Abstract: A mouse model and uses thereof for detecting, treating, characterizing, and diagnosing various diseases are described.
A. **CLASSIFICATION OF SUBJECT MATTER**

**IPC(8)** - G01 33/00; C40B 30/06; C07H 21/04 (201 2.01 )

**USPC** - 800/3; 506/1 0; 536/24.5

According to International Patent Classification (IPC) or to both national classification and IPC

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B. **FIELDS SEARCHED**

Minimum documentation searched (classification system followed by classification symbols)

**USPC:** 800/3; 506/10; 536/24.5

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

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Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

PubWEST (PGP;USPT;USOC;EPAB;JPAB);

Google; PubMed: miR-29, transgenic, mouse, B cells, acute, lymphoblastic, lymphoma, leukemia, V sub.H, Igh-E.mu., TCL1

GenCore 6.3: SEQ ID NO:1

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C. **DOCUMENTS CONSIDERED TO BE RELEVANT**

<table>
<thead>
<tr>
<th>Category</th>
<th>Citation of document, with indication, where appropriate, of the relevant passages</th>
<th>Relevant to claim No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>X</td>
<td>SANTANAM et al. Chronic lymphocytic leukemia modeled in mouse by targeted miR-29 expression. Proc. Natl Acad Sci. USA. 21 June 2010, 107(27):12210-12215; abstract; pg 12210, para 1, 6; pg 12211 1, para 2-4; pg 12214, para 2-3</td>
<td>1-12, 27-28, 31-33, 35-37</td>
</tr>
</tbody>
</table>

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Date of the actual completion of the international search

30 January 2012 (30.01.2012)

Date of mailing of the international search report

05 MAR 2012

Name and mailing address of the ISA/US

Mail Stop PCT, Attn: ISA/US, Commissioner for Patents

P.O. Box 1450, Alexandria, Virginia 22313-1450

Facsimile No. 571-272-3201

Authorized officer:

Lee W. Young

PCT Helpdesk: 571-272-4300

PCT OSP: 571-272-7774
<table>
<thead>
<tr>
<th>Box No. I</th>
<th>Nucleotide and/or amino acid sequence(s) (Continuation of item 1c of the first sheet)</th>
</tr>
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<tbody>
<tr>
<td>1.</td>
<td>With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of a sequence listing filed or furnished:</td>
</tr>
<tr>
<td>a.</td>
<td>(means)</td>
</tr>
<tr>
<td></td>
<td>[ ] on paper</td>
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<tr>
<td></td>
<td>[x] in electronic form</td>
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<td>b.</td>
<td>(time)</td>
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<td>[ ] in the international application as filed</td>
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<td></td>
<td>[x] together with the international application in electronic form</td>
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<td></td>
<td>[ ] subsequently to this Authority for the purposes of search</td>
</tr>
<tr>
<td>2.</td>
<td>[ ] In addition, in the case that more than one version or copy of a sequence listing has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.</td>
</tr>
<tr>
<td>3.</td>
<td>Additional comments:</td>
</tr>
</tbody>
</table>
Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [ ] Claims Nos.:___
because they relate to subject matter not required to be searched by this Authority, namely:

2. [ ] Claims Nos.:___
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out. Specifically:

3. [x] Claims Nos.:___
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

This application contains the following inventions or groups of inventions which are not so linked as to form a single general inventive concept under PCT Rule 13.1.

Group I: claims 1-12, 27-28, 31-38, drawn to a transgenic animal whose genome comprises: a nucleic acid construct comprising at least one transcriptional regulatory sequence capable of directing expression to B cells operably linked to a nucleic acid sequence encoding miR-29.

Group II: claim 13, drawn to a transgenic animal whose genome comprises a nucleic acid construct comprising a nucleic acid sequence encoding miR-29, wherein the sequence is operably linked to a VH promoter and to a iH-Ef.1 enhancer, wherein miR-29 is expressed in immature and mature B cells of the animal.

(Continued on extra sheet) 

1. [ ] As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. [ ] As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of additional fees.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:__

4. [x] No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims: it is covered by claims Nos.: 1-12, 27-28, 31-38

Remark on Protest

[ ] The additional search fees were accompanied by the applicant’s protest and, where applicable, the payment of a protest fee.

[ ] The additional search fees were accompanied by the applicant’s protest but the applicable protest fee was not paid within the time limit specified in the invitation.

[ ] No protest accompanied the payment of additional search fees.

Form PCT/ISA/2 10 (continuation of first sheet (2)) (July 2009)
Continuation of Box III - Observations where unity of producing is lacking:

Group III: claim 14, drawn to a method of producing animals having a lymphoproliferative disorder comprising the steps of: a) obtaining white blood cells from a transgenic animal whose genome comprises: a nucleic acid construct comprising at least one transcriptional regulatory sequence capable of directing expression to B cells operably linked to a nucleic acid sequence encoding miR-29; b) counting the cells; and, c) injecting a number of the cells into a recipient animal syngeneic with the transgenic animal, wherein the number of the cells so injected is effective to produce a lymphoproliferative disorder in the recipient animal.

Group IV: claims 15-19, 39-43, drawn to a method of determining the ability of a therapeutic modality to affect a lymphoproliferative disorder by a) providing a first transgenic animal whose genome comprises: a nucleic acid construct comprising at least one transcriptional regulatory sequence capable of directing expression to B cells operably linked to a nucleic acid sequence encoding miR-29; b) administering the therapeutic modality to the first transgenic animal; c) performing an analysis of the population of B cells in the transgenic animal; d) providing a control animal, wherein the control animal is a second transgenic animal whose genome comprises: a nucleic acid construct comprising at least one transcriptional regulatory sequence capable of directing expression to B cells operably linked to a nucleic acid sequence encoding miR-29, wherein the control animal does not receive the therapeutic modality; e) performing an analysis of the population of B cells in the control animal; and, f) comparing the analysis of step e) with the analysis of step c), wherein the ability of the therapeutic modality to affect a lymphoproliferative disorder is evidenced by a difference in the B cell population between the first transgenic animal and the control animal.

Group V: claims 20-26 and 29, drawn to a transgenic mouse whose genome comprises a nucleic acid sequence encoding a human B-CLL, wherein the sequence is operably linked to a VH promoter and to a IgH-Emu enhancer, wherein the transgenic mouse develops an expanded population of CD5+ B cells compared to a control mouse.

Group VI: claim 30, drawn to a method for evaluating the efficacy of a therapeutic agent used in the treatment of chronic lymphocytic leukemia, comprising determining whether miR-29a is up-regulated, wherein up-regulation of miR-29a is indicative of indolent human B-CLL as compared with aggressive BCLL and normal CD19+ B cells.

The inventions listed as Groups I-VI do not relate to a single general inventive concept under PCT Rule 13.1 because, under PCT Rule 13.2, they lack the same or corresponding special technical features for the following reasons:

The inventions of Groups I-V do not include the inventive concept of a method for evaluating the efficacy of a therapeutic agent used in the treatment of chronic lymphocytic leukemia, as required by Group VI.

The inventions of Groups I-III and V-VI do not include the inventive concept of a method of producing animals having a lymphoproliferative disorder, as required by Group III, or a method of determining the ability of a therapeutic modality to affect a lymphoproliferative disorder by a) providing a first transgenic animal whose genome comprises: a nucleic acid construct comprising at least one transcriptional regulatory sequence capable of directing expression to B cells operably linked to a nucleic acid sequence encoding miR-29, as required by Group III.

The inventions of Group I do not include the inventive concept of a transgenic animal whose genome comprises a nucleic acid construct comprising a nucleic acid sequence encoding miR-29, wherein the sequence is operably linked to a VH promoter and to a IgH-Emu enhancer, wherein miR-29 is expressed in immature and mature B cells of the animal, as required by Group II, or a transgenic mouse whose genome comprises a nucleic acid sequence encoding a human B-CLL, wherein the sequence is operably linked to a VH promoter and to a IgH-Emu enhancer, wherein the transgenic mouse develops an expanded population of CD5+ B cells compared to a control mouse, as required by Group V.

The inventions of Groups I-II, IV-V share the technical feature of a transgenic animal of claim 1. However, this shared technical feature does not represent a contribution over prior art as being anticipated by an article titled "Chronic lymphocytic leukemia modeled in mouse by targeted miR-29 expression" by Santanana, et al. (PNAS ePUB 21 June 2010, 107(27):12210-12215) (hereinafter "Santanana") that discloses said transgenic animal (Abstract, "To study the role of miR-29 in B-CLL, we generated Emu-miR-29 transgenic mice overexpressing miR-29 in mouse B cells": pg 3, para 2."... we reported development and characterization of a Teh-driven mouse model of CLL (15, 20, 21) and showed that miR-29 can target TCL1 expression... In this mouse model, the TCL1 ORF (lacking 3' UTR) was under the control of a VH promoter-Igh-Emu enhancer... To determine if transgenic miR-29 expression can accelerate CLL in Emu-TCL1 transgenic mice, we crossed Emu-miR-29 and Emu-TCL1 transgenic mice. Emu-miR-29/Emu-TCL1 mice and their Emu-TCL1 littersmates were killed at -8 mo of age and analyzed"). As said transgenic animal was known in the art at the time of the invention, this cannot be considered a special technical feature that would otherwise unify the groups.

Groups I-VI therefore lack unity under PCT Rule 13 because they do not share a same or corresponding special technical feature.