 2020, DOI: 10.1038/s41586-019-1922-8.
- [15] A. T. Ruffin, A. R. Cillo, T. Tabib, A. Liu, S. Onkar, S. R. Kunning, C. Lampenfeld, H. I. Atiya, I. Abecassis, C. H. L. Kürten, Z. Qi, R. Soose, U. Duvvuri, S. Kim, S. Oesterrich, R. Lafyatis, L. G. Coffman, R. L. Ferris, D. A. A. Vignali, and T. C. Bruno, “B cell signatures and tertiary lymphoid structures contribute to outcome in head and neck squamous cell carcinoma,” *Nat Commun*, vol. 12, no. 1, p. 3349, 2021, DOI: 10.1038/s41467-021-23355-x.
- [16] M. C. Dieu-Nosjean, J. Goc, N. A. Giraldo, C. Sautès-Fridman, and W. H. Fridman, “Tertiary lymphoid structures in cancer and beyond,” *Trends Immunol*, vol. 35, no. 11, pp. 571-580, 2014, DOI: 10.1016/j.it.2014.09.006.
- [17] B. Serafini, B. Rosicarelli, D. Franciotta, R. Magliozzi, R. Reynolds,

- P. Cinque, et al, "Dysregulated Epstein-Barr virus infection in the multiple sclerosis brain," *J Exp Med*, vol. 204, no. 12, pp. 2899-2912, 2007, DOI: 10.1084/jem.20071030.
- [18] A. Manzo, M. Bombardieri, F. Humby, and C. Pitzalis, "Secondary and ectopic lymphoid tissue responses in rheumatoid arthritis: from inflammation to autoimmunity and tissue damage/remodeling," *Immunol Rev*, vol. 233, no. 1, pp. 267-285, 2010, DOI: 10.1111/j.0105-2896.2009.00861.x.
- [19] O. Thaunat, "Pathophysiologic significance of B-cell clusters in chronically rejected grafts," *Transplantation*, vol. 92, no. 2, pp. 121-126, 2011, DOI: 10.1097/tp.0b013e31821f74fe.
- [20] G. González-Alcaide, "Bibliometric studies outside the information science and library science field: un-containable or uncontrollable?" *Scientometrics*, vol. 126, no. 5, pp. 6837-6870, 2021, DOI: 10.1007/s11192-021-04061-3.
- [21] Y. Liu, S. Wong, Y. Han, X. Li, L. Yuan, M. Xiong, L. Zhao, H. Xu, T. Wang, and Q. Liang, "Global Research Trends in Adipose Stem Cell Tissue Engineering: A Scientometric Research," *Tissue Eng Part C Methods*, vol. 29, no. 9, pp. 505-525, 2023, DOI: 10.1089/ten.tec.2023.0152.
- [22] H. Q. Niu, Y. Yang, B. B. Wang, W. T. Xiao, H. R. Wu, Y. D. Liu, et al, "A global bibliometric and visualized analysis in the status and trends of corneal tissue engineering research from 1991 to 2021," *J Tissue Eng*, vol. 13, p. 20417314221138188, 2022, DOI: 10.1177/20417314221138188.
- [23] X. Yang, X. Wang, X. Li, D. Gu, C. Liang, K. Li, G. Zhang, and J. Zhong, "Exploring emerging IoT technologies in smart health research: a knowledge graph analysis," *BMC Med Inform Decis Mak*, vol. 20, no. 1, p. 260, 2020, DOI: 10.1186/s12911-020-01278-9.
- [24] L. Zhang, H. Zheng, S. T. Jiang, Y. G. Liu, T. Zhang, J. W. Zhang, et al, "Worldwide research trends on tumor burden and immunotherapy: a bibliometric analysis," *Int J Surg*, vol. 110, no. 4, pp. 1699-1710, 2024, DOI: 10.1097/j.s9.0000000000001022.
- [25] C. Pitzalis, G. W. Jones, M. Bombardieri, and S. A. Jones, "Ectopic lymphoid-like structures in infection, cancer and autoimmunity," *Nat Rev Immunol*, vol. 14, no. 7, pp. 447-462, 2014, DOI: 10.1038/nri3700.
- [26] K. Silina, A. Soltermann, F. M. Attar, R. Casanova, Z. M. Uckeley, H. Thut, et al, "Germinal Centers Determine the Prognostic Relevance of Tertiary Lymphoid Structures and Are Impaired by Corticosteroids in Lung Squamous Cell Carcinoma," *Cancer Res*, vol. 78, no. 5, pp. 1308-1320, 2018, DOI: 10.1158/0008-5472.can-17-1987.
- [27] S. Xiong, K. Liu, F. Yang, Y. Dong, H. Zhang, P. Wu, et al, "Global research trends on inflammatory bowel diseases and colorectal cancer: A bibliometric and visualized study from 2012 to 2021," *Front Oncol*, vol. 12, p. 943294, 2022, DOI: 10.3389/fonc.2022.943294.