Transcription factor immunohistochemistry

Transcription factor immunohistochemistry is a laboratory technique used in histology and pathology to visualize and study the presence and localization of specific transcription factors within cells and tissues. Transcription factors are proteins that play a critical role in gene expression by binding to DNA and regulating the transcription of specific genes. By using immunohistochemistry, researchers and pathologists can better understand the distribution and activity of these transcription factors in different tissues and under various conditions.

Overview of the process

Sample Preparation: Tissue samples, often obtained from biopsy or surgical specimens, are embedded in paraffin wax or frozen in optimal cutting temperature (OCT) compound to preserve the tissue structure.

Sectioning: Thin sections (slices) of the tissue samples are cut using a microtome or cryostat, depending on whether the tissue is paraffin-embedded or frozen.

Antigen Retrieval: In many cases, the tissue sections are subjected to antigen retrieval methods. This step helps expose the target antigen (in this case, the transcription factor) by reversing the cross-linking that may have occurred during tissue fixation.

Blocking: The tissue sections are treated with blocking reagents to prevent nonspecific binding of antibodies. This ensures that the antibodies used in the assay bind specifically to the target transcription factors.

Primary Antibody Incubation: Tissue sections are incubated with primary antibodies specific to the transcription factor of interest. These antibodies are designed to bind to the transcription factor, marking its location in the tissue.

Washing: Excess primary antibodies are washed away to reduce background staining.

Secondary Antibody Incubation: The tissue sections are incubated with secondary antibodies that are conjugated to a marker, such as a fluorescent dye or an enzyme. These secondary antibodies bind to the primary antibodies, further enhancing the signal.

Washing: Any unbound secondary antibodies are washed away.

Detection: In the case of enzymatic detection, a substrate is applied, and an enzymatic reaction produces a visible signal, such as a colored precipitate. For fluorescent detection, no additional steps are required.

Counterstaining: In some cases, a counterstain (e.g., DAPI for nuclei) may be applied to visualize the overall tissue structure.

Microscopy: The tissue sections are examined under a microscope, and images are captured to analyze the location and intensity of the transcription factor staining within the tissue.

Transcription factor immunohistochemistry is a valuable tool in both research and clinical settings, as

it allows for the assessment of transcription factor expression and localization in various diseases, development, and physiological processes. This information can aid in the understanding of gene regulation and may have diagnostic or prognostic implications in certain medical conditions.

The application of transcription factor immunohistochemistry to pituitary neuroendocrine tumor (PitNET) assessment has allowed the identification of tumors that do not conform to a single lineage. Multilineage PIT1 and SF1 PitNETs are a rare and relatively newly described tumor subtype. These tumors express both transcription factors and may also express combinations of hormones corresponding to both lineages. Histological and clinical characteristics can vary, and overall clinical behavior and prognosis are not known. They describe the clinical outcomes and somatostatin receptor status (SSTR) of a series of nine cases identified from our cohort of pituitary tumors at Westmead Hospital. Eight PitNETs (88.9%) expressed growth hormone and caused acromegaly at presentation. Of the 7 macro tumors that caused acromegaly, one had cavernous sinus invasion. Ki 67 labeling index ranged from 0.6 to 3.6%. 88% of tumors that secreted excess growth hormone exhibited strong immunostaining for SSTR 2 and all tumors displayed weak immunoreactivity for SSTR5. In 62.5% of patients with acromegaly, the cure was achieved after surgical resection. Somatostatin receptor ligands resulted in clinical remission in cases where medical treatment was initiated. There was no new tumor recurrence or regrowth over an overall mean follow-up period of 62.5 months [1)].

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Fookeerah P, Varikatt W, Shingde M, Dexter MAJ, McLean M. Somatostatin receptor expression and clinical outcome of multilineage PIT1 and SF1 pituitary tumours. Endocr Connect. 2023 Sep 26;12(11):e230328. doi: 10.1530/EC-23-0328. Epub ahead of print. PMID: 37751411; PMCID: PMC10563595.