Supplementary tables

**Article title:** TGFβ pathway deregulation and abnormal phospho-SMAD2/3 staining in hereditary cerebral hemorrhage with amyloidosis-Dutch type

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Supplementary Table 1  list of antibodies used in this study

| Reference | Name or clone | Antigen /Epitopes recognized | Host | Dilution IHC | Company & Type |
|-----------|--------------|------------------------------|------|-------------|----------------|
| 3101S<sup>a</sup> | pSMAD2/3 | Phospho-SMAD2 (Ser465/467)/SMAD3 (Ser423/425) | Rb (pc) | 1:500 or 1:1000 +TSA | Cell Signaling |
| 3108<sup>a</sup> | pSMAD2/3 | Phospho-SMAD2 (Ser465/467)/SMAD3 (Ser423/425) | Rb (mc) | 1:100 | Cell Signaling |
| sc-11769-R<sup>a</sup> | pSMAD2/3 | Phospho-SMAD2 (Ser465/467)/SMAD3 (Ser423/425) | Rb (pc) | 1:500 or 1:1000 +TSA | Santa Cruz |
| 9515 | SMAD4 | SMAD4 | Rb (pc) | 1:100 | Cell Signaling |
| A1474 | 6E10 | Aβ protein [1-17] | Ms (mc) | 1:1000 | Sigma |
| A1349 | 4G8<sup>b</sup> | Aβ protein [17-24] | Ms (mc) | 1:1000 | Sigma |
| G3893 | GA5 | Glial Fibrillar Acidic Protein (GFAP) | Ms (mc) | 1:5000 | Sigma |
| MAB364 | HM-2 | Microtubule-associated protein 2 (MAP-2) | Ms (mc) | 1:2000 | Chemicon |
| BR-003 | AT-8 | Human PHF-tau, phosphorylated at Ser202 and Thr205 | Ms (mc) | 1:2000 | Innogenetics |
| mab1510 | Ubi-1 | Ubiquitin | Ms (mc) | 1:3000 | Millipore |
| ab6586 | Collagen IV | Collagen IV native protein | Rb (pc) | 1:200<sup>c</sup> | Abcam |
| ab11575 | Laminin | Laminin native protein | Rb (pc) | 1:100<sup>d</sup> | Abcam |
| M0851 | SMA (IA4) | Smooth muscle actin | Ms (mc) | 1:2000 | Dako |

Rb Rabbit, Ms Mouse, pc polyclonal, mc monoclonal

<sup>a</sup> the present study was conducted with #3101S antibody; sc-11769-R recognizing the same epitope gave exact identical pattern (Supplementary Figure 1C). #3108, a monoclonal version of #3101S, also stained the pSMAD2/3 granules although the signal was weaker (Supplementary Figure 1D).

<sup>b</sup> data not shown, identical to 6E10 staining

<sup>c</sup> no antigen retrieval

<sup>d</sup> proteinase K antigen retrieval
### Supplementary Table 2 Occurrence of pSMAD2/3 deposits in occipital cortex of HCHWA-D subjects and clinical details

| Code | Age | Gender | pSMAD2/3 angiopathic vessels\(^a\) | Perivascular pSMAD2/3 granules\(^b\) | Delay last ICH to death | Interval 1st stroke-death | Previous stroke | Cause of death |
|------|-----|--------|-----------------------------------|-------------------------------------|------------------------|--------------------------|-----------------|---------------|
| H7   | 71  | M      | 77% * +/−                         | No major bleeding                   | 26 y                   | No major bleeding         | hematemesis     |               |
| H6   | 61  | M      | 70% * +/−                         | 8 y                                 | 15 y                   | 5                        | pneumonia       |               |
| H4   | 55  | F      | 72% +                            | 3 d (operated)                     | 3 d                    | 0                        | ICH             |               |
| H5   | 50  | M      | 46% +                            | n.a.                               | 6 y                    | 2                        | ICH             |               |
| H1   | 48  | M      | 36% ++                           | 4 d                                 | 3 y                    | 1                        | ICH             |               |
| H2   | 57  | M      | 3% −                             | 17 d (remission)                   | 9 y                    | 2                        | pneumonia       |               |
| H3   | 53  | F      | 0% +/−                           | 1 d                                 | 1 d                    | 0                        | ICH             |               |
| H8   | 51  | M      | 43% +++                          | 3 d                                 | 4 y                    | 1                        | ICH             |               |
| H11  | 81  | F      | 39% − \(^#\)                     | No major bleeding                   | 23 y                   | No major bleeding         | pneumonia       |               |
| H9   | 67  | F      | 84% * − \(^#\)                   | unknown                             | 21 y                   | 3 (1 ischemic)           | suffocation     |               |
| H10  | 71  | M      | 63% − \(^#\)                     | 1 d                                 | 17 y                   | 3                        | ICH             |               |

ICH: intracerebral hemorrhage, y years, d day(s), n.a. not available

\(^a\) percentage from average grading occipital cortex, \(^*\) capCAA involved

\(^b\) ranked based on single slide observation (+/−: scarcely found, + to +++: occurrence of finding),

\(^#\) DAB procedure only (although visible with DAB staining; the presence of perivascular granules is more easily assessed in immunofluorescence and preferably with TSA enhancement)