Iğdır Üniversitesi Fen Bilimleri Enstitüsü Dergisi, 14(2), 888-893, 2024
Journal of the Institute of Science and Technology, 14(2), 888-893, 2024

ISSN: 2146-0574, eISSN: 2536-4618

Moleculer Biology and Genetic **DOI: 10.21597/jist.1447517**

Research Article

Received: 07.03.2024 **Accepted: 17.04.2024**

Atıf İçin: Aslan, E.S., Abuaisha, A., Emiroğlu, S., Doğan, M.A. (2024). Meme Kanserli Hastalarda Anti-Nükleer Antikorlar ve Anti-Çekilebilir Nükleer Antijen Seviyelerinin Değerlendirilmesi. *Iğdır Üniversitesi Fen Bilimleri Enstitüsü Dergisi*, 14(2), 888-893.

To Cite: Aslan, E.S., Abuaisha, A., Emiroğlu, S. & Doğan, M.A. (2024). Assessment of Anti-Nuclear Antibodies and Anti-Extractable Nuclear Antigen Levels in Breast Cancer Patients. *Journal of the Institute of Science and Technology*, 14(2), 888-893.

Assessment of Anti-Nuclear Antibodies and Anti-Extractable Nuclear Antigen Levels in Breast Cancer Patients

Elif Sibel ASLAN[1], Asmaa ABUAISHA[2], Selman EMİROĞLU[3], Mehmet Ali DOĞAN[4]

Öne Çıkanlar:
- Lümenal meme kanseri hastalarında ANA'ların yaygınlığı
- Tümör alt tiplerinde anti-nükleer antikorların farklı konsantrasyonları
- Otoantikor seviyeleri ile klinik özellikler arasındaki ilişki.

Anahtar Kelimeler:
- Anti-nükleer antikorlar (ANAs)
- Otoantikorlar
- Meme Kanseri
- Anti-Çekilebilir Nükleer Antijenler (ENAs)

ÖZET:

Anti-nükleer antikorlar (ANAs), hücre çekirdek içeriğine karşı üretilen otoantikorlardır ve sistemik otoimmün hastalıkların biyobelirteçleri olarak kullanılırlar. İnflamasyon, apoptoz ve hücre nekrozu, meme kanseri ile ilişkili olarak ortaya çıkan hücresel sonuçlardır ve bu duruma karşı otoantikorlar üretilebilir. Bu çalışmada, meme kanserinde ANA'ların ve anti-ekstrakte edilebilir nükleer antijenlerin (anti-ENAs) varlığını değerlendirmeyi amaçladık. Toplam 33 lümenal A ve lümenal B meme kanseri hastası, ANA'ların ve anti-ENAs'ın varlığı açısından değerlendirildi. Tüm hastalar, testlerden en az 6 ay önce hormon terapisi almışlardır. Hastalar, insan epitel tip 2 (HEp-2) hücrelerinde endirekt immünoflorasansta ANA'lar için taranmıştır. U1-snRNP, snRNP/Sm, SmD1, dsDNA, SS-A/Ro 60, SS-A/Ro 52, SS-B/La antikor konsantrasyonlarını belirlemek için AESKUBLOTS® ANA-17 comp kiti kullanıldı. Onbeş hastanın (%45.5) lümenal A ve 18 hastanın (%54.5) lümenal B olduğu belirlendi. Ortalama yaş 57, tümör boyutu ise 25 tir. Hastaların %57.6'sında I veya II derece, %42.4'ünde III derece tümör vardı ve üç hasta ANA testi pozitifti. Pozitif ANA testi olan tüm hastalar lümenal A meme kanserliydi ve I veya II derece tümöre sahip olup pozitif lenf noduna sahipti; ancak patolojik tümör evresi değişkenlik gösteriyordu. ANA pozitifliği ile moleküler alt tip, yaş, vücut kitle indeksi (VKİ), derece, tümör evresi veya lenf nodu tutulumu arasında istatistiksel olarak anlamlı bir ilişki bulunamamıştır. Ayrıca, anti-U1-snRNP ve anti-dsDNA'nın Ki-67 ile negatif, anti-snRNP/Sm ve anti-SS-A/Ro 52'nin ise pozitif bir korelasyonu bulunmuştur. Lümenal A ile karşılaştırıldığında, anti-U1-snRNP ve anti-snRNP/Sm konsantrasyonları lümenal B tümörlerinde istatistiksel olarak önemli ölçüde düşük bulunmuştur (sırasıyla p= 0.015 ve 0.016). Yüksek dereceli tümöre sahip hastalar, düşük anti-snRNP/Sm konsantrasyonları gösterirken, lenf nodu metastazına sahip olanlar yüksek anti-U1-snRNP konsantrasyonları sergilemiştir (sırasıyla p=0.027 ve 0.031). ANA pozitifliği, lümenal A meme kanseri hastalarında lümenal B'ye kıyasla daha yaygın bulunmuştur. Anti-U1-snRNP ve anti-snRNP/Sm konsantrasyonları lümenal B'de daha düşüktür. Ayrıca, yüksek dereceli tümöre sahip hastalar düşük anti-snRNP/Sm konsantrasyonları gösterirken, lenf nodu metastazı olanlar yüksek anti-U1-snRNP konsantrasyonları sergilemiştir.

Assessment of Anti-Nuclear Antibodies and Anti-Extractable Nuclear Antigen Levels in Breast Cancer Patients

Highlights:
- Prevalence of ANAs in Luminal Breast Cancer Patients
- Differential Concentrations of Anti-nuclear Antibodies in Tumor Subtypes
- Association between Autoantibody Levels and Clinical Characteristics.

Keywords:
- Anti-nuclear antibodies (ANAs)
- Autoantibodies
- Breast Cancer
- Anti-Extractable Nuclear Antigens (ENAs).

ABSTRACT:

Anti-nuclear antibodies (ANAs) are autoantibodies synthesized in response to the cell nucleus contents and use as biomarkers of systemic autoimmune diseases. Inflammation, apoptosis and necrosis of the cells are consequences that accompany breast cancer against which autoantibodies will be produced. In this study, we aimed to evaluate the presence of ANAs and anti-extractable nuclear antigens (anti-ENAs) in breast cancer. A total of 33 luminal A and luminal B breast cancer patients were assessed for presence of ANAs and anti-ENAs. All the patients had received hormone therapy at least for 6 months before the tests. Patients were screened to ANAs by indirect immunofluorescence on human epithelial type 2 (HEp-2) cells. AESKUBLOTS® ANA-17 comp kit was used to identify the concentrations of U1-snRNP, snRNP/Sm, SmD1, dsDNA, SS-A/Ro 60, SS-A/Ro 52, SS-B/La antibodies. Fifteen (45.5%) patients were luminal A and 18 (54.5%) patients were luminal B. The median of age was 57 and the median of tumor size was 25. 19 (57.6%) patients had grade I or II and 14 (42.4%) had grade III. 3 patients had ANAs test positive. All the patients who had positive ANA test were luminal A breast cancer and had grade I or II tumors and positive lymph node, whereas, pathological tumor stage were varied. No statistically significant association was found between ANAs positivity and molecular subtype, age, body mass index (BMI), grade, tumor stage or lymph node involvement. Moreover, there were negative correlations between the anti-U1-snRNP and anti-dsDNA with Ki-67 and a correlation between anti-snRNP/Sm and anti-SS-A/Ro 52 was found. Comparing with luminal A, anti-U1-snRNP and anti-snRNP/Sm concentrations were statistically significantly lower in luminal B tumors (p= 0.015 and 0.016 respectively). Patients who had high grade tumors showed low concentrations of anti-snRNP/Sm (p=0.027), whereas patients who had lymph node metastasis showed high concentrations of anti-U1-snRNP (p=0.031). ANAs positivity was more common in luminal A breast cancer patients compared with luminal B. Anti-U1-snRNP and anti-snRNP/Sm concentrations were lower in luminal B. Moreover, patients who had high grade tumors showed low concentrations of anti-snRNP/Sm, whereas those who had lymph node metastasis showed high concentrations of anti-U1-snRNP.

[1] Elif Sibel ASLAN (**Orcid ID:** 0000-0002-3081-9004), Department of Molecular Biology and Genetics, Biruni University, Istanbul, Türkiye

[2]Asmaa ABUAISHA (**Orcid ID:** 0000-0003-2869-6620), Department of Genetics, Institute of Health Sciences, Istanbul University, Istanbul, Türkiye

[3]Selman Emiroğlu (**Orcid ID:** 0000-0001-9333-6926), Breast Surgery Division, General Surgery Department, Istanbul Faculty of Medicine, Istanbul University, Istanbul, Türkiye

[4]Mehmet Ali Doğan. (**Orcid ID:** 0000-0003-3324-3028), Department of Immunology and Medical Microbiology, Basic Medical Sciences, Istanbul Faculty of Medicine, Istanbul University, Istanbul, Türkiye

***Corresponding Author:** Elif Sibel ASLAN, e-mail: easlan@biruni.edu.tr

Ethics Committee Approval: This study has been approved by Ethical committee of Istanbul University's Istanbul Faculty of Medicine (file no. 2023/195).

INTRODUCTION

Anti-nuclear antibodies (ANAs) are autoantibodies synthesized in response to the macromolecular components (DNA, RNA, protein and complexes of them) of the cell nucleus. As a result to apoptosis or cell necrosis, cells contents appear in the blood to which ANAs may be synthesized (Ling and Murali, 2019). ANAs is used as biomarkers of systemic autoimmune diseases and it may represent a pre-autoimmunity state such in systemic lupus or rheumatoid arthritis (Choi and Costenbader, 2022). Not only in the autoimmune diseases, but ANAs may also be a marker of some other diseases (Choi and Costenbader, 2022). A positive ANAs can be seen in different infections and malignancies including lung, breast, head and neck cancer (Admou B., et al. 2019; Morimoto, K. et al. 2019; Otsuka T. et al. 2019; Fernández Madrid, F., 2015). Different studies have showed that cancers may trigger the production of autoantibodies that may be used for early diagnosis (Turgutalp, K. et al. 2013; Sexauer, and Gray 2022; Lacombe, J. et al. 2014). Other studies showed that autoantibodies have role in angiogenesis and cancer prognosis (Lacombe, J. et al. 2014; Liu, B.C., et al. 2012) and might be used as targets for cancer drugs (Hong, C. Q. et al. 2021).

Moreover, ANAs may be helpful during the early diagnosis of breast cancer. Inflammation and cell necrosis are consequences that accompany breast cancer. Tumor cell death is a source of antigen stimulation that leads to ANAs production in cancers. Although, it may represent an immune response to stop tumor spreading (Chapman, C., et al. 2007). Nisihara et al. 2018 investigated the prevalence of ANAs in breast cancer patients and showed its association with tumor characteristics. 91 healthy individuals, 72 patients had malignant lesions and 19 patients had benign lesions were included in their study. 44.4% of the patients with malignant tumors and 15.7% of those with benign lesions and 5.4% of the controls had positive ANAs. They showed that there was a high prevalence of ANAs positivity in breast cancer patients with a negative association with hormonal receptor positivity. In addition, (Mohammed et al. 2015), investigated the concentration of ANAs and anti-dsDNA in 35 newly diagnosed breast cancer patients and compared to 18 age- and sex-matched control subjects. They reported that ANAs concentration was significantly high in breast cancer patients, whereas anti-dsDNA was not.

In this study, we aimed to evaluate ANA concentrations and the concentration of U1-snRNP, snRNP/Sm, SmD1, dsDNA, SS-A/Ro 60, SS-A/Ro 52, SS-B/La in Luminal subtypes of breast cancer patients.

MATERIALS AND METHODS

This study has been approved by Ethical commitee of Istanbul University's Faculty of Medicine (file no. 2023/195). All patients were informed about the study's purpose, content, and intervention, and their oral and written consent was obtained. A total of 33 luminal subtypes of breast cancer were evaluated for the clinicopathological characteristics and for the presence of ANAs and anti-extractable nuclear antigens (ENAs) (anti-U1-snRNP, anti-snRNP/Sm, anti-SmD1, anti-dsDNA, anti-SS-A/Ro60, anti-SS-A/Ro52 and anti-SS-B/La). All the patients had received hormone therapy at least for 6 months before the tests. Two (2) mL of venous blood were drawn and serum samples were preserved for ANAs and anti-ENAs panel tests. All the samples were investigated for ANAs by indirect immunofluorescence on human epithelial type 2 (HEp-2) cells, using the commercially available kit ANA HEp-2 (AESKUSLIDES–ANA-HEp-2, Germany), as recommended by the manufacturer. A titre of 1:80 or higher was considered to indicate positive result. AESKUBLOTS®ANA-17comp kit was used to evaluate anti-ENAs.ANA-17comp is a membrane based enzyme immunoassay for qualitative detection of IgG antibodies against nuclear and cytoplasmic antigens in human serum. For instance, SS-A/Ro and

SS-B/La antibodies are associated with the sjogren's (Sjögren's) syndrome (SS) and anti-dsDNA, anti-SM, anti-histone and anti-nucleosomes are associated with Systemic Lupus, Erythematosus systemic lupus erythematosus (SLE). Anti-RNP antibodies with mixed connective tissue diseases (MCTD) and SLE with mixed connective tissue diseases (MCTD) and SLE.

RESULTS AND DISCUSSION

Patients and Tumor General Characteristics

Thirty-three luminal A and luminal B breast cancer patients were included in this study. The median of age was 57 and the median of body mass index (BMI) was 29. Fifteen (45.5%) patients were luminal A and eighteen (54.5%) patients were luminal B. The median of Ki-67 was twenty (20%) and the median of tumor size was twenty five mm. Nineteen (57.6%) patients had grade I or II and fourteen (42.4%) had grade III. Nine (27.3%) patients had pathological tumor stage I and twenty four (72.7%) had stage II or III. Lymph node positivity were observed in seventeen (51.5) patients (Table 2 and Table 3).

ANAs Positivity in Patients According to the Cinicopathological Characteristics

Three patients had ANAs positive. All the patients who had a positive ANAs were luminal A and had grade I or II tumors with lymph node metastasis, whereas, pathological tumor stage were varied in those patients. No statistically significant association was found between ANAs positivity and molecular subtype, age, BMI, grade, tumor stage or lymph node involvement (Table 1).

Table 1. ANA positivity rates in patients according to clinicopathological characteristics

	ANA		
Variables	**Positive**	**Negative**	***p*-Value[a]**
All, n(%)	**3(9.1)**	**30(90.9)**	
Age, n(%)			0.999
<60	2(10)	18(90)	
≥60	1(7.7)	12(92.3)	
BMI, n(%)			0.999
<30	2(11.1)	16(88.9)	
≥30	1(6.7)	14(93.3)	
Grade, n(%)			0.244
I-II	3(15.8)	16(84.2)	
III	0(0)	14(100)	
pT stage, n(%)			0.999
I	1(11.1)	8(88.9)	
II-III	2(8.3)	22(91.7)	
pN involvement, n(%)			0.227
N(-)	0(0)	16(100)	
N(+)	3(17.6)	14(82.4)	
Molecular subtype, n(%)			0.083
Luminal A	3(20)	12(80)	
Luminal B	0(0)	18(100)	

p>0.05, Fisher's Exact Test. **BMI**: body mass index, **pT stage**: pathological tumor stage. **pN**: Pathological lymph node

Comparison Between Different Variables of Patients

There was no correlation between the different variables of the patients during these comparsions, whereas, there was a negative correlation between the anti-U1-snRNP and anti-dsDNA with Ki-67. Additionally, there was a correlation between anti-snRNP/Sm and anti-SS-A/Ro 52 concentrations (Table 2).

Table 2. Median, interquartile range(IQR) and Correlations

No.	Variables	Median(IQR)	1	2	3	4	5	6	7	8	9	10
1	**Age (year)**	57(46.50-65.5)	NA									
2	**Ki-67 (%)**	20(10-30)	-0.272									
3	**Tumor size (mm)**	25(18-37.5)	-0.154	0.247								
4	**BMI**	29.1(25.6-32.6)	**0.592****	-0.114	-0.048							
5	**U1-snRNP**	0.30(0.15-0.40)	0.022	**-0.380***	-0.099	0.067						
6	**snRNP/Sm**	0.30(0.10-0.50)	-0.056	-0.229	-0.169	-0.186	0.085					
7	**SmD1**	0.30(0.20-0.50)	0.028	-0.282	-0.052	-0.102	0.127	0.243				
8	**dsDNA**	0.40(0.20-0.50)	0.091	**-0.434***	-0.095	0.020	0.329	-0.081	-0.024			
9	**SS-A/Ro 60**	0.30(0.20-0.45)	0.117	-0.123	-0.017	0.279	0.018	-0.051	0.278	0.042		
10	**SS-A/Ro 52**	0.40(0.20-0.40)	0.192	-0.111	0.194	0.145	0.059	**0.363***	0.106	0.048	0.222	
11	**SS-B/La**	0.30(0.20-0.45)	0.022	0.144	-0.216	0.062	0.048	0.335	0.023	0.132	0.183	0.319

p<0.01, *p<0.05, Spearman's correlation test, **NA:Not available

ENAs Panel According to Tumor Characteristics

Table 3. ENA panel according to tumor characteristics

	All	U1-snRNP	snRNP/Sm	SmD1	dsDNA	SS-A/Ro 60	SS-A/Ro 52	SS-B/La
Variables	**n(%)**	**Median(IQR)**	**Median(IQR)**	**Median(IQR)**	**Median(IQR)**	**Median(IQR)**	**Median(IQR)**	**Median(IQR)**
Grade								
I-II	19(57.6)	0.30(0.20-0.50)	0.40(0.30-0.70)	0.40(0.20-0.60)	0.40(0.20-0.50)	0.30(0.20-0.40)	0.30(0.20-0.40)	0.30(0.20-0.50)
III	14(42.4)	0.25(0.10-0.40)	0.20(0.10-0.33)	0.25(0.10-0.43)	0.30(0.10-0.50)	0.30(0.20-0.50)	0.40(0.30-0.43)	0.30(0.18-0.43)
***p*-Value[a]**		0.426	**0.027***	0.055	0.311	0.467	0.401	0.698
pT stage								
I	9(27.3)	0.40(0.20-0.45)	0.30(0.15-0.50)	0.30(0.15-0.40)	0.50(0.30-0.50)	0.30(0.20-0.35)	0.30(0.15-0.40)	0.30(0.20-0.60)
II-III	24(72.7)	0.25(0.10-0.38)	0.30(0.10-0.65)	0.35(0.20-0.68)	0.40(0.20-0.50)	0.30(0.20-0.50)	0.40(0.30-0.48)	0.30(0.13-0.40)
***p*-Value[a]**		0.242	0.743	0.390	0.437	0.301	0.165	0.400
pN stage								
N(-)	16(48.5)	0.20(0.10-0.30)	0.30(0.20-0.65)	0.25(0.10-0.40)	0.30(0.20-0.48)	0.35(0.30-0.50)	0.40(0.30-0.48)	0.30(0.20-0.48)
N(+)	17(51.5)	0.30(0.20-0.50)	0.30(0.10-0.50)	0.40(0.20-0.55)	0.50(0.25-0.60)	0.30(0.20-0.35)	0.30(0.20-0.40)	0.30(0.15-0.45)
***p*-Value[a]**		**0.031***	0.422	0.126	0.113	0.053	0.555	0.913
Molecular subtype								
Luminal-A	15(45.5)	0.30(0.20-0.60)	0.50(0.30-0.70)	0.40(0.20-0.60)	0.50(0.20-0.60)	0.30(0.20-0.40)	0.40(0.20-0.70)	0.20(0.10-0.50)
Luminal-B	18(54.5)	0.20(0.10-0.33)	0.20(0.10-0.33)	0.25(0.10-0.43)	0.35(0.18-0.50)	0.30(0.20-0.50)	0.35(0.20-0.40)	0.30(0.20-0.43)
***p*-Value[a]**		**0.015***	**0.016***	0.138	0.124	0.711	0.344	0.409

Comparing with luminal A, anti-U1-snRNP and anti-snRNP/Sm concentrations were statistically significantly lower in luminal B tumors (p= 0.015 and 0.016 respectively). Patients who had high grade tumors showed low concentrations of anti-snRNP/Sm (p=0.027), whereas patients who had lymph node metastasis showed high concentrations of anti-U1-snRNP (p=0.031).

DISCUSSION

ANAs are autoantibodies against the cell nucleus components. Inflammation, apoptosis and necrosis of the cells are consequences that accompany breast cancer. In this study, thirty three 33 luminal A and luminal B breast cancer patients were been assessed for presence of ANAs and anti-ENAs. Three 3 patients were ANAs positive who had luminal A and had grade I or II tumors and lymph node metastasis. Comparing with luminal A, patients with anti-U1-snRNP and anti-snRNP/Sm concentrations were lower levels in luminal B patients. Patients who had high grade tumors showed low concentrations of anti-snRNP/Sm, whereas patients who had lymph node metastasis showed high concentrations of anti-U1-snRNP.

Autoantibodies found in a cancer patient may be classified into two categories; (a) specific antibodies to antigens which are not associated directly with the tumor such as ANA and (b) antibodies against specific tumor antigens (tumour-associated antigens) such as tumor suppression proteins, oncoproteins, onconeural antigens (Nisihara et al. 2018). Immune response function is against tumor-associated antigens to remove pre-cancerous lesions within during the early carcinogenesis. Thus, ANAs may have a protective role against tumor spread (Tan, H. T. et al. 2009; Erkanli, A. et al. 2009).

Compering to other studies, Nisihara et al. 2018 showed that there was a high prevalence of ANAs positivity in breast cancer patients with a negative association with hormone receptors positivity. Moreover, Mohammed et al 2015, investigated the concentration of ANAs and anti-dsDNA in 35 newly diagnosed breast cancer patients compared to 18 control subjects. They found that ANA concentration was significantly increased in the patients irrespective of the grade or stage, whereas anti-dsDNA had no significant difference between the patients and controls. (Shiel and Jason 1989), reported that 2.9% of patients who had positive ANA had neoplasia. As the number of the studies related to ANA in breast cancer is so limited we were not able to discuss this topic more (Cleaton and Bateman 2020). Further studies are needed to be done for a better understanding of the protective or may be the diagnostic role of ANAs in breast cancer.

CONCLUSION

ANAs positivity was more prevalent in luminal A breast cancer patients compared with luminal B. No statistically significant association was found between ANAs positivity and molecular subtype, age, BMI, grade, tumor stage or lymph node involvement. There was a negative correlation found between the levels of anti-U1-snRNP and anti-dsDNA with Ki-67 expression was found. Additionally, there was a correlation between anti-snRNP/Sm and anti-SS-A/Ro 52 concentrations.

ACKNOWLEDGEMENTS

Authors thanks to Atilla Bozdoğan for his statistical support.

Conflict of Interest

The article authors declare that there is no conflict of interest between them.

Author's Contributions

The authors declare that they have contributed equally to the article.

REFERENCES

Admou, B., Eddehbi, F. E., Elmoumou, L., Elmojadili, S., Salami, A., Oujidi, M., ... & Hazime, R. (2022). Anti-double stranded DNA antibodies: A rational diagnostic approach in limited-resource settings. *Practical Laboratory Medicine*, 31, e00285.

Chapman, C., Murray, A., Chakrabarti, J., Thorpe, A., Woolston, C., Sahin, U., ... & Robertson, J. (2007). Autoantibodies in breast cancer: their use as an aid to early diagnosis. *Annals of oncology*, 18(5), 868-873.

Choi, M.Y., Costenbader, K.H. Understanding the concept of pre-clinical autoimmunity: prediction and prevention of systemic lupus erythematosus: identifying risk factors and developing strategies against disease development. *Frontiers in Immunology*, 2022, 13: 890522.

Cleaton, N., & Bateman, J. (2020). P92 Referral with a positive antinuclear antibody? Lessons from prospective triage and diagnosis in a single-centre service evaluation of 1,547 new rheumatology referrals. *Rheumatology*, 59(Supplement_2), keaa111-090.

Erkanli, A., Taylor, D. D., Dean, D., Eksir, F., Egger, D., Geyer, J., ... & Roden, R. B. (2006). Application of Bayesian modeling of autologous antibody responses against ovarian tumor-associated antigens to cancer detection. *Cancer research*, 66(3), 1792-1798.

Fernández Madrid, F., Maroun, M. C., Olivero, O. A., Long, M., Stark, A., Grossman, L. I., ... & Peebles, C. (2015). Autoantibodies in breast cancer sera are not epiphenomena and may participate in carcinogenesis. *BMC cancer*, 15(1), 1-15.

Hong, C. Q., Weng, X. F., Huang, X. C., Chu, L. Y., Wei, L. F., Lin, Y. W., ... & Peng, Y. H. (2021). A panel of tumor-associated autoantibodies for the detection of early-stage breast cancer. *Journal of Cancer*, 12(9), 2747.

Lacombe, J., Mangé, A., & Solassol, J. (2014). Use of autoantibodies to detect the onset of breast cancer. *Journal of immunology research*, 2014.

Ling, M.; Murali, M. Antinuclear antibody tests. *Clinics in Laboratory Medicine*, 2019, 39.4: 513-524.

Liu, B. C., DiJohnson, D. A., & O'Rourke, D. J. (2012). Antibody profiling with protein antigen microarrays in early stage cancer. *Expert opinion on medical diagnostics*, 6(3), 187-196.

Mohammed, M. E. A., & Abdelhafiz, K. (2015). Autoantibodies in the sera of breast cancer patients: antinuclear and anti-double stranded DNA antibodies as example. *Journal of Cancer Research and Therapeutics*, 11(2), 341-344.

Morimoto, K., Yamada, T., Nakamura, R., Katayama, Y., Tanaka, S., Takumi, C., ... & Takayama, K. (2020). Impact of preexisting antinuclear antibodies on combined immunotherapy and chemotherapy in advanced non-small cell lung cancer patients. *Medical Oncology*, 37, 1-7.

Nisihara, R., Machoski, M. C. C., Neppel, A., Maestri, C. A., Messias-Reason, I., & Skare, T. L. (2018). Anti-nuclear antibodies in patients with breast cancer. Clinical & Experimental Immunology, 193(2), 178-182.

Otsuka, T., Hashii, Y., Murayama, S., Ishizuka, Y., Kojitani, Y., Nishio, M., & Kudo, T. (2023). Checkpoint inhibitor-induced gastritis followed by delayed severe hepatitis in a patient with lung metastases of head and neck squamous cell carcinoma: a case report. *Frontiers in Oncology*, 13, 1164236

Sexauer, D., Gray, E., & Zaenker, P. (2022). Tumour-associated autoantibodies as prognostic cancer biomarkers-a review. *Autoimmunity reviews*, 21(4), 103041.

Shiel Jr, W. C., & Jason, M. (1989). The diagnostic associations of patients with antinuclear antibodies referred to a community rheumatologist. *The Journal of Rheumatology*, 16(6), 782-785.

Tan, H. T., Low, J., Lim, S. G., & Chung, M. C. (2009). Serum autoantibodies as biomarkers for early cancer detection. *The FEBS journal*, 276(23), 6880-6904.

Turgutalp, K., Oğuz Gök, E., Akça, M., & Kıyım, A. A. (2013). Anti-dsDNA positivity in a patient with prostate cancer and acute kidney ınjury: a case report. *Türk Nefroloji Diyaliz ve Transplantasyon Dergisi,* 22(2), 221-223.