

SChLAP1, three exosomes genes (ERG, PCA3, SPDEF), a urinary two gene combination (HOXC6 and DLX1), as well as any other prostate cancer biomarker known to the skilled person (see e.g., McGrath et al., *Prostate Int.* 2016, 4(4): 130-135).

5 In a particular embodiment of the invention, a method according to the present invention comprises the determination of the level of progastrin over time in samples from a patient who has been or is being treated for prostate cancer.

The characteristics of the embodiments of the invention will become further apparent from the following detailed description of examples below.

FIGURE LEGENDS

10 **Figure 1**

Progastrin concentration was measured in 40 plasma samples from prostate cancer patients and 119 plasma samples from healthy donors using the ELISA Kit DECODE Lab (capture antibody: Mab14, detection antibody: anti-hPG polyclonal).

EXAMPLES

15 **Example 1: Detection of plasmatic progastrin concentration using polyclonal antibodies**

Plasma progastrin levels were quantified by ELISA through the use of two specific anti-progastrin antibodies: capture antibodies are coated on the wells of the plate, whereas revelation antibodies are used to detect progastrin and mediates
20 revelation of the signal.

In the present example, quantification is based on the ELISA method which allows, through the use of a substrate whose reaction emits light, to assign a value proportional to the luminescence amount of antibodies bound to the antigen retained by capture antibodies.

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CLAIMS

1. A method for the *in vitro* diagnosis of prostate cancer in a subject, comprising the steps of:

- a) contacting a biological sample from said subject with at least one
5 progastrin-binding antibody, or an antigen-binding fragment thereof,
- b) detecting the binding of said progastrin-binding antibody, or an antigen-binding fragment thereof to progastrin in said sample, wherein said binding indicates the presence of prostate cancer in said subject,

wherein said biological sample is chosen among: blood, serum, and plasma,
10 and

wherein said antibody binding to progastrin is a monoclonal antibody chosen from the group consisting of:

- A monoclonal antibody comprising a heavy chain comprising CDR-H1, CDR-H2 and CDR-H3 of amino acid sequences SEQ ID N° 4, 5 and 6, respectively,
15 and a light chain comprising CDR-L1, CDR-L2 and CDR-L3 of amino acid sequences SEQ ID N° 7, 8 and 9, respectively,
- A monoclonal antibody comprising a heavy chain comprising CDR-H1, CDR-H2 and CDR-H3 of amino acid sequences SEQ ID N° 10, 11 and 12, respectively, and a light chain comprising CDR-L1, CDR-L2 and CDR-L3 of
20 amino acid sequences SEQ ID N° 13, 14 and 15, respectively,
- A monoclonal antibody comprising a heavy chain comprising CDR-H1, CDR-H2 and CDR-H3 of amino acid sequences SEQ ID N° 16, 17 and 18, respectively, and a light chain comprising CDR-L1, CDR-L2 and CDR-L3 of amino acid sequences SEQ ID N° 19, 20 and 21, respectively,
- A monoclonal antibody comprising a heavy chain comprising CDR-H1, CDR-H2 and CDR-H3 of amino acid sequences SEQ ID N° 22, 23 and 24, respectively, and a light chain comprising CDR-L1, CDR-L2 and CDR-L3 of
25 amino acid sequences SEQ ID N° 25, 26 and 27, respectively,
- A monoclonal antibody comprising a heavy chain comprising CDR-H1, CDR-H2 and CDR-H3 of amino acid sequences SEQ ID N° 28, 29 and 30,
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respectively, and a light chain comprising CDR-L1, CDR-L2 and CDR-L3 of amino acid sequences SEQ ID N° 31, 32 and 33, respectively, and

- A monoclonal antibody comprising a heavy chain comprising CDR-H1, CDR-H2 and CDR-H3 of amino acid sequences SEQ ID N° 34, 35 and 36, respectively, and a light chain comprising CDR-L1, CDR-L2 and CDR-L3 of amino acid sequences SEQ ID N° 37, 38 and 39, respectively, and

wherein said antigen binding fragment thereof comprises the 6 CDRs of the antibody it derives from.

2. The method of claim 1, wherein step b) further comprises determining the concentration of progastrin and wherein a concentration of progastrin at least 10 pM in said biological sample is indicative of the presence of prostate cancer in said subject.

3. The method of claim 2, comprising the further steps of:

- c) determining a reference concentration of progastrin in a reference sample,
- d) comparing the concentration of progastrin in said biological sample with said reference concentration of progastrin,
- e) determining, from the comparison of step d), the presence of prostate cancer.

4. The method of any of claims 1 to 3, wherein said antibody, or antigen-binding fragment thereof, is selected among N-terminal anti-progastrin monoclonal antibodies and C-terminal anti-progastrin monoclonal antibodies.

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5. The method of any one of claims 1 to 4, wherein said antibody is selected in the group consisting of:

- A monoclonal antibody comprising a heavy chain of amino acid sequence SEQ ID N° 41 and a light chain of amino acid sequence SEQ ID N° 42;
- A monoclonal antibody comprising a heavy chain of amino acid sequence SEQ ID N° 43 and a light chain of amino acid sequence SEQ ID N° 44;

- A monoclonal antibody comprising a heavy chain of amino acid sequence SEQ ID N° 45 and a light chain of amino acid sequence SEQ ID N° 46;
 - A monoclonal antibody comprising a heavy chain of amino acid sequence SEQ ID N° 47 and a light chain of amino acid sequence SEQ ID N° 48;
 - 5 • A monoclonal antibody comprising a heavy chain of amino acid sequence SEQ ID N° 49 and a light chain of amino acid sequence SEQ ID N° 50; and
 - A monoclonal antibody comprising a heavy chain of amino acid sequence SEQ ID N° 51 and a light chain of amino acid sequence SEQ ID N° 52.
- 10 6. The method of any one of claims 1 to 5, wherein said biological sample is contacted with a first antibody or an antigen-binding fragment thereof, which binds to the C-terminal part of progastrin, and with a second antibody or an antigen-binding fragment thereof, which binds to the N-terminal part of progastrin, wherein the C-terminal part of progastrin is defined by SEQ ID No 3 and wherein the N-terminal part of progastrin is defined by SEQ ID No 2.
- 15 7. The method of claim 6, wherein the first progastrin-binding antibody, or an antigen-binding fragment thereof binds an epitope within the C-terminus of progastrin, wherein said epitope is selected in the group consisting of:
- an amino acid sequence corresponding to residues 71-74 of SEQ ID No 1,
 - 20 • an amino acid sequence corresponding to residues 69-73 of SEQ ID No 1,
 - an amino acid sequence corresponding to residues 71-80 of SEQ ID No 1,
 - an amino acid sequence corresponding to residues 76-80 of SEQ ID No 1, and
 - an amino acid sequence corresponding to residues 67-74 of SEQ ID No 1.
- 25 8. The method of claim 6 or 7, wherein the second progastrin-binding antibody, or an antigen-binding fragment thereof binds an epitope within the N-terminus of progastrin, wherein said epitope is selected in the group consisting of:
- 30 • an amino acid sequence corresponding to residues 10-14 of SEQ ID No 1,
 - an amino acid sequence corresponding to residues 9-14 of SEQ ID No 1,
 - an amino acid sequence corresponding to residues 4-10 of SEQ ID No 1,

- an amino acid sequence corresponding to residues 2-10 of SEQ ID No 1, and
 - an amino acid sequence corresponding to residues 2-14 of SEQ ID No 1.
- 5 9. The method of any one of claims 6 to 8, wherein said progastrin-binding antibody, or an antigen-binding fragment thereof is a monoclonal antibody produced by the hybridoma deposited at the CNCM, Institut Pasteur, 25-28 rue du Docteur Roux, 75724 Paris CEDEX 15, France, on 27 December 2016, under reference I-5158.
- 10 10. The method of any one of claims 6 to 9, wherein said second progastrin-binding antibody, or an antigen-binding fragment thereof is a polyclonal antibody binding an epitope within the N-terminus of progastrin or a monoclonal antibody comprising a heavy chain comprising the following three CDRs, CDR-H1, CDR-H2 and CDR-H3 of amino acid sequences SEQ ID N°16, 17 and 18, respectively, and a light chain comprising the following three CDRs, CDR-L1, CDR-L2 and CDR-L3 of amino acid sequences SEQ ID N°19, 20 and 21, respectively.
- 15 11. The method of any one of claims 6 to 11, wherein the first antibody is bound to an insoluble or partly soluble carrier and the second antibody is labelled with a detectable moiety.
- 20 12. The method of any one of claims 1 to 11, wherein said biological sample is plasma, and wherein a concentration of progastrin of at least 10 pM is indicative of the presence of prostate cancer in said subject.
- 25 13. Use of a progastrin-binding antibody, or an antigen-binding fragment thereof, as in any one of claims 1 to 11 for the in vitro diagnosis of prostate cancer.