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| Gene | FISH probes | | | Immunohistochemistry | | | | |
| **Source** | **Probe** | **Methods** | **Source** | **Clone** | **Dilution** | **Methods** |
| *EGFR* | Leica biosystems | EGFR (7p11) / SE 7 (D7Z1) | Samples are cut from FFPE at 3 microns. Leave them overnight at 60 ºc   1. Deparaffined and hydrated 2. Wash with buffer 3. Pretreatment 10 minutes at 100ºc 4. Allow to temperate 15 minutes 5. Wash with buffer (2 times) 6. Digestion with pepsin for 7 minutes at 37ºc 7. Wash with buffer (2 times) 8. 70º alcohol 3 minutes 9. 85º alcohol 3 minutes 10. 100º alcohol 3 minutes 11. Hybridize in the hybridizer 12. Denaturalization 80º for 15 minutes 13. Hybridization at 37º overnight 14. Post-hybridation wash with astringency buffer at 65º for 10 minutes 15. Wash with buffer 16. Mount with DAPI and Vectashield | Dako products - Agilent | H11 | 1:200 | **Bond™ automated system**  Leica biosystems  bond™ III-polymer refine detection.  Samples are cut from FFPE at 3 microns. Leave them overnight at 60 ºc   1. Dewaxing with bond dewax solution 2. Wash with alcohol 3. Wash with bond wash solution (buffer) 4. Antigenic recovery enzyme 1 during 10 minutes at 100ºc 5. Wash with bond wash solution (buffer) 6. Peroxidase blocker 8 minutes 7. Wash with bond wash solution (buffer) 8. *EGFR* antibody 60 minutes 9. Wash with bond wash solution (buffer) 10. Primary post 8 minutes 11. Wash with bond wash solution (buffer) 12. Polymer 10 minutes 13. Wash with bond wash solution (buffer) 14. Wash with distilled water. 15. Dab(chromogen) 10 minutes 16. Wash with distilled water. 17. Hematoxylin 9 minutes 18. Wash with bond wash solution (buffer) 19. Wash with distilled water. 20. Dehydrate and mount. |
| *MDM2* | Leica biosystems | MDM2 (12q15) / SE 12 (D12Z3) | Vitro (Master Diagnóstica)  &  LifeSpan BioSciences | 1A7 | 1:200 |