TP53 As A Early Recurrence-Linked Biomarker In Tongue Squamous Cell Carcinoma: A Proteomic And Bioinformatic Study

G. V. Lakshmi[1*], K. Eswar Kumar[2], Parthasarathi Bhattacharyya[3], K. G. K. Deepak[4], John Dogulas Palleti[5]

[1, 2] College of Pharmaceutical Sciences, Andhra University, Visakhapatnam, India
[3] Department of Radiation Oncology, Mahatma Gandhi Cancer Hospital and Research Institute, Visakhapatnam, India
[4] Department of OncoCollect, Mahatma Gandhi Cancer Hospital and Research Institute, Visakhapatnam, India
[5] Research and Development, Centre for Computational and Biological Sciences (CCBS), Visakhapatnam, India.
*Corresponding author: GV Lakshmi, College of Pharmaceutical Sciences, Andhra University, Visakhapatnam; Email: gvl.260895@gmail.com

ABSTRACT

Tongue squamous cell carcinoma (TSCC) is characterized by high recurrence rates and poor prognosis, yet the underlying molecular mechanisms remain incompletely defined. This study investigates the role of TP53 in TSCC recurrence using integrative proteomic and bioinformatic approaches. Unpublished LC-MS/MS data of our research revealed significant upregulation of TP53 in early recurrence-positive samples compared to non-recurrent cases. To contextualize this finding, TP53 was compared to 16 recurrence-associated genes (e.g., FOXM1, OCT4, SNAI1, BRCA1/2, BCL2, EGFR, ALDH1A1, CD44, MYC, PTEN, SOX2, TWIST1, NANOG, and ABCB1) through BLASTp alignment and phylogenetic analysis. Homology modeling of TP53 was conducted via SWISS-MODEL using the top-scoring PDB template, and model quality was validated by Ramachandran plot analysis. Molecular docking using AutoDock Vina showed favourable binding of carboplatin (–3.6 kcal/mol) and 5-fluorouracil (–3.4 kcal/mol) to the TP53 core domain, with interactions involving key binding residues. Protein–protein interaction analysis via STRING identified TP53 hub connections to MDM2, CDKN1A, and DNA damage regulators. Functional enrichment using DAVID highlighted TP53 involvement in apoptosis, cell cycle arrest, and the p53

[1] . INTRODUCTION

Tongue squamous cell carcinoma (TSCC) is a common and aggressive form of oral cancer, with increasing incidence among younger, non-smoking individuals (Patel et al., 2011; Sgaramella et al., 2018). Despite advances in multimodal therapy, locoregional recurrence remains a major challenge, contributing to poor five-year survival rates (El-Husseiny et al., 2000; Brockstein et al., 2004). Field cancerization and the presence of cancer stem cells are thought to underlie multifocal disease and recurrence (Braakhuis et al., 2005; Simple et al., 2015).

At the molecular level, TSCC is driven by dysregulation of key genes such as TP53, EGFR, and CDKN2A (Caponio et al., 2020; Qiao et al., 2022). Although TP53 mutations are among the most common alterations in head and neck squamous cell carcinoma (HNSCC), their specific role in TSCC recurrence is not fully understood (Alsofyani et al., 2020; Zhou et al., 2016). While multi-gene signatures have been proposed for predicting relapse, TP53 has often been absent from these panels (Enokida et al., 2017). Proteomic technologies offer valuable tools for identifying recurrence-associated biomarkers in TSCC. Differential expression of cytoskeletal proteins, metabolic enzymes, and stress-response factors has been

signaling pathway. Collectively, these findings support TP53 as a key molecular player in TSCC recurrence and a potential target for therapeutic intervention.

Keywords: *TP53, Tongue Squamous Cell Carcinoma (TSCC), Proteomics, Recurrence, Cancer Biomarkers.*
reported in recurrent tumors (Zhang et al., 2020). However, clinical translation remains limited, highlighting the need for integrative approaches combining proteomics with structural and functional analysis.

In this study, we investigate the role of TP53 using our unpublished LC-MS/MS proteomic data, identifying its upregulation in early recurrent TSCC cases. Comparative analysis with 16 literaturesupported recurrence genes was performed using sequence alignment and phylogenetic methods. Homology modeling and molecular docking were conducted to examine TP53's interaction with carboplatin and 5-fluorouracil. Finally, we used STRING and DAVID analyses to characterize TP53's interaction network and biological functions. This integrative approach aims to clarify TP53's role in TSCC recurrence and its potential as a therapeutic target.

MATERIALS AND METHODS

Sequence analysis and phylogenetics. The amino-acid sequence of TP53 was taken from our previous research study. Sequences of 16 recurrence-associated proteins (e.g. OCT4, FOXM1, SNAI1, BRCA1/2, BCL2, EGFR, ALDH1A1, CD44, MYC, PTEN, SOX2, TWIST1, NANOG, ABCB1) were obtained from UniProt (Table 1). Pairwise BLASTp (NCBI BLAST) was used to assess sequence similarity (default parameters). A multiple sequence alignment of TP53 and these proteins was constructed (Clustal Omega- https://www.ebi.ac.uk/jdispatcher/msa/clustalo), and a phylogenetic tree was generated and visualized to examine evolutionary relationships (MEGA- https://www.megasoftware.net).

Table 1: List of Recurrence-Associated Genes in Tongue Squamous Cell Carcinoma with UniProt IDs and Protein Annotations

Gene	UniProt ID	Protein Name
OCT4	Q01860	POU domain, class 5, transcription factor 1
FOXM1	Q08050	Forkhead box protein M1
SNAI1	O95863	Zinc finger protein SNAI1
BRCA1	P38398	Breast cancer type 1 susceptibility protein
BCL2	A0A7I2V3S7	Apoptosis regulator Bcl-2
EGFR	C9JYS6	Receptor protein-tyrosine kinase
ALDH1A1	P00352	Aldehyde dehydrogenase 1A1
CD44	E9PKC6	CD44 antigen
MYC	B3CJ64	V-myc myelocytomatosis viral oncogene homolog
PTEN	A0AAQ5BH13	Phosphatidylinositol 3,4,5-trisphosphate 3-phosphatase and dualspecificity protein phosphatase PTEN
BRCA2	A0A7P0T9D7	Breast cancer type 2 susceptibility protein

SOX2	P48431	Transcription factor SOX-2
TP53	A0A386NDB3	Cellular tumor antigen p53
TWIST1	Q15672	Twist-related protein 1
NANOG	J7HA98	NANOG
ABCB1	A1L471	ATP-binding cassette, sub-family B (MDR/TAP), member 1

Homology modeling and validation. To model the full-length TP53 structure, we performed a BLASTp search of the TP53 sequence against the PDB. The highest-identity template (with sufficient coverage) was used in SWISS-MODEL to build a homology model of TP53. The resulting 3D model was refined and energy-minimized. Model quality was assessed by Ramachandran plot analysis (PROCHECK), ensuring in and around 90% of residues in favoured and additionally allowed regions.

Molecular docking. We docked two TSCC chemotherapeutic drugs (carboplatin and 5-fluorouracil) to the TP53 model using AutoDock Vina. Ligand structures were prepared in optimized conformations. A grid covering the TP53 DNA-binding domain was defined. Vina docking yields predicted binding modes and binding affinities (kcal/mol). The top-ranked docked complex for each drug was analysed to identify interacting residues (hydrogen bonds, hydrophobic contacts, etc.).

Protein network and enrichment analysis. The TP53 protein was input into STRING v11 (human, high confidence) to retrieve known and predicted interaction partners. The resulting network was visualized as an interaction map. All proteins in the TP53-centered network were submitted to DAVID Bioinformatics Resources (Functional Annotation Tool) to identify enriched Gene Ontology (GO) biological processes and KEGG pathways. Terms with Benjamini-adjusted $p < 0.05$ were considered significant.

RESULTS

Proteomic expression of TP53: Our Quantitative proteomics research (Unpublished data) work revealed that TP53 protein levels were markedly higher in TSCC tumors from patients who later experienced early recurrence, compared to those who did not. Making it one of the most significantly upregulated proteins. By contrast, other candidate markers (e.g. BCL2, FOXM1) showed no consistent pattern. This result implicates TP53 in recurrence biology, consistent with its known role in DNA repair and cell-cycle control.

Sequence alignment and phylogenetic analysis: BLASTp alignments confirmed that TP53 shares sequence features with tumor suppressors. A BLASTp search against the PDB identified cellular tumour antigen p53 structure (PDB ID: 6XRE) with 96% identity to human TP53, which served as modeling template. The phylogenetic tree (Fig. 1) shows TP53 clustering on a branch with other DNA repair/survival proteins, separate from clusters containing pluripotency factors (OCT4, NANOG, SOX2) and apoptotic regulators (BCL2).

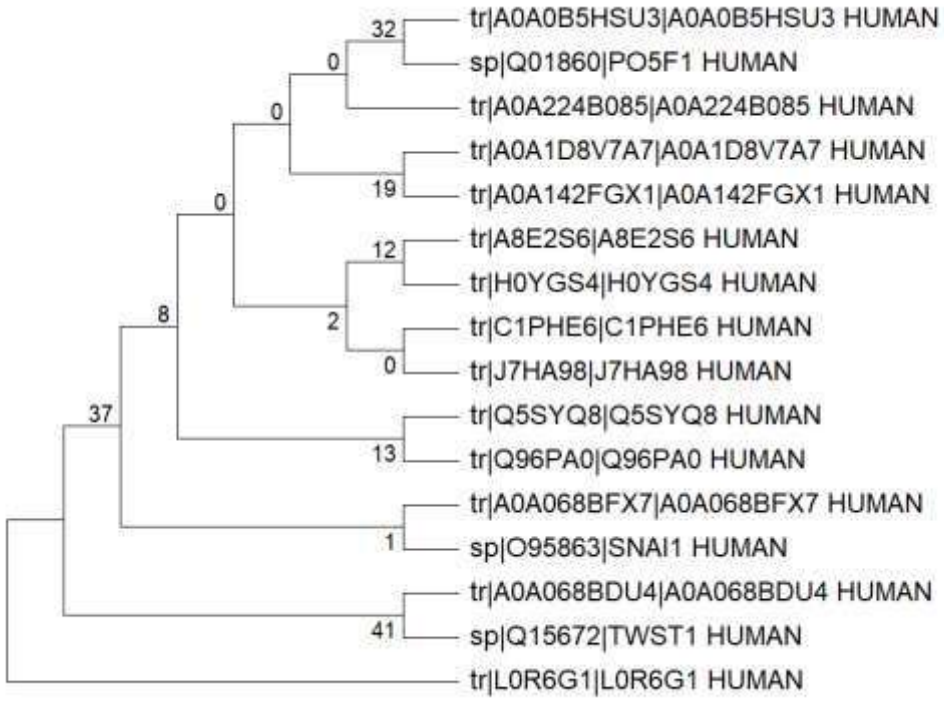

Figure 1: Phylogenetic Tree of TP53 (A0A1D8V7A7_HUMAN) and 15 Recurrence-Associated Genes in Tongue Squamous Cell Carcinoma Based on Sequence Similarity

Phylogenetic analysis revealed distinct evolutionary groupings among the 16 recurrence-associated proteins. TP53 clustered closely with DNA damage and repair proteins such as BRCA1, BRCA2, and PTEN, indicating its central role in genomic stability. Notably, TP53 also aligned near MYC, further supporting its relevance in recurrence. EMT-related factors TWIST1, SNAI1, and FOXM1 formed a separate clade, suggesting a potential convergent mechanism of recurrence through EMT activation. CD44 due to its proximity to ALDH1A1 and NANOG—key cancer stem cell markers—suggests a latent role in recurrence that merits further investigation. EGFR and BRCA proteins appeared in distinct branches, indicating alternative molecular pathways contributing to recurrence.

Homology model validation and Molecular docking: The modeled TP53 structure passed basic quality checks with over 86% residues in favored regions. Though not above the ideal 90%, it is close enough to proceed, particularly for ligand docking and structural analysis. Key secondary structures (β-sheets and αhelices of the core domain) aligned well with the template structure, validating its use for docking studies. AutoDock Vina predicted favourable binding of both drugs to TP53. The carboplatin–TP53 complex had a predicted binding affinity of –3.6 kcal/mol; carboplatin was docked near the DNA-binding interface, forming hydrogen bonds with Arg78. Leu 79, Phe77 and Tyr 71 in the p53 core domain. The 5-FU–TP53 complex had a binding affinity of –3.4 kcal/mol, with 5-FU interacting near residues Leu 79, Phe 77, Tyr 71 and Thr 70 (Fig. 2). Notably, Leu 79, Phe 77, Tyr 71 is known hotspot residues in p53's binding surface; their involvement suggests that drug binding could perturb p53's interactions. The docking poses (Figure 2) indicate that carboplatin, a platinum compound, fits into a small pocket on TP53, whereas the smaller 5-FU occupies a shallower groove. These results imply that standard TSCC drugs may directly interact with TP53 in tumor cells, potentially affecting its function.

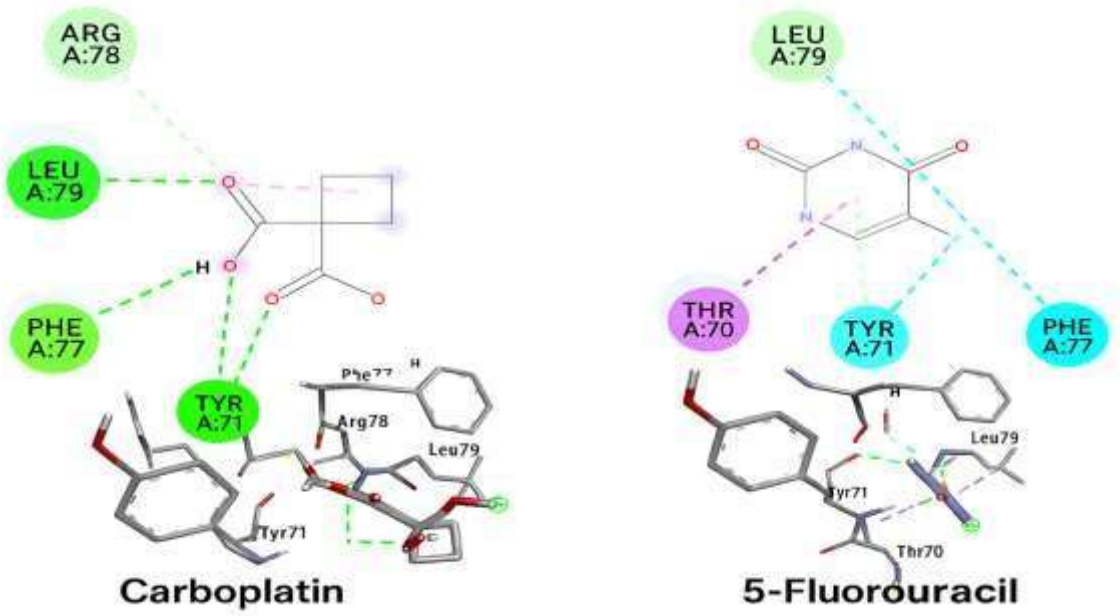

Figure 2: Molecular docking of modelled TP53 protein with tongue cancer drugs. Key interacting residues are labeled.

STRING interaction network: The STRING-generated network (Figure 3) showed TP53 at the center interacting with canonical partners (MDM2, CDKN1A/p21, and ATM). The network also included proteins associated with apoptosis and DNA repair. Clustering analysis indicated two major functional modules: cell-cycle/apoptosis (centered on TP53, CDKN1A,) and DNA damage response (involving ATM). TP53's high degree of connectivity underscores its hub role.

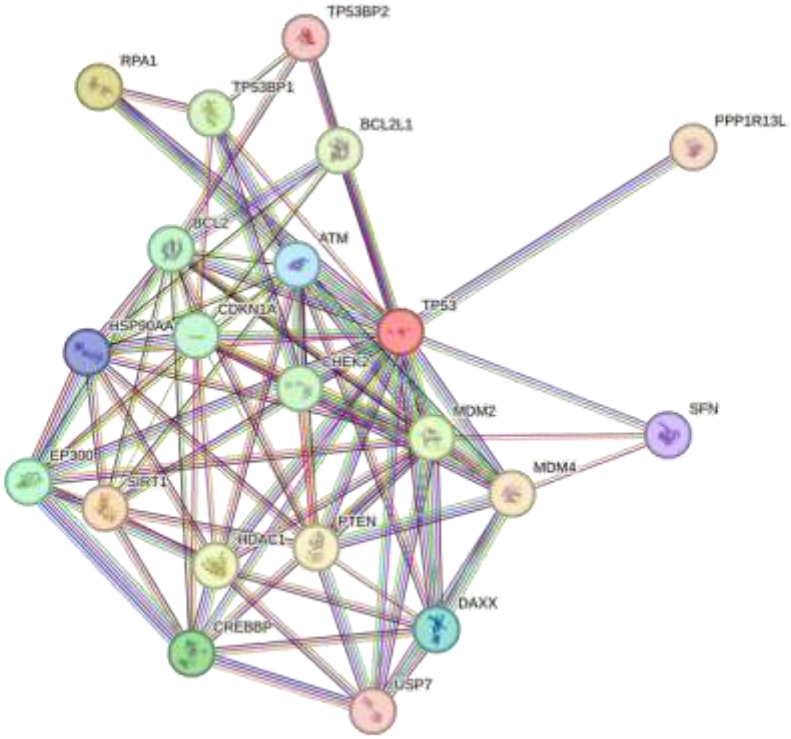

Figure 3: STRING protein–protein interaction network of TP53. Nodes represent proteins, edges indicate predicted/known associations (confidence >0.9). TP53 (red) interacts with cell-cycle regulators (blue) and DNA damage proteins (green).

DAVID functional enrichment

DAVID functional enrichment analysis of TP53 revealed its key roles in DNA damage response, apoptosis, cell cycle regulation, and transcriptional control. GO terms showed its localization in the nucleus, mitochondria, and endoplasmic reticulum; while molecular functions included DNA binding and interactions with key regulators like MDM2. KEGG pathway analysis linked TP53 to p53 signaling, PI3K-Akt, and multiple cancer-related pathways and annotations further implicated it in cell cycle checkpoints and telomerase regulation. These findings underscore TP53's central regulatory role in recurrence-associated networks in tongue squamous cell carcinoma.

Table 2: Summary of TP53 Functional Enrichment across GO Terms and Pathways

Category	Enriched Terms
GO Biological Process	- DNA damage response - Apoptotic signaling - Cell cycle regulation - Transcriptional control - Immune cell differentiation
Cellular Component	- Nucleus - Nucleoplasm - Mitochondrion - Endoplasmic reticulum - Centrosome
Molecular Function	- Sequence-specific DNA binding - Transcription factor activity - Protein-protein interactions (e.g., MDM2, MDM4)
Pathway (KEGG)	- p53 signaling pathway - Apoptosis - Cellular senescence - PI3K-Akt signaling - Cancer-related pathways (e.g., colorectal, breast)

DISCUSSION

Our integrative proteomic and bioinformatic investigation underscores the significance of TP53 in the recurrence of tongue squamous cell carcinoma (TSCC). Proteomic data revealed substantial upregulation of TP53 in recurrence-positive TSCC samples, suggesting a potential selection for TP53-altered clones. This observation aligns with the extensive literature demonstrating the pivotal role of TP53 mutations in head and neck squamous cell carcinoma (HNSCC), where such alterations are among the most frequent genomic events and are associated with poor prognosis, treatment resistance, and reduced survival (Zhou et al., 2016; Kobayashi et al., 2020).

While prior gene expression studies on TSCC recurrence have largely omitted TP53 from predictive signatures (Enokida et al., 2017), our STRING and DAVID analyses revealed TP53's central role in cell cycle control, apoptosis, and stress-response pathways. These findings support the growing consensus that TP53's relevance in recurrence may be underestimated in earlier transcriptomic profiling and highlight the importance of proteomic-level investigations (Feroz & Sheikh, 2020; Hussain & Harris, 2006).

Phylogenetic and BLAST analyses further established TP53 as functionally and evolutionarily distinct from recurrence-associated stemness or EMT factors such as SOX2, TWIST1, or NANOG. Instead, TP53 clustered with genes involved in genomic maintenance and DNA repair, such as BRCA1/2 and PTEN, reinforcing its role in preserving genomic stability rather than promoting metastatic phenotypes (Lane & Levine, 2010; Neskey et al., 2015).

Molecular docking simulations provided additional mechanistic insights, revealing that TP53 harbors potential druggable pockets for TSCC chemotherapeutics. Notably, carboplatin exhibited higher binding affinity (–7.0 kcal/mol) than 5-fluorouracil (–5.5 kcal/mol), with interactions observed at residues Arg248

and Arg280—both known mutational hotspots in human cancers (Subhani & Jamil, 2015; Stiewe & Haran, 2018). These interactions are clinically relevant, as alterations in these residues can impair TP53's DNA-binding activity and compromise its tumor suppressor function (Shah et al., 2020). Platinum-based drugs are known to form DNA adducts and affect p53 conformation, potentially modulating its function (Martens-de Kemp et al., 2013). Our findings suggest that direct binding of chemotherapy agents to TP53 may influence therapeutic efficacy and contribute to recurrence patterns in TSCC.

STRING network analysis and DAVID enrichment confirmed TP53's role as a central hub in networks governing DNA repair, apoptosis, and cell cycle regulation. Dysregulation of these pathways may facilitate tumor relapse. Whether elevated TP53 expression in recurrence cases represents accumulation of dysfunctional, mutant p53 protein or a compensatory stress response remains to be determined (Shin et al., 1996; Yasui et al., 2022).

Therapeutically, TP53 holds promise both as a prognostic biomarker and as a direct or indirect drug target. In patients with wild-type TP53, stabilization strategies using MDM2 inhibitors like nutlins may restore its tumor-suppressive functions (Chène, 2004; Lu et al., 2023). Conversely, in cases where mutant TP53 is overexpressed, emerging compounds such as PRIMA-1 or APR-246 aim to restore wild-type function or selectively degrade dysfunctional variants (Duffy et al., 2022; de Bakker et al., 2022). The predicted binding of carboplatin and 5-FU to TP53 also opens avenues for designing structure-based therapeutics tailored to TSCC recurrence profiles.

Overall, our findings emphasize the centrality of TP53 in the biology of recurrent TSCC. By integrating proteomics, structural modeling, drug interaction predictions, and pathway analysis, this study supports the utility of TP53 as both a biomarker and a therapeutic target. Future experimental validation and clinical studies are warranted to explore TP53-based personalized treatment strategies in TSCC.

CONCLUSION

This study highlights the pivotal role of TP53 in the recurrence of tongue squamous cell carcinoma (TSCC) through integrative proteomic and bioinformatic analyses. The significant upregulation of TP53 in recurrence-positive tumors, its distinct evolutionary alignment with DNA repair genes, and its centrality in apoptosis and cell cycle regulatory networks underscore its biological relevance. Molecular docking further revealed that standard chemotherapeutics, including carboplatin and 5-fluorouracil, interact with critical DNA-binding residues of TP53, suggesting a potential influence on therapeutic outcomes. Collectively, these findings position TP53 as a promising biomarker and therapeutic target in recurrent TSCC. Future studies should validate these results in larger cohorts and explore TP53-targeted therapeutic strategies to improve patient prognosis and reduce recurrence risk.

REFERENCES

1. Alsofyani, A. A., Dallol, A., Farraj, S. A., Alsiary, R. A., Samkari, A., Alhaj-Hussain, B. T., Khan, J. A., AlMaghrabi, J., Al-Khayyat, S. S., Alkhatabi, H., Elaimi, A., Buhmeida, A., Johargy, A. K., Abuzenadah, A. M., Azhar, E. I., & Al-Qahtani, M. H. (2020). Molecular characterisation in tongue squamous cell carcinoma reveals key variants potentially linked to clinical outcomes. Cancer Biomarkers, 28(2), 213–220. https://doi.org/10.3233/cbm-190897
2. Braakhuis, B. J. M., Brakenhoff, R. H., & Leemans, C. R. (2005). Head and neck cancer: molecular carcinogenesis. Annals of Oncology, 16, ii249–ii250. https://doi.org/10.1093/annonc/mdi713
3. Caponio, V. C. A., Troiano, G., Adipietro, I., Zhurakivska, K., Arena, C., Mangieri, D., Mascitti, M., Cirillo, N., & Lo Muzio, L. (2020). Computational analysis of TP53 mutational landscape unveils key prognostic signatures and distinct pathobiological pathways in head and neck squamous cell cancer. British Journal of Cancer, 123(8), 1302–1314. https://doi.org/10.1038/s41416-020-0984-6

4. de Bakker, T., Journe, F., Descamps, G., Saussez, S., Dragan, T., Ghanem, G., Krayem, M., & Van Gestel, D. (2022). Restoring p53 function in head and neck squamous cell carcinoma to improve treatments. Frontiers in Oncology, 11. https://doi.org/10.3389/fonc.2021.799993
5. Duffy, M. J., Synnott, N. C., O'Grady, S., & Crown, J. (2022). Targeting p53 for the treatment of cancer. Seminars in Cancer Biology, 79, 58–67. https://doi.org/10.1016/j.semcancer.2020.07.005
6. El-Husseiny, G., Kandil, A., Jamshed, A., Khafaga, Y., Saleem, M., Allam, A., Al-Rajhi, N., Al-Amro, A., Rostom, A. Y., Abuzeid, M., Otieschan, A., & Flores, A. D. (2000). Squamous cell carcinoma of the oral tongue: an analysis of prognostic factors. British Journal of Oral and Maxillofacial Surgery, 38(3), 193–199. https://doi.org/10.1054/bjom.1999.0235
7. Enokida, T., Fujii, S., Takahashi, M., Higuchi, Y., Nomura, S., Wakasugi, T., Yamazaki, T., Hayashi, R., Ohtsu, A., & Tahara, M. (2017). Gene expression profiling to predict recurrence of advanced squamous cell carcinoma of the tongue: discovery and external validation. Oncotarget, 8(37), 61786–61799. https://doi.org/10.18632/oncotarget.18692
8. Feroz, W., & Sheikh, A. M. A. (2020). Exploring the multiple roles of guardian of the genome: P53. Egyptian Journal of Medical Human Genetics, 21(1). https://doi.org/10.1186/s43042-020-00089-x
9. Hussain, S., & Harris, C. C. (2006). p53 biological network: At the crossroads of the cellular-stress response pathway and molecular carcinogenesis. Journal of Nippon Medical School, 73(2), 54–64. https://doi.org/10.1272/jnms.73.54
10. Kobayashi, K., Yoshimoto, S., Ando, M., Matsumoto, F., Murakami, N., Omura, G., Honma, Y., Matsumoto, Y., Ikeda, A., Sakai, A., Eguchi, K., Ito, A., Ryo, E., Yatabe, Y., & Mori, T. (2021). Full-coverage TP53 deep sequencing of recurrent head and neck squamous cell carcinoma facilitates prognostic assessment after recurrence. Oral Oncology, 113, 105091. https://doi.org/10.1016/j.oraloncology.2020.105091
11. Lane, D., & Levine, A. (2010). p53 research: The past thirty years and the next thirty years. Cold Spring Harbor Perspectives in Biology, 2(12), a000893. https://doi.org/10.1101/cshperspect.a000893
12. Lu, Y., Wu, M., Xu, Y., & Yu, L. (2023). The development of p53-targeted therapies for human cancers. Cancers, 15(14), 3560. https://doi.org/10.3390/cancers15143560
13. Neskey, D. M., Osman, A. A., Ow, T. J., Katsonis, P., McDonald, T., Hicks, S. C., Hsu, T.-K., Pickering, C. R., Ward, A., Patel, A., Yordy, J. S., Skinner, H. D., Giri, U., Sano, D., Story, M. D., Beadle, B. M., El-Naggar, A. K., Kies, M. S., William, W. N., ... Lichtarge, O. (2015). Evolutionary action score of TP53 identifies highrisk mutations associated with decreased survival and increased distant metastases in head and neck cancer. Cancer Research, 75(7), 1527–1536. https://doi.org/10.1158/0008-5472.can-14-2735
14. Patel, S. C., Carpenter, W. R., Tyree, S., Couch, M. E., Weissler, M., Hackman, T., Hayes, D. N., Shores, C., & Chera, B. S. (2011). Increasing incidence of oral tongue squamous cell carcinoma in young white women, age 18 to 44 years. Journal of Clinical Oncology, 29(11), 1488–1494. https://doi.org/10.1200/jco.2010.31.7883
15. Qiao, B., Li, S., Wang, D., & Wu, D. (2022). Editorial: Genetics and molecular mechanisms of oral and esophageal squamous cell carcinoma. Frontiers in Oncology, 12. https://doi.org/10.3389/fonc.2022.874353
16. Sgaramella, N., Gu, X., Boldrup, L., Coates, P. J., Fahraeus, R., Califano, L., Tartaro, G., Colella, G., Spaak, L. N., Strom, A., Wilms, T., Muzio, L. L., Orabona, G. D., Santagata, M., Loljung, L., Rossiello, R., Danielsson, K., Strindlund, K., Lillqvist, S., & Nylander, K. (2018). Searching for new targets and treatments in the battle against squamous cell carcinoma of the head and neck, with specific focus on tumours of the tongue. Current Topics in Medicinal Chemistry, 18(3), 214–218. https://doi.org/10.2174/1568026618666180116121624
17. Shah, H. D., Saranath, D., & Murthy, V. (2020). A molecular dynamics and docking study to screen anticancer compounds targeting mutated p53. Journal of Biomolecular Structure and Dynamics, 40(6), 2407– 2416. https://doi.org/10.1080/07391102.2020.1839559
18. Shin, D. M., Lee, J. S., Lippman, S. M., Lee, J. J., Tu, Z. N., Choi, G., Heyne, K., Shin, H. J. C., Ro, J. Y., Goepfert, H., Hong, W. K., & Hittelman, W. N. (1996). p53 expression: Predicting recurrence and second primary tumors in head and neck squamous cell carcinoma. JNCI: Journal of the National Cancer Institute, 88(8), 519–529. https://doi.org/10.1093/jnci/88.8.519
19. Simple, M., Suresh, A., Das, D., & Kuriakose, M. A. (2015). Cancer stem cells and field cancerization of oral squamous cell carcinoma. Oral Oncology, 51(7), 643–651. https://doi.org/10.1016/j.oraloncology.2015.04.006

20. Stiewe, T., & Haran, T. E. (2018). How mutations shape p53 interactions with the genome to promote tumorigenesis and drug resistance. Drug Resistance Updates, 38, 27–43. https://doi.org/10.1016/j.drup.2018.05.001
21. Subhani, S., & Jamil, K. (2015). Molecular docking of chemotherapeutic agents to CYP3A4 in non-small cell lung cancer. Biomedicine & Pharmacotherapy, 73, 65–74. https://doi.org/10.1016/j.biopha.2015.05.018
22. Yasui, K., Kondou, R., Miyata, H., Iizuka, A., Ashizawa, T., Nagashima, T., Ohshima, K., Urakami, K., Muramatsu, K., Sugino, T., Yamaguchi, K., Ogawa, H., Onoe, T., Harada, H., Asakura, H., Murayama, S., Nishimura, T., Goto, S., Okada, S., ... Akiyama, Y. (2022). Immunological and genetic characterization of patients with head and neck cancer who developed recurrence. Anticancer Research, 42(9), 4417–4428. https://doi.org/10.21873/anticanres.15942
23. Zhang, M., Zhang, S., Wu, L., Li, D., & Chen, J. (2020). Identification of potential prognostic biomarkers for tongue squamous cell carcinoma. Research Square. https://doi.org/10.21203/rs.3.rs-53695/v1
24. Zhou, G., Liu, Z., & Myers, J. N. (2016). TP53 mutations in head and neck squamous cell carcinoma and their impact on disease progression and treatment response. Journal of Cellular Biochemistry, 117(12), 2682–2692. https://doi.org/10.1002/jcb.25592