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BIOMARKERS OF OXIDATIVE STRESS AND GENOTOXICITY WITH T-CELL IMMUNE PROFILING IN KIDNEY CANCER PATIENTS

Nida Nisar[1], Muhammad Saqib Khalil[*2], Muhammad Ateeq[3], Hafsa[4], Naila Gulfam[5], Sulha Syed[6], Ihtisham Ul Haq[7]

[1,*2,3,4,6,7] *Sarhad Institute of Allied Health Sciences, Sarhad University of Science and Information Technology*
[5]*Jinnah College for Women, University of Peshawar*

[*2]saqib.siahs@suit.edu.pk

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Abstract

Kidney cancer is also called as renal cancer. It is a type of cancer, which occurs in the kidneys. It is believed that certain inflammatory responses have anti-tumorigenic effect. Cytotoxic T-lymphocytes, including CD8+ T-cells and CD4+ T-cells have potent anti-tumor activity. The study aimed to investigate the significance of T-lymphocyte cells in patients who have been diagnosed with kidney cancer. Twenty milliliter blood was collected under aseptic condition from 30 kidney cancer patients and 30 healthy individuals by vein puncture and transported to Alpha Genomic Laboratory, Islamabad for flow cytometric, comet assay analysis and evaluation of oxidative stress markers including MDA, SOD and GSH. Data of all the biochemical parameters were compiled in Microsoft Excel and analyzed statistically. Results showed that the CD4+ T-cells were higher in kidney cancer patients of both age groups (8.23±3.88 cells/µL; 12.06±4.71 cells/µL). The CD8+ T lymphocytes counts were also elevated (20.13±2.56; 20.11±3.33 cells/µL) in kidney cancer patients. Furthermore, the CD14+ T-cell counts were also higher (17.53±2.42 cells/µL; 19.75±3.96 cells/µL) in affected patients of both age groups. The DNA length in case of kidney cancer patients was higher than the control group. Similarly, the MDA and SOD concentration was also higher in kidney cancer patients in both cases. Whereas, the GSH concentration was lower (52.63±3.41 Nmol-mL; 55.65±4.44 Nmol-mL) in affected patients when compared with the healthy people (61.44±1.80 Nmol-mL; 67.17±2.86 Nmol-mL). Hence, it concluded that Malondialdehyde, superoxide dismutase and reduced glutathione are important biomarker that can be used to measure and monitor oxidative stress in cancer patients.

INTRODUCTION

Kidney cancer, also known as renal cancer, is a type of cancer that develops from the tissues of the kidneys (Alzubaidi et al., 2022). Kidney cancer is often characterized by the formation of an abdominal mass, hematuria (blood in the urine), and back or flank pain (Vasudev et al., 2020). Other symptoms include fatigue, unexplained weight loss, and high-grade fever. In its advanced form, kidney cancer can spread to other parts of the body, including the lungs and brain (Bahadoram et al., 2022). However, in its early stage, kidney cancer is often symptom-free and cannot be easily diagnosed by physical examination (Krishna et al., 2020).

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Kidney cancer consists of multiple histological types which include Wilms tumor and Transitional Cell Carcinoma and Renal Cell Carcinoma according to research by Kamal and Ramzan (2023). The most common type of kidney cancer is RCC which constitutes about 80 percent of all kidney cancer cases while TCC exists as a less common type according to research by Moore et al. (2020). Researchers have linked multiple risk factors to both RCC and TCC which include previous bladder cancer cases and hypertension and smoking and obesity and family history and chemical and analgesic exposure according to Leach, (2023). Smoking accounts for about 25 percent of all kidney cancer cases according to Verma, (2021)while smokers face a 1.3 times greater risk of developing kidney cancer than non-smokers according to research by Lin et al. (2021). Wilms tumor shows a genetic link to WAGR syndrome and familial predisposition as its primary genetic association according to research by Spreafico et al. (2021).

Globally, kidney cancer is a major concern for public health. Recently, approximately 403,300 people were diagnosed with kidney cancer, and in 2018, nearly 175,000 deaths were recorded due to this cancer (Esser et al., 2020). Males are more commonly affected by kidney cancer than females, and people above 45 years of age are at a higher risk (Bukavina et al., 2022). The five-year survival rate in the United States is about 75% (National Cancer Institute, 2022). The comparative survival rate has been found to be 68% in Canada, 70% in China, and 60% in Europe (Zeng et al., 2018; Delam et al., 2023). The survival rate is dependent on the stage of cancer. If the cancer is limited to the kidney, the five-year survival rate is about 93% (National Cancer Institute, 2022). However, if the cancer affects the regional lymph nodes, the survival rate is 70% (Donelan et al., 2023). The survival rate further drops to almost 12% if the cancer has metastasized to distant parts (Padala et al., 2020). Kidney cancer is the 13th most common cancer globally, and its incidence has been steadily rising since 1930 (Yang et al., 2023).

The clinical presentation varies according to disease progression. The early stages of the disease show no symptoms which creates difficulties for medical professionals to identify the condition (Krishna et al., 2020). As the disease progresses patients will develop multiple symptoms including flank pain and a palpable renal mass and swollen lymph nodes and a persistent cough and night sweats and bilateral lower limb swelling (Challacombe & Bultitude, 2021; Yi, 2016). The typical symptomatic profile appears in only 15 percent of patients (Blumberg et al., 2020). The systemic biological effects of kidney cancer extend to its paraneoplastic syndromes (Novacescu et al., 2022). Medical professionals classify these syndromes into two main groups which include endocrine and non-endocrine disorders. Endocrine disorders cause hypertension and hypercalcemia and galactorrhea and polycythemia and Cushing's syndrome, while non-endocrine disorders lead to anemia and vasculopathy and neuromyopathies and coagulopathy and amyloidosis (Palapattu et al., 2020).

Diagnosis is mainly done through imaging. Most kidney masses are found incidentally during abdominal ultrasonography or computed tomography (CT) scans done for other complaints in the abdomen (Hines et al., 2021). About 60% of kidney cancers are diagnosed incidentally during abdominal imaging. Kidney masses can be differentiated according to cell type and imaging characteristics (Campbell & Lane, 2012). Ultrasonography is commonly employed in the initial assessment (Moses & Fernandez, 2022), though CT scans remain one of the most accurate imaging tools for diagnosis (Raty et al., 2020). Magnetic resonance imaging (MRI) of the abdomen is useful in staging and vascular involvement. Renal mass biopsy (RMB) is a reliable tool for histopathological confirmation of malignancy (Rouviere et al., 2020). Treatment modalities depend on histopathology and stage. Radiotherapy and chemotherapy are of limited use in kidney cancer (Campbell et al., 2021). Thus, surgical excision is the main and most successful treatment modality (Motzer et al., 2022). Targeted biologic therapies such as

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axitinib, sorafenib (Nexavar), everolimus, sunitinib (Sutent), and temsirolimus (Torisel) are commonly employed. Immunotherapeutic agents such as interleukin-2 and interferon have shown efficacy in about 10-15% of cases (Hemminki et al., 2022; Xu et al., 2020).

The connection between inflammation and cancer development has particular importance for the study of RCC. The immune system functions as a double-edged sword because it enables cancer development yet helps eliminate tumors. Tumor development occurs through chronic inflammation, while immune system stimulation leads to cancer defense mechanisms. CD8+ and CD4+ T-cells which make up cytotoxic T-lymphocytes, show strong anti-cancer effects. Their presence inside tumors leads to better patient outcomes according to Stockis et al. (2019) study. Regulatory T-cells (Treg cells) inhibit CD8+ and CD4+ T-cell function, which protects against excessive autoimmune reactions and allergic reactions. This mechanism enables tumors to escape immune system detection. Researchers have found CD8+ and CD4+ T-cell presence in RCC patients at both thymus and various blood and tissue sites including pleural effusion and intestinal tissues from infected and cancer patients according to Nishida et al. (2020) study. Scientists must study how effector and regulatory T-cells interact because this knowledge will help them create better immunotherapy treatments.

Oxidative stress and DNA damage further contribute to kidney carcinogenesis. Reactive oxygen species (ROS) can alter proteins, lipids, and nucleic acids, disrupting cellular function. An imbalance between ROS production and antioxidant defense mechanisms leads to oxidative stress and subsequent pathological conditions (Jelic et al., 2021). ROS are generated from endogenous sources such as mitochondria, inflammatory cells, and peroxisomes. Mitochondrial respiratory chain reactions produce superoxide ions, while inflammatory cells reduce molecular oxygen to generate ROS. Peroxisomes produce both superoxide and hydrogen peroxide, although antioxidant enzymes including superoxide dismutase (SOD), glutathione peroxidase (GPx), and catalase (CAT) help neutralize these reactive molecules (Jelic et al., 2021). Exogenous sources of ROS include tobacco smoke, alcohol, ionizing radiation, chemicals, chemotherapeutic agents, environmental toxins, and infectious agents (Jomova et al., 2023). Radiation induces DNA damage and apoptosis, while cigarette smoking causes DNA lesions that contribute to carcinogenesis (Birben et al., 2012). When ROS levels exceed critical thresholds, extensive cellular damage and cell death occur (Villalpando-Rodriguez et al., 2021). Collectively, oxidative stress, immune dysregulation, and environmental risk factors interact to drive the initiation and progression of kidney cancer.

METHODOLOGY

The present research study was performed in the Institute of Radiotherapy and Nuclear Medicine (IRNUM) Cancer hospital, Peshawar. The experimental steps were further conducted in Alpha Genomic Laboratory, Islamabad.

Total Subjects

The research was conducted on 60 humans in the age range of 20-40 years. Among them, 30 were healthy and 30 were diagnosed for kidney cancer. About 30 healthy vounteers with normal renal function and 30 patients diagnosed with kidney cancer by nephrologists were included. Patients with kidney problems other than cancer were excluded. The subjects were distributed as follows.

Table. 1. The distribution of samples

S. No.	Group	Types	No. of Subjects
1.	Control	Healthy subjects	30
2.	Affected	Kidney cancer patients	30

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Blood Sample Collection

The blood samples (3 mL) were collected under aseptic condition from 30 kidney cancer patients and 30 control individuals by vein puncture. These samples were added to heparin tubes, specifically used for blood collection and transported to Alpha Genomic Laboratory, Islamabad for flow cytometric, comet assay analysis and evaluation of oxidative stress markers including MDA, SOD and GSH.

Counting T-lymphocytes Using Flow Cytometry

From the collected blood, 2 µL was transferred to trucount tube or absolute count tube, which has a known amount of fluorescent beads. Following this, 20 µL of colored fluorescent antibodies were added to the blood in trucount tube. The mixture was vortexed properly and incubated in dark at room temperature for 20 minutes. Then, 450 µL of FACS hemolysin was added and the tubes were mixed thoroughly. The tubes were incubated in the dark at room temperature for 15 minutes. Fluorescent events were counted using the BD FACS Calibur flow cytometer (Zhang *et al.*, 2021).

Evaluating DNA Damage Using Comet Assay

One percent agarose gel was prepared for comet assay by mixing 1 gram of agarose powder in 100 mL of distilled water. Melt this powder in oven for 2 minutes. Clean glass slide was dipped in agarose. One side of the slide was clean with a lint free wipes. Theses slides were dried. The cell suspension was prepared and mixed with 1 % agarose (1:10 v/v). This cell suspension was thoroughly mixed by pipetting it. Following this, 30 µL of cell suspension was added to a clean microscopic slide and a thin layer was made using pipette. Incubate the slide in the dark for 10 minutes at 4 °C. The slides were immersed in electrophoresis chamber for 30 minutes at 4 °C. After electrophoresis, the excess buffer was washed and dried in the dark at 37 °C for 15 minutes. The slides were stained with fluorescent nucleic acid solution (green) and analyzed through epifluorescence microscope and comet assay software (Lu *et al.*, 2017).

Determination of Malondialdehyde (MDA)

Lipid peroxidation was determined by the synthesis of thiobarbituric acid reactive substances (TBARS). Tricholoroacetic acid (TCA 15%) was added to the samples to deproteinize it. Then thiobarbituric acid was mixed with the sample and this mixture was heated for 15 minutes in a hot water bath. The mixture was cooled and then centrifuged for 10 minutes. The synthesized TBARS was measured using spectrophotometer at 535 nm. The total TBARS calculated was expressed as MDA in nmol/mL.

Determination of Reduced Glutathione (GSH) Concentration

The GSH test was based on the appearance of yellow color after the addition of 2-nitrobenzoic acid to the sulphydryl compounds. This yellow color was stable for 10 minutes. Thus reaction mixture was immediately analyzed at 412 nm. The blood sample was mixed with distilled water and precipitated. Following this, 3 mL of this solution was transferred to a tube containing hemolysate and filtered. Eight milliliter of phosphate buffer was taken in a test tube and 2 mL of filtrate and 1 mL of DTNB was mixed with it. Absorbance of this solution was measured at 412 nm.

Determination of Superoxide Dismutase (SOD)

Pyrogallol autoxidation assay was performed to determine SOD concentration. For this, 100 µL pyrogallol solution (2.6 mM) was prepared and mixed with the HCL (10 mM). A final concentration of 0.13 mM was achieved and analyzed with spectrophotometer (Zahra *et al.*, 2021).

Statistical Analysis of Data

Data of all the biochemical parameters including $CD4^+$, $CD8^+$, $CD14^+$ T-cells, DNA damage, SOD, MDA and reduced GSH concentrations were compiled in Microsoft Excel and analyzed. Mean, standard deviation and p value was calculated. Mean is the average of all the given values. Standard deviation is the variation of variables expected about its mean and t test is the

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statistical test used for the comparison of mean of two groups.

RESULTS

Counting T-lymphocytes Using Flow Cytometry

The T-lymphocytes including CD4+, CD8+ and CD14+ were analyzed in kidney cancer patients and control group.

Concentration of CD4+ T-cells in Kidney Cancer Patients

In the control group aged 20-40 years, the CD4+ T-subsets analyzed were 2.74±0.58 cells/µL, which in case of affected group of age 20-40 years were higher in number (8.23±3.88 cells/µL) compared to the control group. Similarly, in case of control group aged 41-60 years, the CD4+ T-subsets were lower (2.95±0.57 cells/µL) as compared to affected group (12.06±4.71 cells/µL). Figure 1. summarizes the comparative statistical measure of the CD4+ T-cells in control and affected group. All the CD4+ T-cell values are shown as mean ± standard deviation.

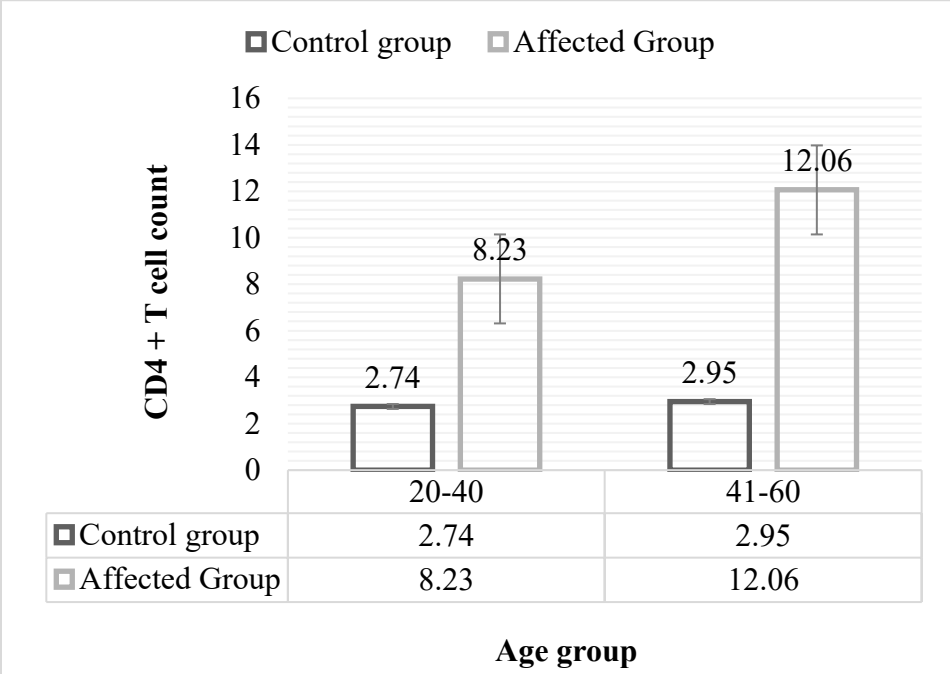


Fig. 1. Comparative statistical measure of the CD4+ T-cells

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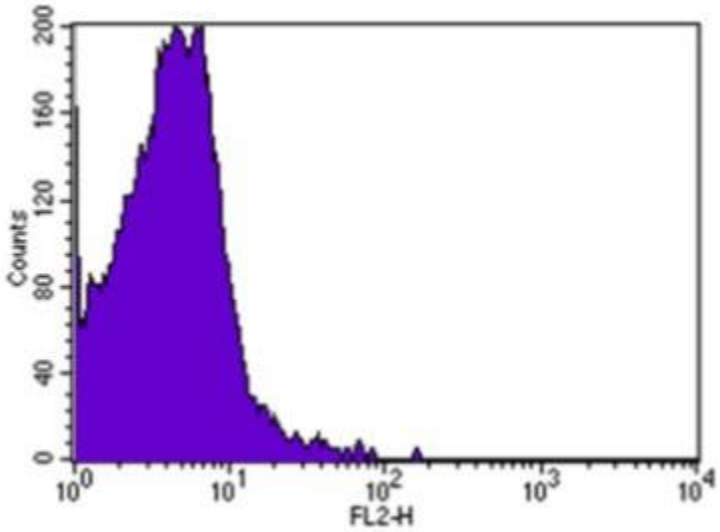


Fig. 2. Graphical depiction of CD4+ T-cells in affected and control group

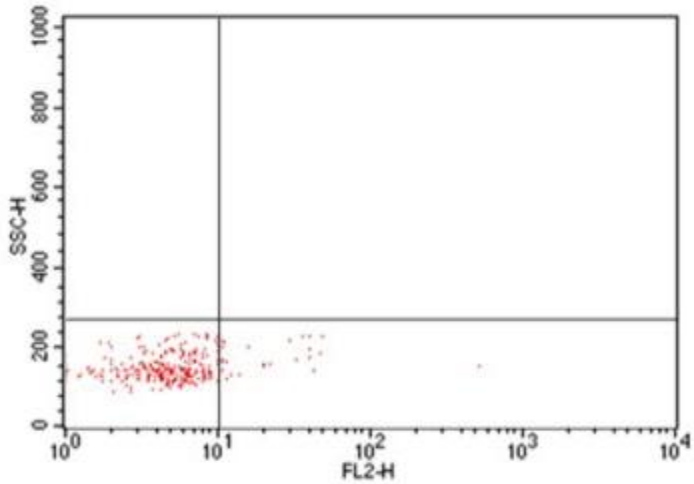


Fig. 3. Histogram A or light scatter parameter of CD4+ T-cells in affected group

Concentration of CD8 + T-cells in Kidney Cancer Patients

In control group aged 20-40 years, the CD8+ T-subsets analyzed were 2.55±0.67 (cells/µL). Which in case of affected group aged 20-40 years were higher in number (20.13±2.56 (cells/µL).) as compared to the control group.

Similarly, in control group aged 41-60 years, the CD8+ T-subsets analyzed were lower (2.90±0.55 cells/µL) as compared to the affected group (20.11±3.33 cells/µL). The statistical analysis of the CD8+ T-cells in control group and affected categorized by age groups is given in Figure 4.

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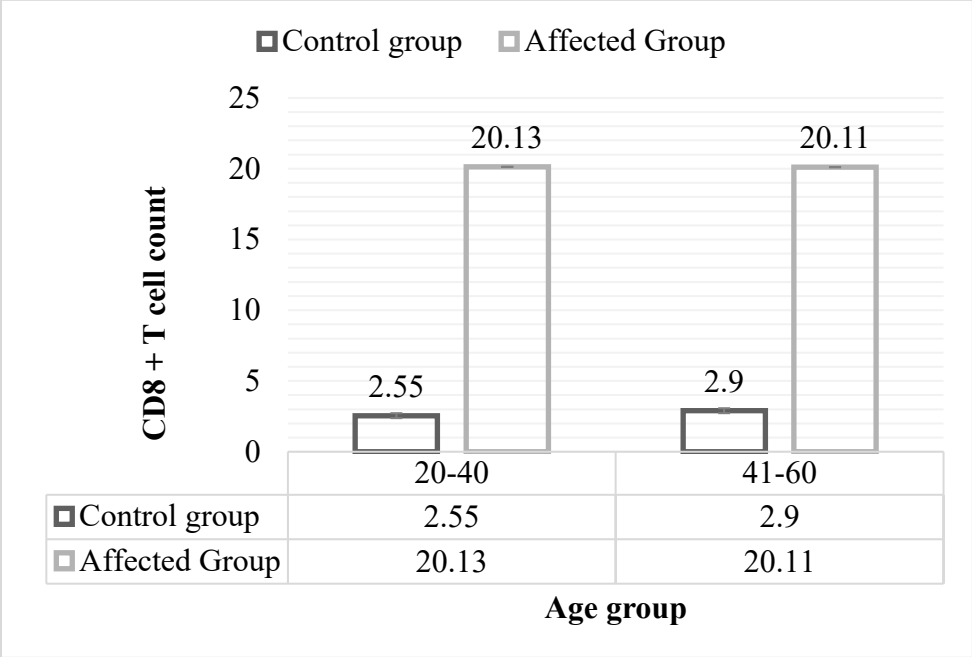


Fig. 4. Statistical analysis of CD8+ T-cells in control and affected group

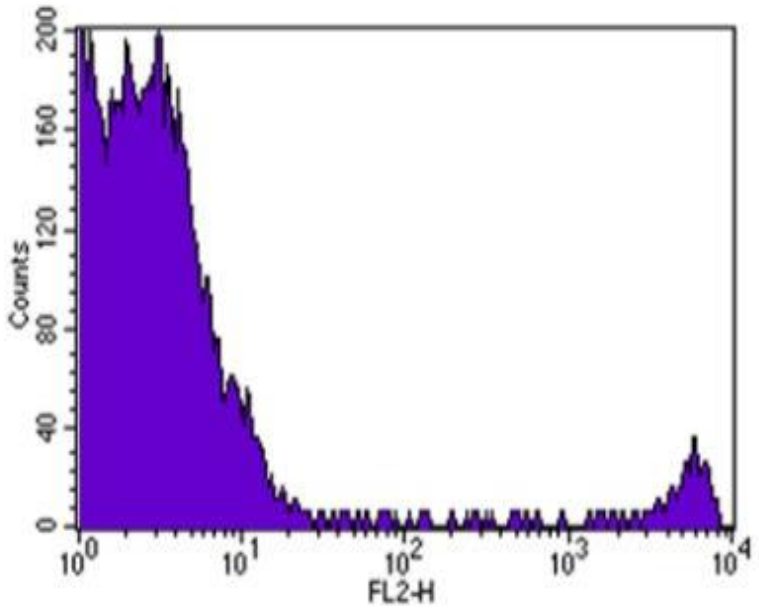


Fig. 5. Quantitative analysis of CD8+ T-cells in affected and control group

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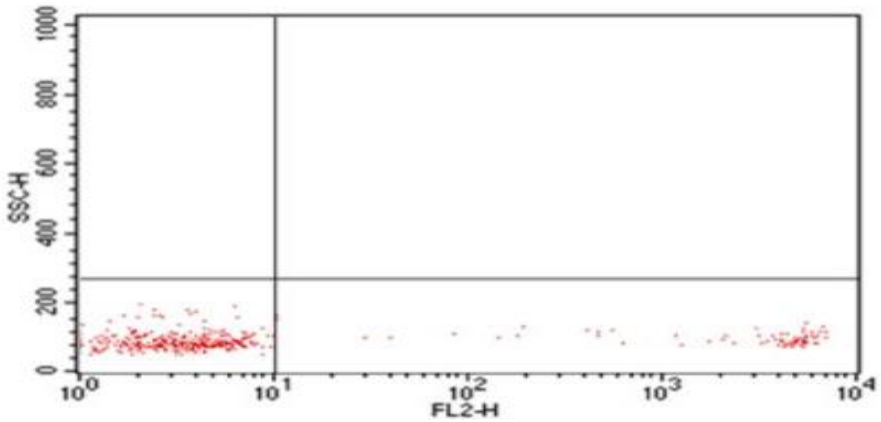


Fig. 6. Light scatter analysis of CD8+ T-cells in affected group

Concentration of CD14+ T-cells in Kidney Cancer Patients

The CD14+ T-cells subsets were 6.38±2.01 cells/µL in control group aged 20-40 years, which were lower as compared with the affected group aged 20-40 years (17.53±2.42 cells/µL). Whereas, in control group aged 41-60 years, these counts were lower (6.46±1.91 cells/µL) as compared to the affected group aged 41-60 years (19.75±3.96 cells/µL). The comparative statistical analysis of the CD14+ T-cells in control and affected group categorized by age groups is given in Figure 7.

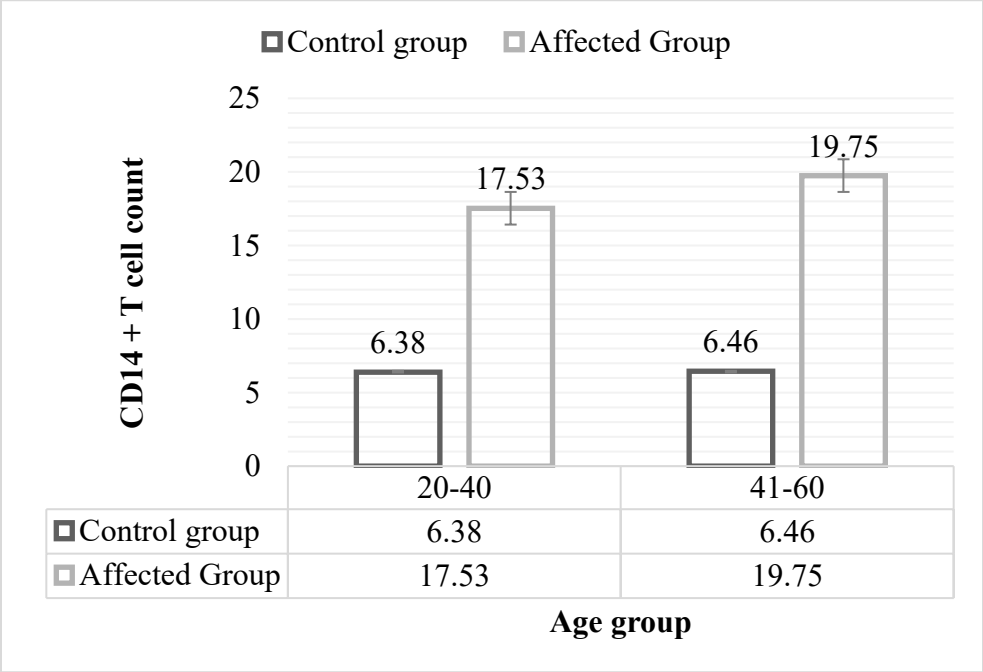


Fig. 7. Comparative statistical analysis of the CD14+ T-cells

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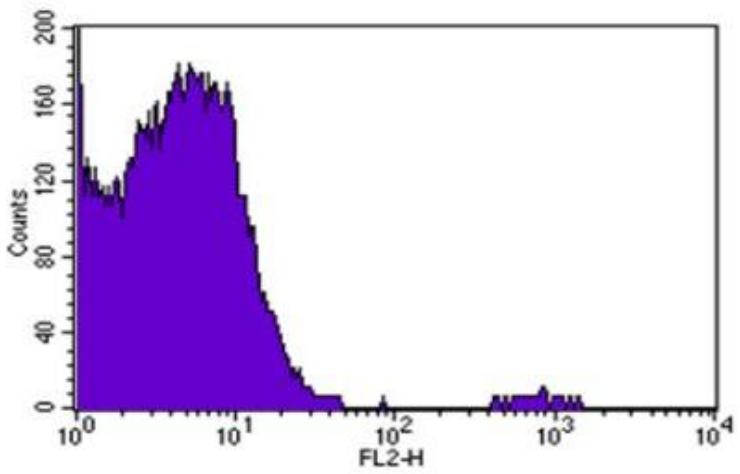


Fig. 8. Quantitative comparison of CD14+ T-cells of affected and control group

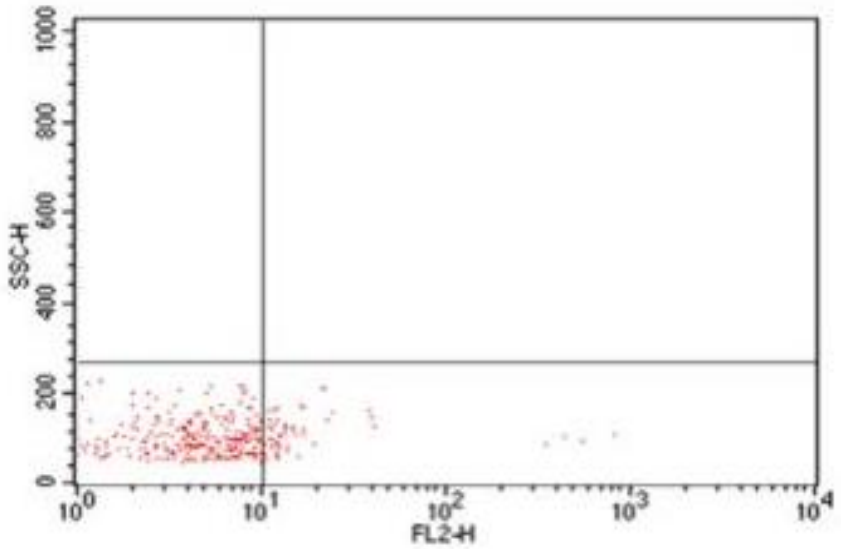


Fig. 9. Quantitative analysis of CD14+ T-cell light scatter in affected patients

Evaluating DNA Damage Using Comet Assay

The mean DNA length in control group aged 20-40 years calculated was 9.52±2.13 µm. The DNA length in case of affected group aged 20-40 years was slightly higher than the control group i-e., 24.58±4.73 µm. Whereas, in control group aged 41-60 years, these counts were lower (7.71±2.42 µm) as compared to the affected group aged 41-60 years (22.60±4.69 µm). Figure 10. shows the statistical measure of the DNA tail length in control and affected group of both age groups.

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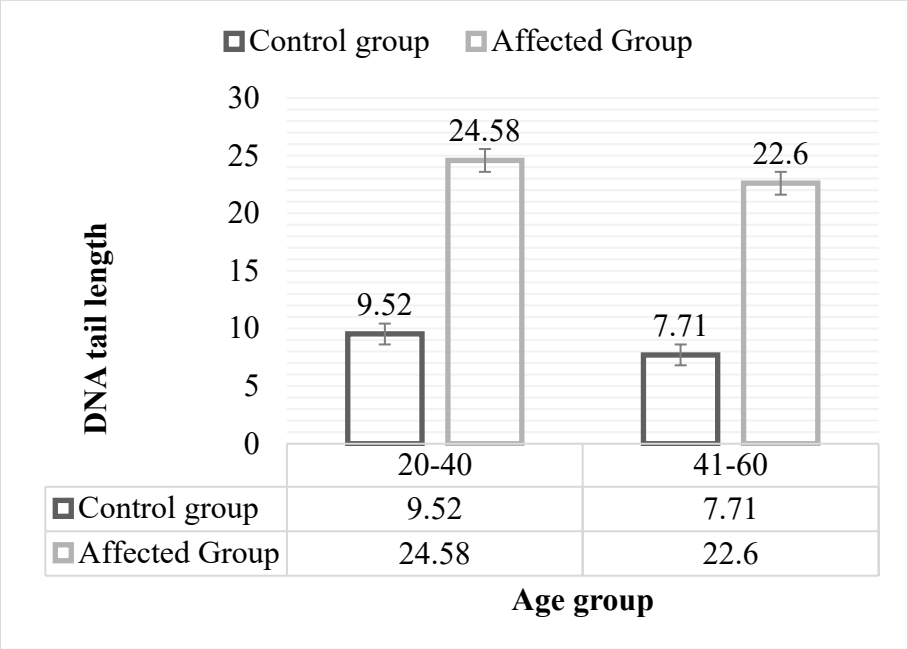


Fig. 10. Statistical measure of the DNA tail length in control and affected group

Malondialdehyde (MDA) concentration

The mean value of MDA concentration across age group 20-40 years in control group was 6.99±2.18 Nmol-mL. Whereas in same age group of affected patients, the MDA levels were higher (9.05±3.14 Nmol-mL). Similarly, the MDA concentration was also higher in affected patients aged 41-60 years when compared with the control group of the same age (6.77±2.57 Nmol-mL). Figure 11. shows significant differences in MDA concentration in both control group and kidney cancer patients. The MDA and SOD concentration was also higher in cancer group as compared to the control group. Whereas, the GSH concentration was lower in cancer group.

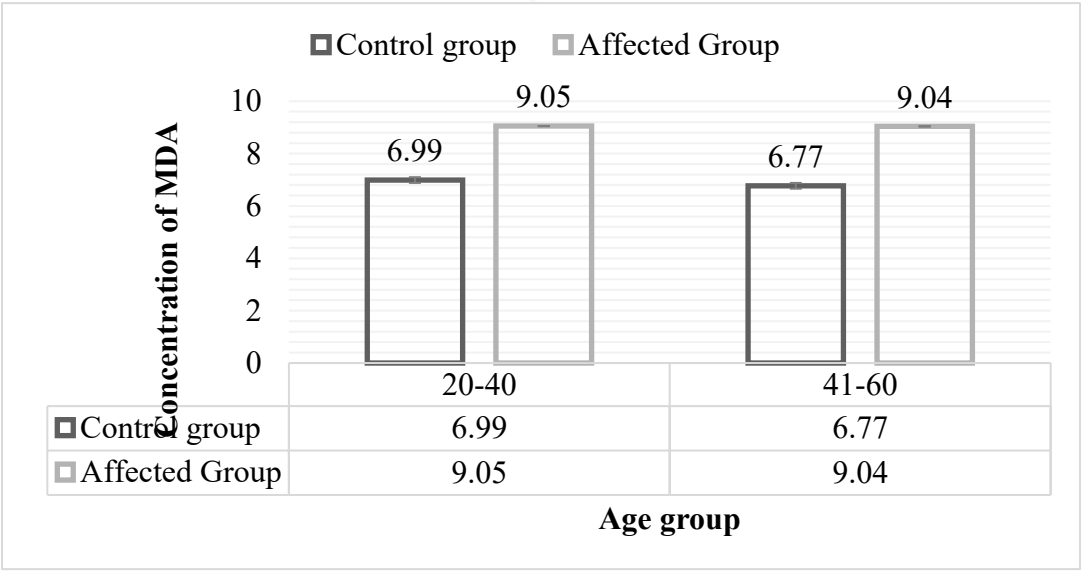


Fig. 11. Concentration of MDA in control and affected patients

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Reduced Glutathione (GSH) Concentration

The GSH levels were lower (52.63±3.41) in affected patients aged 20-40 years as compared to the control group aged 20-40 years. Moreover, in case of age group 41-60 years, the GSH levels were higher in control group (67.17±2.86 Nmol-mL) as compared to the affected patients (55.65±4.44 Nmol-mL). Figure 12. shows the comparative reduced GSH concentration among the 2 groups i-e. control and affected group.

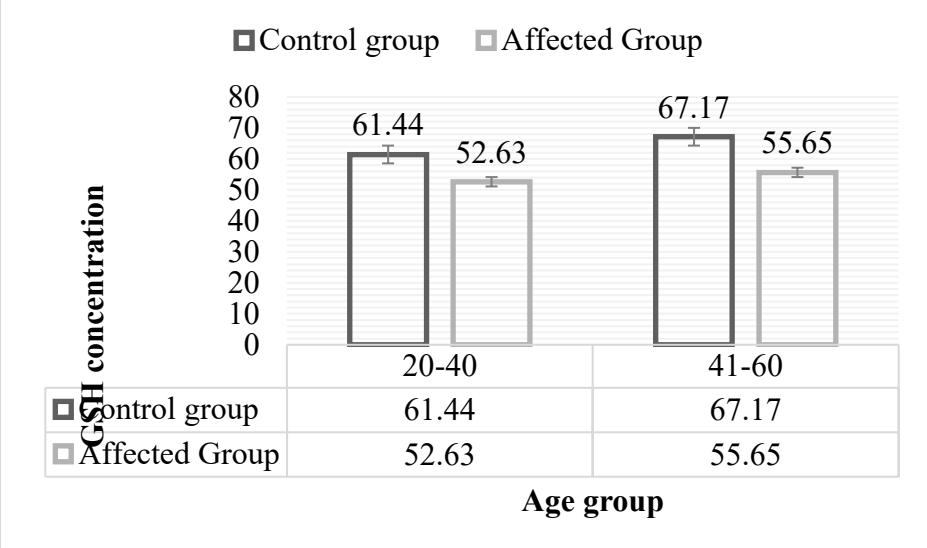


Fig. 12. Comparative statistical measure of the GSH concentration in control and affected group

Superoxide Dismutase (SOD) Concentration

In the control group individuals aged 20-40 years showed a mean SOD concentration of 57.36±1.24 Nmol-mL, significantly lower than the SOD concentration of affected group aged 20-40 years. Similarly, individuals of age 41-60 years in control group showed an average SOD concentration of 65.08±1.95 Nmol-mL, significantly lower compared to the affected patients (74.61±3.97 Nmol-mL). Figure 13 shows significant differences in SOD concentrations in different age groups.

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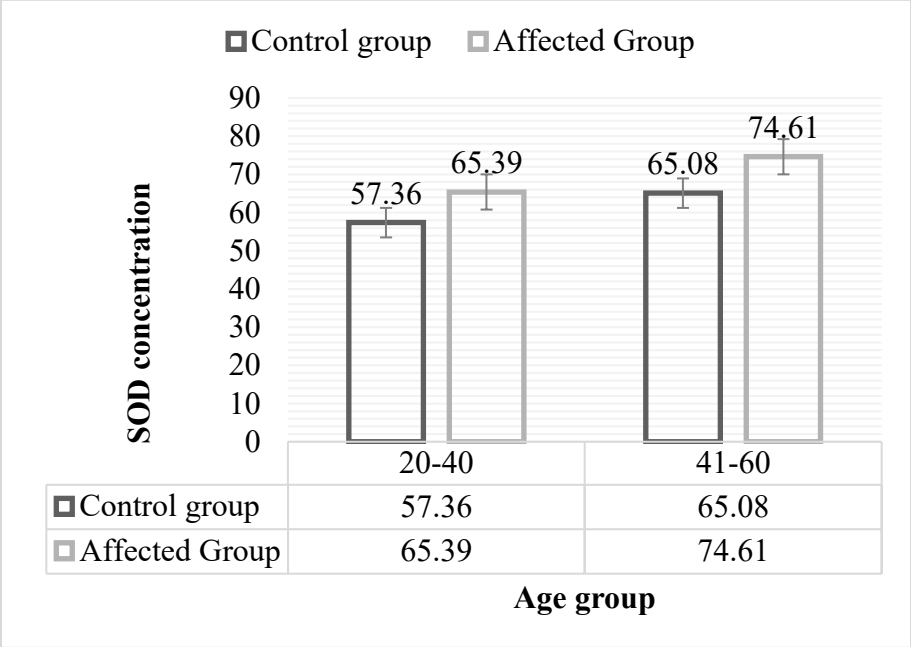


Fig. 13. Quantitative analysis of SOD concentration in both group

DISCUSSION

Cancer is the uncontrolled, continuous abnormal replication of body cells, which leads to the tumor formation (Rajendra & Kedari. 2020). Among all cancers, kidney cancer is one of the frequent urological tumor. Owing to the high death rate and prevalence, kidney cancer is the 3rd most common malignant tumor of urinary system, which is followed by bladder and prostate cancer. It is resistant to chemo and radiotherapy and can develop drug resistance easily (Mao *et al.*, 2021). It is the 15th most common malignancy globally, with a higher occurrence rate in developed countries (Harrison *et al.*, 2021).

The current study was undertaken on 30 kidney cancer patients of age 20-60 years. This study also included 30 healthy individuals of same age group. The T-lymphocytes including CD4 +, CD8 + and CD14 +were analyzed in kidney cancer patients and control group in the current study. The results presented here in showed that in case of control group (20-40 years), the CD4 + cells were 2.74±0.58 with $p<0.0001$. In case of cancer group aged 20-40 years, the CD4 + cells were higher in number (8.23±3.88) compared to the control group. Similarly, in case of control group (41-60 years), the CD4 + T-subsets were lower (2.95±0.57) compared with the affected group (12.06±4.71). Su *et al.* (2021) also reported CD4 + T-cells ($p<0.01$) from stage 4 renal cell carcinoma (RCC) patients. Similarly, Nishida *et al.* (2020) reported up-regulated expression of CD4 + and CD8 + T-cells in renal cell carcinoma. According to Menard *et al.* (2018), their study showed the expansion of CD4 + and CD8 + T-cells in renal cell carcinoma patients. According to Kawashima *et al.* (2020), the expression of CD4 + and CD8 + T cells should be evaluated because these T lymphocytes play a crucial role in targeting tumor cells. The results of their study implied that the T-lymphocytes were upregulated in RCC patients of grade 3 and grade 4 ($p<0.001$).

The CD8 + and CD14 + T-cells were also higher in affected patients in the current study. Su *et al.* (2021) also reported that CD4 + and CD8 + are the major immune cells in renal cell carcinoma patients. Chevrier *et al.* (2017) performed experimental study, which showed that macrophages followed by CD4 + and CD8 + T-cells were the common immune cells reported from kidney cancer patients. These results were

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in agreement with Qi *et al.* (2020), who found poor prognosis of CD8 + T-cells ($p<0.0001$). Another study by Miao *et al.* (2022) showed statistical differences with $p<0.05$ in CD4 + T-cells. Similarly, Borcherding *et al.* (2021) reported high CD8 + T cells in tumor infiltering cells. On the other hand, the control tissues showed low concentration of CD8 + T cells.

The blood MDA levels in the current study were higher in affected patients than the control group in both 20-40 and 41-60 years age group. Lepara *et al.* (2020) reported higher MDA levels (0.86-1.05 µmol/L) in cancer patients as compared to the control group (0.6-0.7 µmol/L). Vida *et al.* (2021) reported higher MDA concentration in their study.

The current study also reported higher SOD and lower GSH concentration in blood of affected patients compared to the control group. Similarly, Vida *et al.* (2021) analyzed impaired antioxidant enzymes and altered redox including SOD, GSH and GPx, when compared with the blood concentration of healthy individual. Rajendra and Kidari (2020) observed superoxide dismutase (SOD), malondialdehyde (MDA), glutathione reductase (GR) and glutathione peroxidase (GPx). They reported higher MDA levels (9.91±4) in cancer patients as compared to the control (8.56±3.6). The SOD levels were lower (9.4±5.1) in cancer patients. Furthermore, GR and GPx concentration was also lower (12.7±2.4; 43.5±10.5) in cancer patients when compared with the healthy people.

Kaba *et al.* (2024) observed reported higher concentration of MDA levels in affected patients. Whereas, CAT, SOD and GPx levels were lower as compared to MDA levels. According to Van der Slikke *et al.* (2021), oxidative damage markers, mitochondrial mass and DNA damage was upregulated in renal and sepsis patients as compared to healthy controls. The oxidative DNA damage was observed in tubular epithelial cells.

Conclusion

The research was conducted on 60 subjects. Among these, 30 were healthy subjects of age 20-60 years and 30 were kidney cancer subjects of same age group. The T-lymphocytes including CD4+, CD8+ and CD14+ were analyzed in kidney cancer patients and control group. In case of kidney cancer patients, T-lymphocytes including CD4+, CD8+ and CD14+ were higher as compared to the control group. The mean DNA length was also higher in kidney cancer patients when compared with the control group. Similarly, the MDA and SOD concentration was also higher in kidney patients in both cases. Whereas, the GSH concentration was lower in cancer patients when compared with the healthy group.

REFERENCES

Alzubaidi, A. N., Sekoulopoulos, S., Pham, J., Walter, V., Fuletra, J. G., & Raman, J. D. (2022). Incidence and distribution of new renal cell carcinoma cases: 27-year trends from a statewide cancer registry. *J. Kidney Cancer VHL*, *9*(2), 1-7.

Bahadoram, S., Davoodi, M., Hassanzadeh, S., Bahadoram, M., Barahman, M., & Mafakher, L. (2022). Renal cell carcinoma: an overview of the epidemiology, diagnosis, and treatment. *G. Ital. Nefrol*, *39*(3), 1-8.

Birben, E., Sahiner, U. M., Sackesen, C., Erzurum, S., & Kalayci, O. (2012). Oxidative stress and antioxidant defense. *WAO J*, *5*(7), 9-19.

Blumberg, J. M., Elkhoury, F. F., & Gollapudi, K. (2020). Blood in Urine. *Surgery: A Case Based Clinic. Rev*, 607-615.

Borcherding, N., Vishwakarma, A., Voigt, A. P., Bellizzi, A., Kaplan, J., Nepple, K., & Zhang, W. (2021). Mapping the immune environment in clear cell renal carcinoma by single-cell genomics. *Commun. Biol*, *4*(1), 1-22.

Bukavina, L., Bensalah, K., Bray, F., Carlo, M., Challacombe, B., Karam, J. A., & Psutka, S. P. (2022). Epidemiology of renal cell carcinoma: 2022 update. *Europ. Urol*, *82*(5), 529-542.

Frontier in Medical & Health Research

ISSN: (e) 3007-1607 (p) 3007-1593

Campbell, S. C., & Lane, B. R. (2012). Malignant renal tumors. *Campbell-Walsh Urol*, 1413-1474.

Campbell, S. C., Clark, P. E., Chang, S. S., Karam, J. A., Souter, L., & Uzzo, R. G. (2021). Renal mass and localized renal cancer: evaluation, management, and follow-up: AUA guideline: part I. *J. Urol*, *206*(2), 199-208.

Challacombe, B., & Bultitude, M. F. (2021). The kidneys, urinary tract and prostate. In *Browse's Introduction to the Symptoms & Signs of Surgical Disease* (pp. 573-587). CRC Press.

Chevrier, S., Levine, J. H., Zanotelli, V. R. T., Silina, K., Schulz, D., Bacac, M., & Bodenmiller, B. (2017). An immune atlas of clear cell renal cell carcinoma. *Cell*, *169*(4), 736-749.

Delam, H., Keshtkaran, Z., Eidi, A., Zare, R., Shahedi, A., Hassanipour, S., & Abdollahpour-Alitappeh, M. (2023). Survival Rate of Kidney Cancer in the Asian Countries: A Systematic Review and Meta-Analysis study. *20*(2), 1-20.

Donelan, W., Brisbane, W., O'Malley, P., Crispen, P., & Kusmartsev, S. (2023). Hyaluronan Metabolism in Urologic Cancers. *Adv. Biol*, *7*(12), 1-16.

Esser, L. K., Branchi, V., Leonardelli, S., Pelusi, N., Simon, A. G., Klümper, N., & Toma, M. I. (2020). Cultivation of clear cell renal cell carcinoma patient-derived organoids in an air-liquid interface system as a tool for studying individualized therapy. *Front. Oncol*, *10*(3), 1-17.

Harrison, H., Thompson, R. E., Lin, Z., Rossi, S. H., Stewart, G. D., Griffin, S. J., & Usher-Smith, J. A. (2021). Risk prediction models for kidney cancer: a systematic review. *Europ. Urol. Focus*, *7*(6), 1380-1390.

Hemminki, O., Perlis, N., Bjorklund, J., Finelli, A., Zlotta, A. R., & Hemminki, A. (2020). Treatment of advanced renal cell carcinoma: immunotherapies have demonstrated overall survival benefits while targeted therapies have not. *Europ. Urol. Open Sci*, *22*(52), 61-73.

Hines, J. J., Eacobacci, K., & Goyal, R. (2021). The incidental renal mass-update on characterization and management. *Radiol. Clinics*, *59*(4), 631-646.

Jelic, M. D., Mandic, A. D., Maricic, S. M., & Srdjenovic, B. U. (2021). Oxidative stress and its role in cancer. *J. Cancer Res. Therap*, *17*(1), 22-28.

Kaba, M., Pirincci, N., Demir, H., & Verep, S. (2024). Serum prolidase activity, oxidative stress, and antioxidant enzyme levels in patients with prostate cancer. In *Urologic Oncology: Seminars and Original Investigations*. *Elsevier*. *3*(91), 1-14.

Kamal, S., & Ramzan, F. (2023). Epidemiology of Kidney Cancer in District "Swat" Khyber Pakhtunkhwa. *Gomal J. Agricul. Biol*, *1*(01), 65-80.

Kawashima, A., Kanazawa, T., Kidani, Y., Yoshida, T., Hirata, M., Nishida, K., & Nonomura, N. (2020). Tumour grade significantly correlates with total dysfunction of tumour tissue-infiltrating lymphocytes in renal cell carcinoma. *Sci. Rep*, *10*(1), 1-20.

Krishna, S., Leckie, A., Kielar, A., Hartman, R., & Khandelwal, A. (2020, April). Imaging of renal cancer. In *Seminars in Ultrasound, CT and MRI* (Vol. 41, No. 2, pp. 152-169). WB Saunders.

Leach, D. (2023). Urological Cancers. In *Stoma Care Specialist Nursing: A Guide for Clinical Practice* (pp. 117-137). Cham: Springer International Publishing.

Lepara, Z., Lepara, O., Fajkić, A., Rebić, D., Alić, J., & Spahović, H. (2020). Serum malondialdehyde (MDA) level as a potential biomarker of cancer progression for patients with bladder cancer. *Roman. J. Intern. Med*, *58*(3), 146-152.

Frontier in Medical & Health Research

ISSN: (e) 3007-1607 (p) 3007-1593

Lin, L., Li, Z., Yan, L., Liu, Y., Yang, H., & Li, H. (2021). Global, regional, and national cancer incidence and death for 29 cancer groups in 2019 and trends analysis of the global cancer burden, 1990–2019. *J. Hematol. Oncol*, *14*, 1-24.

Lu, Y., Liu, Y., & Yang, C. (2017). Evaluating in vitro DNA damage using comet assay. *JoVE* *6*(128), 1-21.

Mao, W., Wang, K., Wu, Z., Xu, B., & Chen, M. (2021). Current status of research on exosomes in general, and for the diagnosis and treatment of kidney cancer in particular. *J. Experiment. Clinic. Cancer Res*, *40*(1), 1-14.

Menard, L. C., Fischer, P., Kakrecha, B., Linsley, P. S., Wambre, E., Liu, M. C., & Nadler, S. G. (2018). Renal cell carcinoma (RCC) tumors display large expansion of double positive (DP) CD4+ CD8+ T cells with expression of exhaustion markers. *Front. Immunol.* *9*(1), 1-27.

Miao, K., Zhang, X., Wang, H., Si, X., Ni, J., Zhong, W., & Zhang, L. (2022). Peripheral Blood Lymphocyte Subsets Predict the Efficacy of Immune Checkpoint Inhibitors in Non–Small Cell Lung Cancer. *Front. Immunol*, *13*(7), 1-18.

Moore, L. E., Stewart, P. A., & Karami, S. (2020). Kidney Cancer. *Occupational Cancers*, *22*(14), 467-486.

Moses, A. A., & Fernandez, H. E. (2022). Ultrasonography in acute kidney injury. *POCUS J*, *7*(10), 1-35.

Motzer, R. J., Jonasch, E., Agarwal, N., Alva, A., Baine, M., Beckermann, K., & Motter, A. (2022). Kidney cancer, version 3.2022, NCCN clinical practice guidelines in oncology. *J. Nat. Compreh. Cancer Network*, *20*(1), 71-90.

National Cancer Institute. (2022). *Cancer stat facts: Kidney and renal pelvis cancer*. https://seer.cancer.gov/statfacts/html/kidrp.html

Nishida, K., Kawashima, A., Kanazawa, T., Kidani, Y., Yoshida, T., Hirata, M., & Wada, H. (2020). Clinical importance of the expression of CD4+ CD8+ T cells in renal cell carcinoma. *Int. Immunol*, *32*(5), 347-357.

Novacescu, D., Feciche, B. O., Cumpanas, A. A., Bardan, R., Rusmir, A. V., Bitar, Y. A., ... & Latcu, S. C. (2022). Contemporary clinical definitions, differential diagnosis, and novel predictive tools for renal cell carcinoma. *Biomed*, *10*(11), 1-29.

Padala, S. A., Barsouk, A., Thandra, K. C., Saginala, K., Mohammed, A., Vakiti, A., & Barsouk, A. (2020). Epidemiology of renal cell carcinoma. *World. J. Oncol*, *11*(3), 1-7.

Palapattu, G. S., Kristo, B., & Rajfer, J. (2020). Paraneoplastic syndromes in urologic malignancy: the many faces of renal cell carcinoma. *Rev. Urol*, *4*(4), 1-6.

Qi, Y., Xia, Y., Lin, Z., Qu, Y., Qi, Y., Chen, Y., & Xu, J. (2020). Tumor-infiltrating CD39+ CD8+ T cells determine poor prognosis and immune evasion in clear cell renal cell carcinoma patients. *Cancer Immunol. Immunother*, *69*(9), 1565-1576.

Rajendra, C. H., & Kedari, G. S. R. (2020). Role of antioxidant levels and lipid peroxidation in patients with breast cancer. *Biomed*, *40*(2), 170-174.

Raty, P., Mentula, P., Lampela, H., Nykänen, T., Helantera, I., Haapio, M., & Sallinen, V. (2020). INtravenous Contrast computed tomography versus native computed tomography in patients with acute Abdomen and impaired Renal functiOn (INCARO): a multicentre, open-label, randomised controlled trial-study protocol. *BMJ open*, *10*(10), 1-13.

Rouviere, O., Cornelis, F., Brunelle, S., Roy, C., André, M., Bellin, M. F., & Renard-Penna, R. (2020). Imaging protocols for renal multiparametric MRI and MR urography: results of a consensus conference from the French Society of Genitourinary Imaging. *Europ. Radiol*, *30*(11), 2103-2114.

Frontier in Medical & Health Research

ISSN: (e) 3007-1607 (p) 3007-1593

Spreafico, F., Fernandez, C. V., Brok, J., Nakata, K., Vujanic, G., Geller, J. I., & Pritchard-Jones, K. (2021). Wilms tumour. *Nature Rev. Dis. Primers*, *7*(1), 1-5.

Stockis, J., Roychoudhuri, R., & Halim, T. Y. (2019). Regulation of regulatory T cells in cancer. *Immunol*, *157*(3), 219-231.

Su, S., Akbarinejad, S., & Shahriyari, L. (2021). Immune classification of clear cell renal cell carcinoma. *Sci. Rep*, *11*(1), 1-38.

Van der Slikke, E. C., Star, B. S., van Meurs, M., Henning, R. H., Moser, J., & Bouma, H. R. (2021). Sepsis is associated with mitochondrial DNA damage and a reduced mitochondrial mass in the kidney of patients with sepsis-AKI. *Critical Care*, *25*(9), 1-13.

Vasudev, N. S., Wilson, M., Stewart, G. D., Adeyoju, A., Cartledge, J., Kimuli, M., & Banks, R. E. (2020). Challenges of early renal cancer detection: symptom patterns and incidental diagnosis rate in a multicentre prospective UK cohort of patients presenting with suspected renal cancer. *BMJ open*, *10*(5), 1-18.

Verma, S. (2021). A Review Study of Cancer in Human Body by Tobacco or Smoking. *TRS*, *14*(10), 2286-2295.

Vida, C., Oliva, C., Yuste, C., Ceprian, N., Caro, P. J., Valera, G., & Carracedo, J. (2021). Oxidative stress in patients with advanced CKD and renal replacement therapy: the key role of peripheral blood leukocytes. *Antioxidants*, *10*(7), 1-11.

Villalpando-Rodriguez, G. E., & Gibson, S. B. (2021). Reactive oxygen species (ROS) regulates different types of cell death by acting as a rheostat. *Oxidat. Med. Cell. Long*, *20*(21), 1-19.

Xu, W., Atkins, M. B., & McDermott, D. F. (2020). Checkpoint inhibitor immunotherapy in kidney cancer. *Nat. Rev. Urol*, *17*(3), 137-150.

Yang, L., Wu, X., Zhou, J., Song, P., Liu, Z., Chen, J., & Dong, Q. (2023). Second malignant tumors and non-tumor causes of death for patients with localized and regional kidney cancer after diagnosis. *Europ. J. Med. Res*, *28*(1), 1-20.

YI, S. M. (2016). Investigations of ym155, an inhibitor of survivin, as a molecular multi-targeted agent for renal cell carcinoma. *Oxidat. Med. Cell. Long*, *27*(6), 1-7.

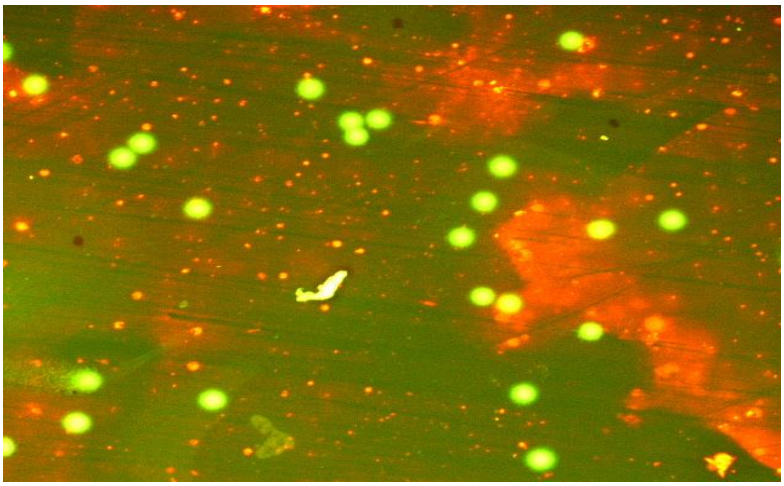
Zahra, K., Patel, S., Dey, T., Pandey, U., & Mishra, S. P. (2021). A study of oxidative stress in cervical cancer-an institutional study. *Biochem. Biophy. Rep*, *25*(8), 1-10.

Zeng, H., Chen, W., Zheng, R., Zhang, S., Ji, J. S., Zou, X., & He, J. (2018). Changing cancer survival in China during 2003–15: a pooled analysis of 17 population-based cancer registries. *The Lancet Glob. Health*, *6*(5), 555-567.

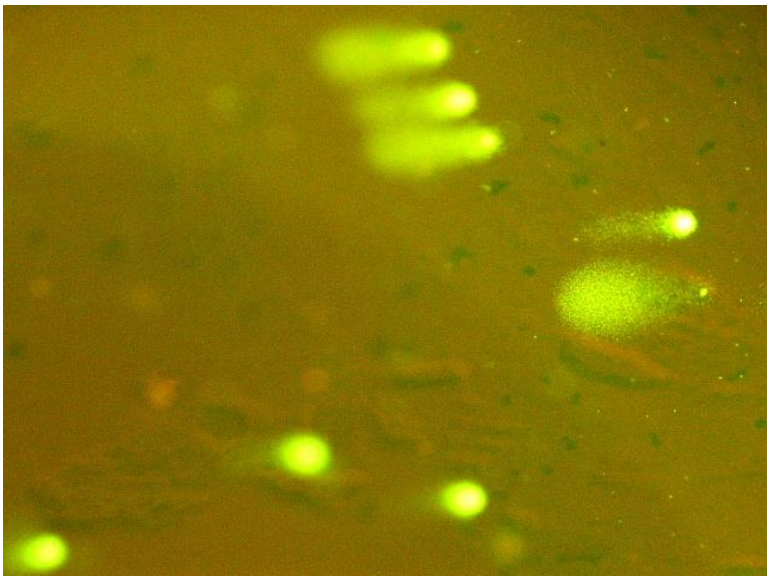
Zhang, Y., Ji, Y., Yu, L., Zhao, Y., Liu, G., & Dong, L. (2021). Characteristics of T lymphocyte subsets and their apparent effect on infection in patients with chronic kidney disease. *BMJ open*, *10*(9), 1-13.

Frontier in Medical & Health Research

ISSN: (e) 3007-1607 (p) 3007-1593



App. 1. DNA of control group



App. 2. DNA of affected group