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**Anti-Conjugated Tyramine  
RAT POLYCLONAL**

**Catalog Number:** AB-T072

**Example of ELISA protocol used to test conjugated tyramine:**

1. Coating of conjugated tyramine (15 $\mu$ g/ml) in maxisorp well plates (Nunc) with a solution of sodium carbonate buffer 0.05M (pH 9.6) containing sodium metabisulfite (SMB) 0.001M, during sixteen hours at 4°C.
2. Saturation of well plates with of a solution of PBS (pH 7.3) containing 2.5g/l of BSA (Acros), 0.05% Tween 20 (Acros) and SMB 0.001M during one hour at 37°C.
3. Wash with PBS Tween (two times).
4. Anti-conjugated tyramine antibodies will be diluted (1/1,000-1/5,000) in PBS containing 2.5g/l BSA, 10% of glycerol and SMB 0.001M, 200 $\mu$ l by well plate (incubating during 2 hours at 37°C).
5. Wash with PBS Tween (three times).
6. 200 $\mu$ l of peroxidase-labeled goat anti-rat (Jackson) diluted (1/5000) in a solution of PBS containing 2.5g/l BSA, 10% of glycerol, 0.5% of Tween and SMB 0.001M, will be applied by well plate (during one hour at 37°C).
7. Well plates will be rinsed with a PBS Tween (three times).
8. And finally the peroxidase will be developed by incubating 200 $\mu$ l by well plate of a citrate 0.1M/phosphate 0.2M (pH 5) solution containing 0.4% of OPD (Sigma) and 0.03% of hydrogen peroxide (Acros) for ten minutes in the dark, after that, we will stop the reaction by the addition of 50 $\mu$ l of 2M HCl.
9. The optical density will be measured at 492nm, to obtain the different values.