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Horia Hulubei National Institute for R&D in Physics and Nuclear Engineering

Process for obtaining immunosorbent trenbolone-3-carboxymethyloxim-ovalbumincarboxymethyl-cellulose (Tren-3-CMO-OVA-CMcellulose)

Patent Number: RO123130/09.12.2007

Abstract

The invention relates to a process for preparing the immunosorbent trenbolone-3-carboxymethylovalbumin-carboxymethyl-cellulose (Tren-3-CMO-OVA-CM-cellulose) used in affinity chromatography for purification of specific antibodies antitrenbolone from complex mixtures of immunoglobulins or polyclonal antitrenbolone antisera and can be applied in pharmaceutical industry. The high specific surface immunosorbent is used in high sensitivity ELISA technique for determination of very low concentrations of food contaminants.

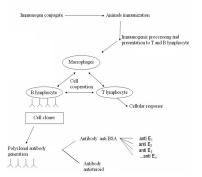
ELISA (Enzyme Linked Immunosorbent Assay) is a biochemical technique used mainly in immunology to detect the presence of an antibody or an antigen in a sample. An unknown amount of antigen is affixed to a surface, and a specific antibody linked to an enzyme is applied over the surface so that it can bind to the antigen. An enzymatic substrate is added that the enzyme can convert to detectable signal - a color change in a chemical substrate which indicates the quantity of antigen in the sample.

Trenbolone $(17\beta$ -Hydroxyestra-4,9-11-trien-3-one) is a steroid used by veterinarians on livestock to increase muscle growth and appetite. Trenbolone compounds have a binding affinity for the androgen receptor five times as high as that of testosterone. Once metabolized, the drugs have the effect of increasing nitrogen uptake by muscles, leading to an increase in the rate of protein synthesis. It also has the secondary effects of stimulating appetite, reducing the amount of fat being deposited in the body and decreasing the rate of catabolism. Short-term side effects on human include high blood pressure, insomnia, night sweats, increased aggression.

Technology stage

The obtained product can be used in ELISA immunochemical technique for dosing of trenbolone steroid from biological samples. It was validated in ELISA dosing technique.

Obtaining the antitrenbolone antibodies



Applications

- ELISA kits for detection of androgenic growth promoter, trenbolone from food samples which leads to increased quality of life by using uncontaminated food;
- Human and veterinary endocrinology: quantitative determination of concentrations of anabolic substance;
- Pharmaceutical industry.

Advantages

- using carboxymethylcellulose (CM-celullose), protein (ovalbumine, bovine serum albumine) can be linked covalent by activating the carboxy group's CM-cellulose by carbodiimide and that coupling to free amino groups of lysine in the structure of the protein with peptide bond formation;
- using the same coupling reagent, carbodiimide, in the same way is activated carboxy derivative of the steroid trenbolone and links the free amino groups of protein with the formation of immunosorbent trenbolone-ovalbumin-CMcellulose;
- resulting immunosorbent presents a large specific surface and affinity of homologue antibody antitrenbolone, it can be used in affinity chromatography to purify these antibody.

Contact

Dan D. ENACHE Tel.: 0040214042303 Fax: 0040214574210 Email: <u>dan.enache@nipne.ro</u>