

The question, whether “fully human” mAbs such as derived from phage display libraries or transgenic mice are less immunogenic than humanized mAbs cannot be answered yet, because full immunogenicity data are available for just two human mAbs. An anti-tumor necrosis factor mAb, developed from phage-displayed human libraries induced antibody responses in 12% of patients - at the higher end of the incidence of anti-antibody responses of the humanized antibodies (Hwang, W. Y., et al. (2005). *Methods*, 36, 3-10).

Evaluation of the immunogenicity of the first registered human mAb generated by the transgenic approach demonstrated that mAb treatment resulted in the generation of antibodies in approximately 5.5% of treated cancer patients (Jakobovits, A., et al. (2007). *Nat. Biotechnol.* 25, 1134., Lofgren, J. A., et al. (2007). *J. Immunol.* 178, 7467).

SUMMARY

In one aspect, the disclosure provides a transgenic non-human mammal or avine animal comprising, integrated in its genome, a rearranged nucleic acid molecule encoding a rearranged human immunoglobulin light chain variable region, wherein said light chain variable region is capable of pairing with at least two different heavy chain variable regions, and wherein the transgenic non-human mammal or avine animal is capable of producing antibodies comprising heavy chain variable regions that can be used in the development of therapeutic antibodies.

In another aspect, the disclosure provides a method for producing a desired antibody comprising exposing a non-human mammal or avine animal according to the disclosure to an antigen such that an antibody response is induced and isolating the antibodies specific for the antigen.

In another aspect, the disclosure provides a method for producing a desired antibody comprising exposing a non-human mammal or avine animal according to the disclosure to an antigen such that an antibody

response is induced and isolating cells producing such antibodies, culturing and harvesting said antibodies.

In another aspect, the disclosure provides a method for producing a desired antibody comprising exposing a non-human mammal or avine
5 animal according to the disclosure to an antigen such that an antibody response is induced and isolating a nucleic acid encoding at least part of such an antibody, inserting said nucleic acid or a copy or a derivative thereof in an expression cassette and expressing said antibody in a host cell.

10 In another aspect, the disclosure provides a progeny of a transgenic non-human mammal or avine animal according to the disclosure, the progeny comprising, integrated in its genome, a rearranged nucleic acid molecule encoding a rearranged human immunoglobulin light chain variable region, wherein said light chain variable region is capable of
15 pairing with at least two different heavy chain variable regions.

In another aspect, the disclosure provides a cell that is isolated from a transgenic non-human mammal or avine animal according to the disclosure, the cell comprising integrated in its genome, a rearranged nucleic acid molecule encoding a rearranged human immunoglobulin light
20 chain variable region, wherein said light chain variable region is capable of pairing with at least two different heavy chain variable regions.

DESCRIPTION OF THE INVENTION

A need therefore remains for a method and means for producing
25 antibodies that are specific for their targets, but are less immunogenic. According to the invention the reduction of immunogenicity is at least partially achieved by providing a transgenic non-human mammal comprising, at least in its B cell lineage, a nucleic acid encoding at least an immunoglobulin light chain or heavy chain, wherein the heavy- or light
30 chain encoding sequence is provided with a means that renders it resistant to DNA rearrangements and/or somatic hypermutations, preferably such a

non-human animal is a rodent, more specifically a mouse. The nucleic acid preferably encodes a human, human-like or humanized immunoglobulin chain.

- 5 In the remainder of the specification mice are typically used as examples of the non-human mammals. The transgenic non-human mammalian hosts are capable of mounting an immune response to an antigen, where the response produces antibodies having primate, particularly human, variable regions. Various transgenic hosts may be

Claims.

1. A transgenic non-human mammal or avine animal comprising,
integrated in its genome, a rearranged nucleic acid molecule encoding a
5 rearranged human immunoglobulin light chain variable region, wherein
said light chain variable region is capable of pairing with at least two
different heavy chain variable regions, and wherein the transgenic non-
human mammal or avine animal is capable of producing antibodies
comprising heavy chain variable regions that can be used in the
10 development of therapeutic antibodies.
2. A transgenic non-human mammal according to claim 1, which
is a rodent.
3. A transgenic non-human mammal according to claim 2, which
is a mouse.
- 15 4. A transgenic non-human mammal or avine animal according to
any one of claims 1-3, wherein said nucleic acid molecule further encodes an
animal light chain constant region.
5. A transgenic non-human mammal or avine animal according to
claim 4, wherein the integration is in a locus that is resistant to silencing.
- 20 6. A transgenic non-human mammal or avine animal according to
claim 5, wherein the integration is in the Rosa-locus.
7. A transgenic non-human mammal or avine animal according to
any one of claims 1-6, wherein the light chain encoding nucleic acid is
provided with a means that allows expression of said nucleic acid
25 essentially limited to cells of B cell lineage.
8. A transgenic non-human mammal or avine animal according to
claim 7, wherein the light chain encoding nucleic acid is provided with a
means that allows expression of the light chain encoding nucleic acid
predominantly during a certain stage of the development of B cells.
- 30 9. A transgenic non-human mammal or avine animal according to
claim 8, wherein said means comprises a promoter selected from the group

of CD19, CD20, μ HC, VpreB1, VpreB2, VpreB3, λ 5, Ig α , Ig β , κ LC, λ LC, and BSAP (Pax5).

10. A transgenic non-human mammal or avine animal according to claim 8 or 9, wherein said means comprises a cre-lox system or the like.

5 11. A transgenic non-human mammal or avine animal according to any one of claims 1-10, wherein at least one endogenous loci encoding an endogenous light chain is functionally silenced.

12. A transgenic non-human mammal or avine animal according to any one of claims 1-11, wherein an endogenous κ light chain locus is
10 functionally silenced.

13. A transgenic non-human mammal or avine animal according to any one of claims 1-12, wherein the sequence of the light chain encoding sequence is a human vk sequence.

14. A transgenic non-human mammal or avine animal according to
15 claim 13 wherein the light chain encoding sequence is a germline-like sequence.

15. A transgenic non-human mammal or avine animal according to claim 14, wherein the light chain encoding sequence is a germline sequence.

16. A transgenic non-human mammal or avine animal according to
20 claim 15, wherein the germline sequence is based on O12.

17. A transgenic non-human mammal or avine animal according to claim 1 wherein the light chain encoding nucleic acid comprises in 5'-3' direction: a vk promoter, a human leader, and a human V gene.

18. A transgenic non-human mammal or avine animal according to
25 claim 17, wherein the light chain encoding nucleic acid comprises in 5'-3' direction: a VK promoter, a human leader, a human V gene, a MoEki enhancer and a rat constant region (K).

19. A transgenic non-human mammal or avine animal according to claim 18, wherein the light chain encoding nucleic acid comprises in 5'-3'
30 direction: a VK promoter, a human leader, a human V gene, a MoEki enhancer, a rat constant region (K) and a (truncated) MoE κ 3' enhancer.

20. A method for producing a desired antibody comprising exposing a non-human mammal or avine animal according to any one of claims 1-19 to an antigen such that an antibody response is induced and isolating the antibodies specific for the antigen.

5 21. A method for producing a desired antibody comprising exposing a non-human mammal or avine animal according to any one of claims 1-19 to an antigen such that an antibody response is induced and isolating cells producing such antibodies, culturing and harvesting said antibodies.

10 22. A method according to claim 21, further comprising immortalizing said cells after the culturing step and prior to harvesting said antibodies.

23. A method for producing a desired antibody comprising exposing a non-human mammal or avine animal according to any one of
15 claims 1-19 to an antigen such that an antibody response is induced and isolating a nucleic acid encoding at least part of such an antibody, inserting said nucleic acid or a copy or a derivative thereof in an expression cassette and expressing said antibody in a host cell.

24. Progeny of a transgenic non-human mammal or avine animal
20 according to any one of claims 1-19, the progeny comprising, integrated in its genome, a rearranged nucleic acid molecule encoding a rearranged human immunoglobulin light chain variable region, wherein said light chain variable region is capable of pairing with at least two different heavy chain variable regions.

25 25. A cell that is isolated from a transgenic non-human mammal or avine animal according to any one of claims 1-19, the cell comprising integrated in its genome, a rearranged nucleic acid molecule encoding a rearranged human immunoglobulin light chain variable region, wherein
30 said light chain variable region is capable of pairing with at least two different heavy chain variable regions.