

**Infection with *Helicobacter pylori* presenting the *vacA* s2m2 haplotype is strongly associated with protection against gastric cancer**

**Short title:** *H. pylori* genotypes in pre and cancerous gastric lesions

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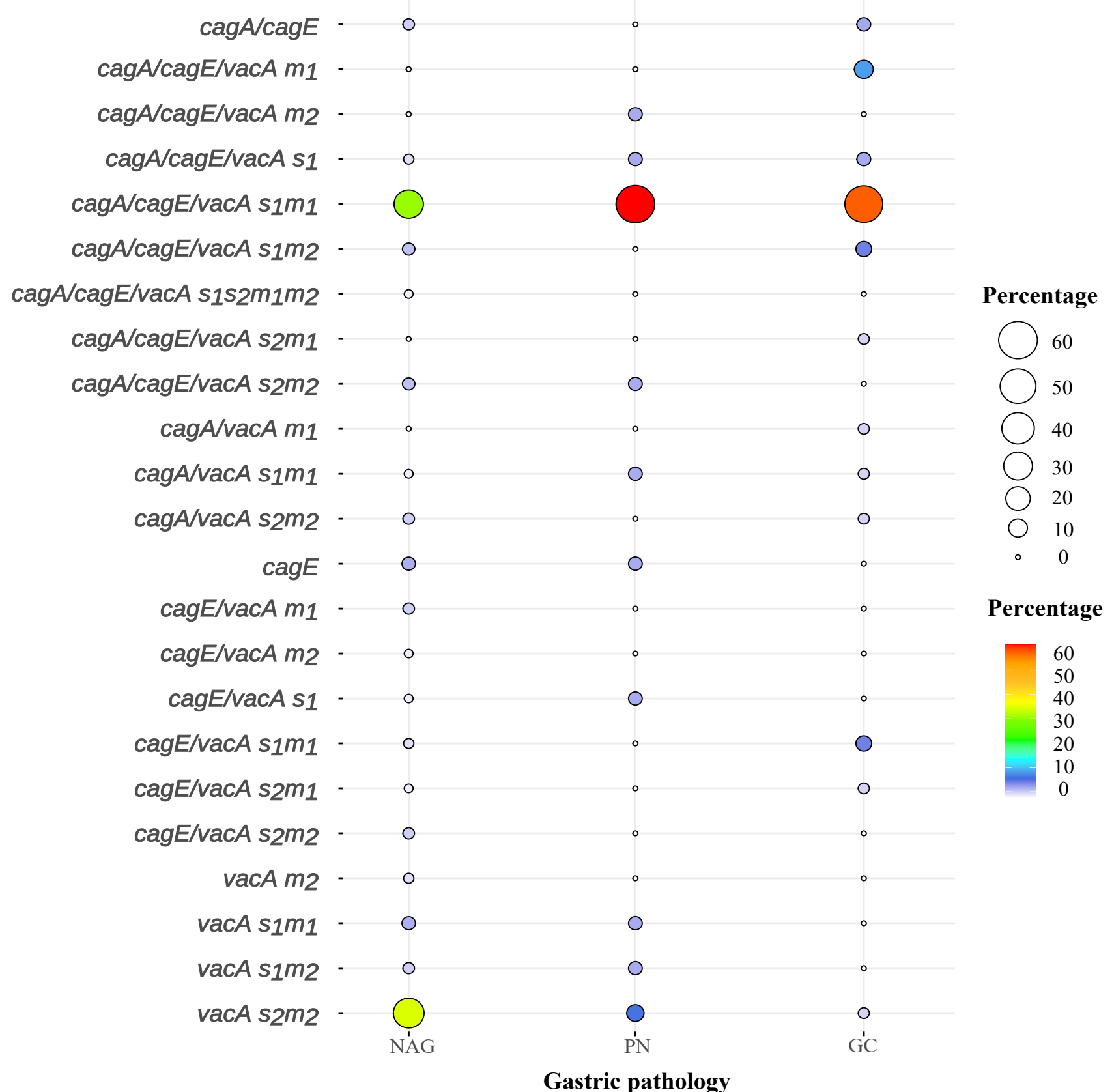
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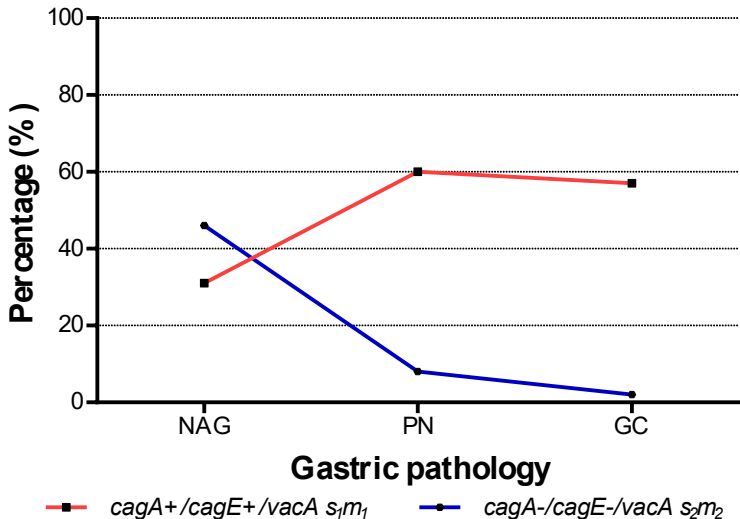
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### Supplementary Files:

- **Supplementary Figure 1.** *H. pylori* Genotypes identified in the patients infected (n = 205).
- **Supplementary Figure 2.** Percent of *cagA*<sup>+</sup>/*cagE*<sup>+</sup>/*vacA* *s*<sub>1</sub>*m*<sub>1</sub> and *cagA*<sup>-</sup>/*cagE*<sup>-</sup>/*vacA* *s*<sub>2</sub>*m*<sub>2</sub> *H. pylori* strains in infected patients.
- **Supplementary Table 1.** Sequence of the primers used to partially amplify of the *cagA*, *cagE* and *vacA* genes.
- **Supplementary Table 2.** Prevalence of *H. pylori* genotypes *vacA* according to gastric pathology of infected patients.
- **Supplementary Table 3.** Prevalence of *H. pylori* genotypes according to gastric pathology of infected patients.



**Supplementary Figure 1.** *H. pylori* Genotypes identified in the patients infected (n = 205).



**Supplementary Figure 2.** Percent of *cagA*+/*cagE*+/*vacA* *s*<sub>1</sub>*m*<sub>1</sub> and *cagA*-/*cagE*-/*vacA* *s*<sub>2</sub>*m*<sub>2</sub> *H. pylori* strains in infected patients.

**Supplementary Table 1.** Sequence of the primers used to partially amplify of the *cagA*, *cagE* and *vacA* genes.

| Genes                               | Primers   | Sequence primer (5'-3')  | Product length (pb) | References                             |
|-------------------------------------|-----------|--------------------------|---------------------|----------------------------------------|
| 16S ADNr                            | act-1     | CTTGCTAGAGTGCTGATTA      | 537                 | López A. et al., 2009. <sup>40</sup>   |
|                                     | act-2     | TCCCACACTCTAGAATAGT      |                     |                                        |
| cagA                                | cagA-F    | TTGACCAACAACCACAAACCGAAG | 183                 |                                        |
|                                     | cagA-R    | CTTCCCTTAATTGCGAGATTCC   |                     |                                        |
| cagE                                | cagE-F    | TGCTGATACGATTAGAGA       | 126                 | Quiroga A. et al., 2005. <sup>33</sup> |
|                                     | cagE-R    | TAGTCCCTTAGTGATGAT       |                     |                                        |
| vacA s <sub>1</sub> /s <sub>2</sub> | vacAs12-F | ATGGAAATACAACAAACACAC    | s1 : 259            | Hou P., et al., 2000. <sup>43</sup>    |
|                                     | vacAs12-R | CTGCTTGAATGCGCCAAAC      | s2 : 286            |                                        |
| vacA m <sub>1</sub>                 | vacAm1-F  | GGTCAAAATCGGTCATGG       | m1 : 570            |                                        |
|                                     | vacAm1-R  | CCATTGGTACCTGTAGAAAC     | m2 : 645            |                                        |
| vacA m <sub>2</sub>                 | vacAm2-F  | GGAGCCCCAGGAAACATTG      | m1=290              | López A. et al., 2009. <sup>40</sup>   |
|                                     | vacAm2-R  | CATAACTAGCGCCTTGAC       | m2=352              |                                        |
| β-globin                            | pco3      | CAACTTCATCCACGTTCCACC    | 110                 |                                        |
|                                     | pco4      | ACACAAACTGTGTTCACTAGC    |                     |                                        |

**Supplementary Table 2.** Prevalence of *H. pylori* genotypes *vacA* according to gastric pathology of infected patients.

| Haplotypes of signal and medium regions of <i>vacA</i> gene | Population<br>n <sup>†</sup> (%) | Gastric pathology |           |           |
|-------------------------------------------------------------|----------------------------------|-------------------|-----------|-----------|
|                                                             |                                  | NACG<br>n(%)      | PN n(%)   | GC n(%)   |
| $s_1 m_1$                                                   | 98 (47.8)                        | 49 (37.4)         | 17 (68.0) | 32 (65.3) |
| $s_1 m_2$                                                   | 11 (5.4)                         | 7 (5.3)           | 1 (4.0)   | 3 (6.1)   |
| $s_2 m_1$                                                   | 3 (1.5)                          | 1 (0.8)           | 0 (0.0)   | 2 (4.1)   |
| $s_2 m_2$                                                   | 61 (29.7)                        | 56 (42.7)         | 3 (12.0)  | 2 (4.1)   |
| $s_1 s_2 m_1 m_2$ <sup>‡</sup>                              | 1 (0.5)                          | 1 (0.8)           | 0 (0.0)   | 0 (0.0)   |
| Others ( $s_1, s_2, m_1, m_2$ )                             | 20 (9.7)                         | 9 (6.9)           | 3 (12.0)  | 8 (16.3)  |
| Negative                                                    | 11 (5.4)                         | 8 (6.1)           | 1 (0.0)   | 2 (4.1)   |
| <b>Total</b>                                                | <b>205</b>                       | <b>131</b>        | <b>25</b> | <b>49</b> |

<sup>†</sup>n(%): number and percent in each category

<sup>‡</sup>Co-infection detected

**Supplementary Table 3.** Prevalence of *H. pylori* genotypes according to gastric pathology of infected patients.

| Pathology | Number of<br><i>H. pylori</i><br>genotypes | <i>H. pylori</i> Genotype (%)                 |                                               |                    |
|-----------|--------------------------------------------|-----------------------------------------------|-----------------------------------------------|--------------------|
|           |                                            | <i>cagA</i> +/ <i>cagE</i> +/ <i>vacAs1m1</i> | <i>cagA</i> -/ <i>cagE</i> -/ <i>vacAs2m2</i> | Others             |
| NAG       | 19                                         | 41 (31.3%)                                    | 46 (35.1%)                                    | 44 (33.6%)         |
| PN        | 10                                         | 15 (60%)                                      | 2 (8%)                                        | 8 (32%)            |
| GC        | 12                                         | 28(57.1%)                                     | 1 (2%)                                        | 20 (40.9%)         |
| NAG vs PN | p-value                                    | 0.006102                                      | 0.00709                                       | 0.887537           |
|           | OR                                         | 3.2927 (1.36-7.95)                            | 0.1607 (0.03-0.71)                            | 0.9305 (0.37-2.32) |
| NAG vs GC | p-value                                    | 0.001499*                                     | 0.000703*                                     | 0.458318           |
|           | OR                                         | 2.9268(1.49-5.75)                             | 0.066 (0.001-0.5)                             | 0.6824 (0.24-1.88) |

n(%): number and percent in each category

\* p-value <0.05