



Microwave Antigen Retrieval Technique

From Shi, Shan-Rong et al, "Antigen retrieval technique: A novel approach to immunohistochemistry on routinely processed tissue sections," in Gu, J, ed. ANALYTICAL MORPHOLOGY: THEORY, LIBRARY AND PROTOCOLS, Eaton Publishing, 1996.

H2550* Laboratory MW Processor (Energy Beam Sciences)

1. This MW processor with temperature readouts is available. Its power output is regulated by a temperature feedback mechanism and a timer, and it is capable of monitoring both temperature and heating time.
2. Fix a Coplin jar on a thick capboard or plastic plate and set it in the center portion of the turntable, filling the jar with distilled water or tap water. Set the temperature probe into the Coplin jar through a hole in the cap to measure the temperature.
3. Set all test jars around the central probe jar as closely as possible.
4. Turn on the MW processor. Set the temperature and the time at temperature as required.
5. Turn on the turntable, making sure that the jars are not moved by the probe and the table.
6. Start the MW heating; the timer is automatically controlled.
7. For heating at 100 degrees C, the heating time should be divided into 5-minute cycles
8. This MW processor is particularly useful for test battery (see below).

Test battery to develop an optimal protocol of AR for certain antibody tested (Shi, S.-R., et al: Cell Vision 2:2-17, 1995)

1. Ten slides numbered 1 to 10 are used for three pH values of AR solution and three different heating conditions as follows:

AR Solution	pH 1	pH6	pH10
Super-high temperature	#1	#4	#7
High temperature	#2	#5	#8
Mid-high temperature	#3	#6	#9

2. A tenth slide stands as a non-AR control.

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3. Super-high temperature: Place slides in the microwave oven at 100 degrees C for 4 x 5 min....Kigh temperature: Microwave at 100 degrees C for 10 min as in the original protocol. Mid-high temperature: Place in a temperature controlled MW oven...at 90 degrees C for 10 min. If possible, a H2550 MW processor may be used to maintain the heating conditions accurately, as listed above.
4. The use of an autostainer is recommended to perform IHC staining.
5. Evaluate the intensity for all 10 slides in order to identify the optimal protocol for the antibody under test.
6. Additional studies, including different AR solutions...may be used if necessary.

The of microwave energy as a means of pretreating formalin-fixed tissue is a technique protected by U.S. Patent No. 5,244,787 assigned to BioGenex Laboratories. Anyone seeking a license to practice this technology should contact BioGenex directly.