Claims

1. A process for the enzymatic regeneration of the redox cofactors NAD^+/NADH and $NADP^+$/NADPH in a one-pot reaction, wherein, as a result of at least two further enzymatically catalyzed redox reactions proceeding in the same reaction batch (product-forming reactions), the redox cofactor NAD^+/NADH accumulates in its reduced form as NADH and, the redox cofactor $NADP^+$/NADPH in its oxidized form as $NADP^+$, characterized in that

   a) in the regeneration reaction which reconverts the NADH into its original oxidized form, oxygen or pyruvate is reduced by means of a NADH oxidase or a lactate dehydrogenase, respectively, and

   b) in the regeneration reaction which reconverts the $NADP^+$ into its original reduced form 2-propanol or malate is oxidized by means of an alcohol dehydrogenase or a malate dehydrogenase, respectively.

2. A process according to claim 1, characterized in that oxidation reaction(s) and reduction reaction(s) take place on the same substrate (molecular backbone).

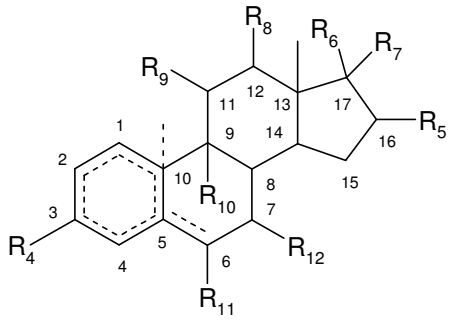
3. A process according to any of claims 1 or 2, characterized in that oxidation reaction(s) and reduction reaction(s) proceed chronologically parallel.

4. A process according to any of claims 1 to 3, characterized in that, in the regeneration reaction which reconverts $NADP^+$ to NADPH, 2-propanol is oxidized to acetone by means of an alcohol dehydrogenase.

5. A process according to any of claims 1 to 3, characterized in that, in the regeneration reaction which reconverts the NADH to NAD^+, pyruvate is reduced to lactate by means of a lactate dehydrogenase.

6. A process according to claim 5, characterized in that, in the regeneration reaction which reconverts $NADP^+$ to NADPH, malate is oxidized to pyruvate and CO_2 by means of a malate dehydrogenase.

7. A process according to any of claims 1 to 5, characterized in that it is used for performing at least one oxidation reaction and at least one reduction reaction, respectively, in the same reaction batch on compounds of general formula



III

wherein

R_4 denotes hydrogen, a methyl group, a hydroxy group or an oxo group,

R_5 denotes hydrogen, a hydroxy group, an oxo group or a methyl group,

R_6 denotes hydrogen or a hydroxy group,

R_7 denotes hydrogen, $–COR_{13}$, wherein R_{13} is a C_1-C_4 alkyl group which is unsubstituted or substituted with a hydroxy group, or a C_1-C_4 carboxy alkyl group which is substituted, in particular with a hydroxy group, or unsubstituted,

or R_6 and R_7 together denote an oxo group,

R_8 denotes hydrogen, a methyl group, a hydroxy group or an oxo group,

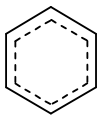
R_9 denotes hydrogen, a methyl group, a hydroxy group or an oxo group,

R_{10} denotes hydrogen, a methyl group or halogen,

R_{11} denotes hydrogen, a methyl group, a hydroxy group, an oxo group or halogen, and

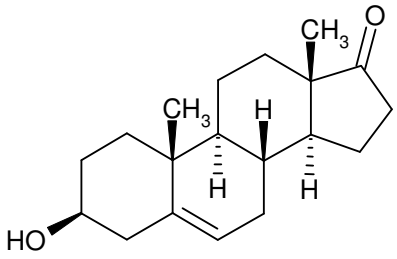
R_{12} denotes hydrogen, a hydroxy group, an oxo group or a methyl group,

wherein the structural element



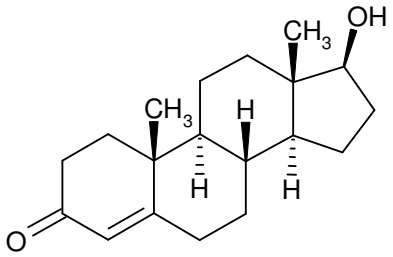
denotes a benzene ring or a ring comprising 6 carbon atoms and 0, 1 or 2 C-C-double bonds; in particular wherein the substrate(s) is/are provided at a concentration of <5% (w/v) in the reaction batch for the reduction reaction(s) involved in the formation of the product.

8. A process according to claim 7, characterized in that it is used for the conversion of dehydroepiandrosterone (DHEA) of formula



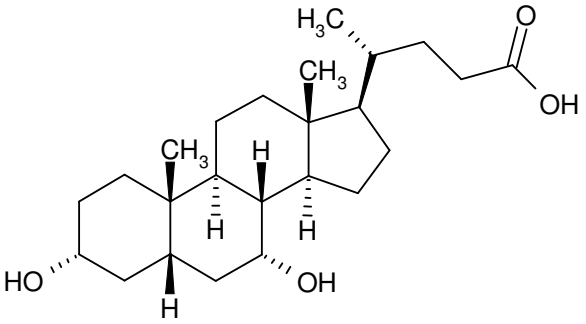
VII

into testosterone of formula



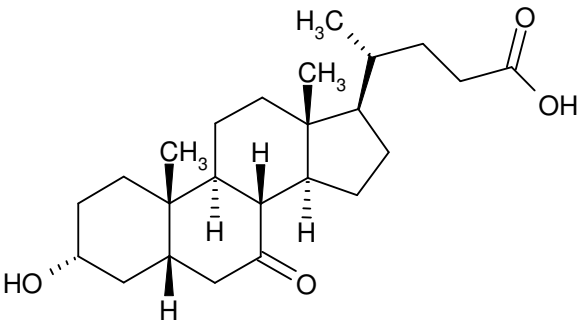
VIII

9. A process according to claim 7, characterized in that it is used for the enzymatic epimerization of 3α,7α-dihydroxy-5β-cholanic acid (chenodeoxycholic acid) of formula



IV

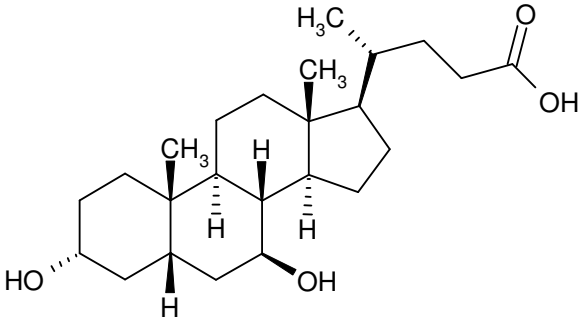
into ketolithocholic acid of formula



V

by oxidation,

and into the stereoisomeric hydroxy compound 3α,7β-dihydroxy-5β-cholanic acid (ursodeoxycholic acid) of formula

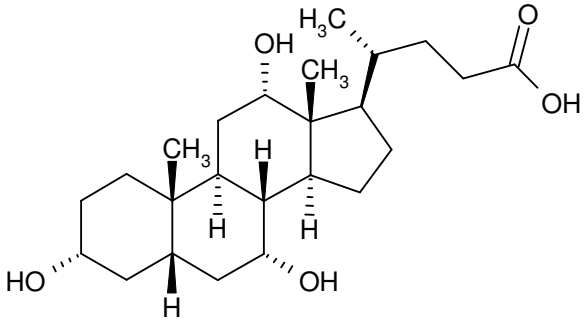


VI

by subsequent reduction,

using two opposite stereospecific hydroxysteroid dehydrogenases, in particular whereby the oxidation reaction is catalyzed by a 7α-hydroxysteroid dehydrogenase from *E. coli*; and/or the reduction reaction is catalyzed by a 7β-hydroxysteroid dehydrogenase from *Ruminococcus torques*.

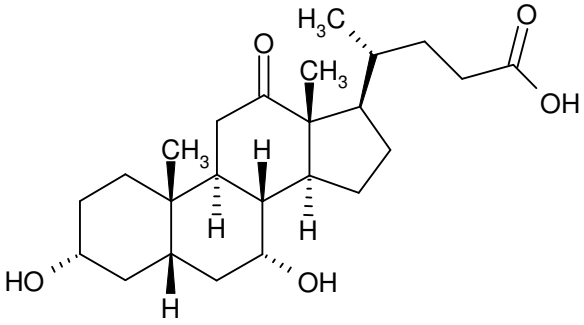
10. A process according to claim 7, characterized in that it is used for the enzymatic epimerization of 3α,7α,12α-trihydroxy-5β-cholanic acid (cholanic acid) of formula



IX

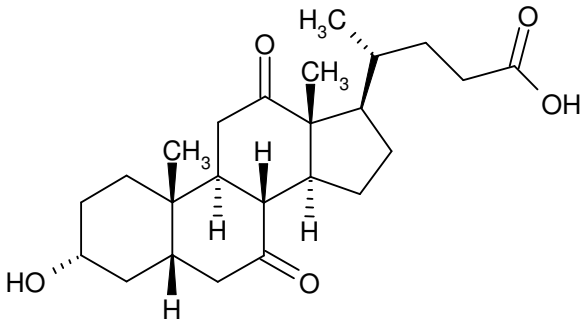
either

A) via oxidation to obtain 3α,7α-dihydroxy-12-oxo-5β-cholanic acid (12-oxo-CDC) of formula



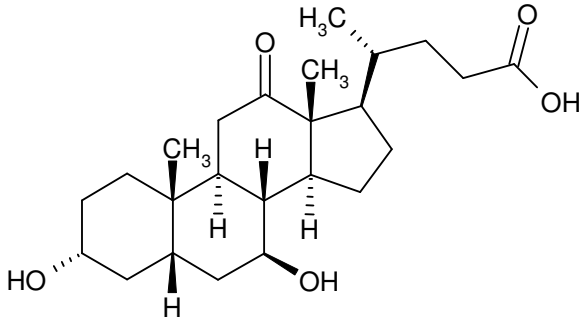
X

which is further reacted to obtain 3α-hydroxy-7,12-dioxo-5β-cholanic acid (12oxo-KLC) of formula



XI

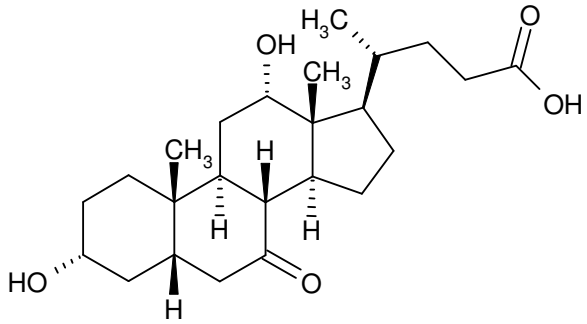
and subsequent reduction to the stereoisomeric hydroxy compound 3α,7β-dihydroxy-12-oxo-5β-cholanic acid (12-keto-ursodeoxycholanic acid) of formula



XII

or

B) via oxidation to obtain 3α,12α-dihydroxy-7-oxo-5β-cholanic acid of formula

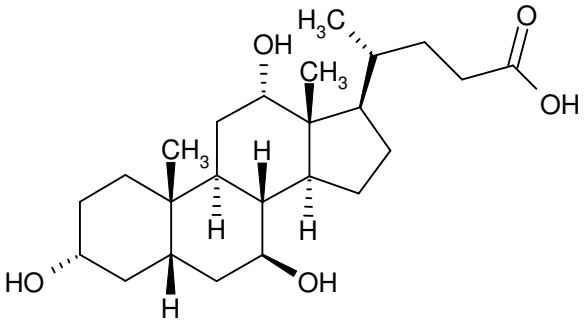


XIII

followed by enzymatic oxidation to obtain 3α-hydroxy-7,12-dioxo-5β-cholanic acid(12oxo-KLC) of formula XI, and subsequent reduction to obtain the stereoisomeric hydroxy compound 3α,7β-dihydroxy-12-oxo-5β-cholanic acid (12-keto-ursodeoxycholanic acid) of formula XII,

or

C) via oxidation to obtain 3α,12α-dihydroxy-7-oxo-5β-cholanic acid of formula XIII, followed by enzymatic reduction to obtain 3α,7β,12α-triydroxy-5β-cholanic acid of formula



XIV

and subsequent oxidation to obtain the stereoisomeric hydroxy compound 3α,7β-dihydroxy-12-oxo-5β-cholanic acid (12-keto-ursodeoxycholanic acid) of formula XII;

using 3 stereospecific hydroxysteroid dehydrogenases, 2 of which have opposite stereospecifity.

11. A process according to any of claims 1 to 6, characterized in that it is used for the isomerization of C_5- or C_6-sugars, in particular for the isomerization of glucose via reduction to sorbitol and subsequent oxidation to fructose.

12. A process according to any of claims 1 to 11, characterized in that the substrate(s) for the oxidation reaction(s) involved in the formation of a product is/are provided in the reaction batch at a concentration of at least 5% (w/v) and more, in particular 7% (w/v) and more, in particular 9% (w/v) and more.