The single-locus model invoked in the present genetic analysis is, however, insufficient to explain any genetic linkage between HWE and febrile convulsions unless differences in genetic penetrance are involved in the appearance of these particular disease phenotypes[13]. Alternatively, it is possible that febrile seizures represent a genetically heterogeneous multigenic disorder; it might then be necessary to invoke the influence of linked loci, modifier loci, and/or environmental factors to account for the co-occurrence of these two syndromes within the same family as was observed in two of these lineages.

We are currently exploring more complex multigenic models that could explain the possible genetic link between HWE and its related syndromes. These analyses would, of course, require substantiation through examination of a much larger number of lineages. We hope that we would be able to discover more instances of HWE in the other parts of the country and identify pedigrees multiply affected by it and its associated epileptic syndromes. Given the paucity of detailed family records so far, however, we are also developing a rat model system to conduct classical and molecular genetic studies on HWE. Such studies should together provide an insight into the genetic basis of HWE, which is essential if we are to understand its mode of inheritance and ætiology, design specific therapies, develop a knowledge base for preventive genetic counseling, and thus, be better able to manage these fairly common, but troubling, epileptic syndromes.

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ACKNOWLEDGEMENTS. We thank Prof. M. N. Srinivas who first drew our attention to the high degree of consanguineous marriages among Muslims, and to Dr Niranjan V. Joshi for his comments on earlier versions of this paper.

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Maintenance of callus growth during subculturing is a genotype-dependent response in rice: Mature seed-derived callus from IR 54 rice cultivar lacks culturability

Following the finding that modified *Agrobacterium tumefaciens*-mediated transformation system works at high-efficiency for stable genetic transformation of rice[1], there is a great deal of activity as well as success in production of transgenic rice plants for varied applications[2]. A common protocol to achieve rice transformation through this approach involves co-cultivation of competent *A. tumefaciens* cells (harbouring the gene of interest) with calli pieces derived from the scutellar portion of mature seeds[1,3]. This method warrants that mature seed-derived calli pieces must be able to survive a series of subcultures, in order to (i) produce and maintain sufficient amounts of calli; (ii) subject calli to different concentrations of selection agent; (iii) check over-growth of *A. tumefaciens* cells on the culture plates following co-cultivation for a limited period and then washing off of the excess bacterial cells; (iv) subject calli to various

pre-treatments (such as with abscisic acid) for inducing high-level regeneration response; (v) transfer calli to regeneration medium, etc[3]. For the large-scale exploitation of the transgenic rice technology, it is therefore imperative that diverse rice cultivars must withstand and respond favourably to different treatments during the tissue culture experiments. It has been shown that anther culturability of rice is significantly variable among rice species, subspecies or varieties[4]. The regeneration response in rice is also found to be variable amongst different genotypes[5–7]. In the course of optimizing for a genetic transformation of different rice cultivars in our laboratory, we encountered the problem that mature seed-derived calli from IR 54 rice cultivar fail to withstand the subculturing treatment. On the other hand, there was no such problem with culturability of seed-derived calli from Pusa Basmati 1, Basmati 370 and Taipei 309 rice cultivars.

Seeds of different rice cultivars were obtained from diverse sources [Taipei 309 and IR 54, International Rice Research Institute (IRRI), Philippines; Basmati 370 and Pusa Basmati 1, Indian Agricultural Research Institute (IARI), New Delhi] and were multiplied at IARI. Mature seeds of the above cultivars were dehusked, surface-sterilized with 70% ethanol for 30–45 s and 0.1% $HgCl_2$ for 20 min and rinsed five times with sterile distilled water (SDW). Seeds of Taipei 309 and Pusa Basmati 1 cultivars were soaked overnight in dark and were cultured on NB medium[8] containing 2 mg l^{-1} 2,4-D (NB callus induction medium, Figure 1 *a*) at 26 ± 1°C, in the dark[3]. After 5–8 days, the scutellar portions excised from these seeds were inoculated onto fresh NB callus induction medium (Figure 1 *a*). The proliferating daughter calli were selected and subcultured every fourteen days on the same medium at

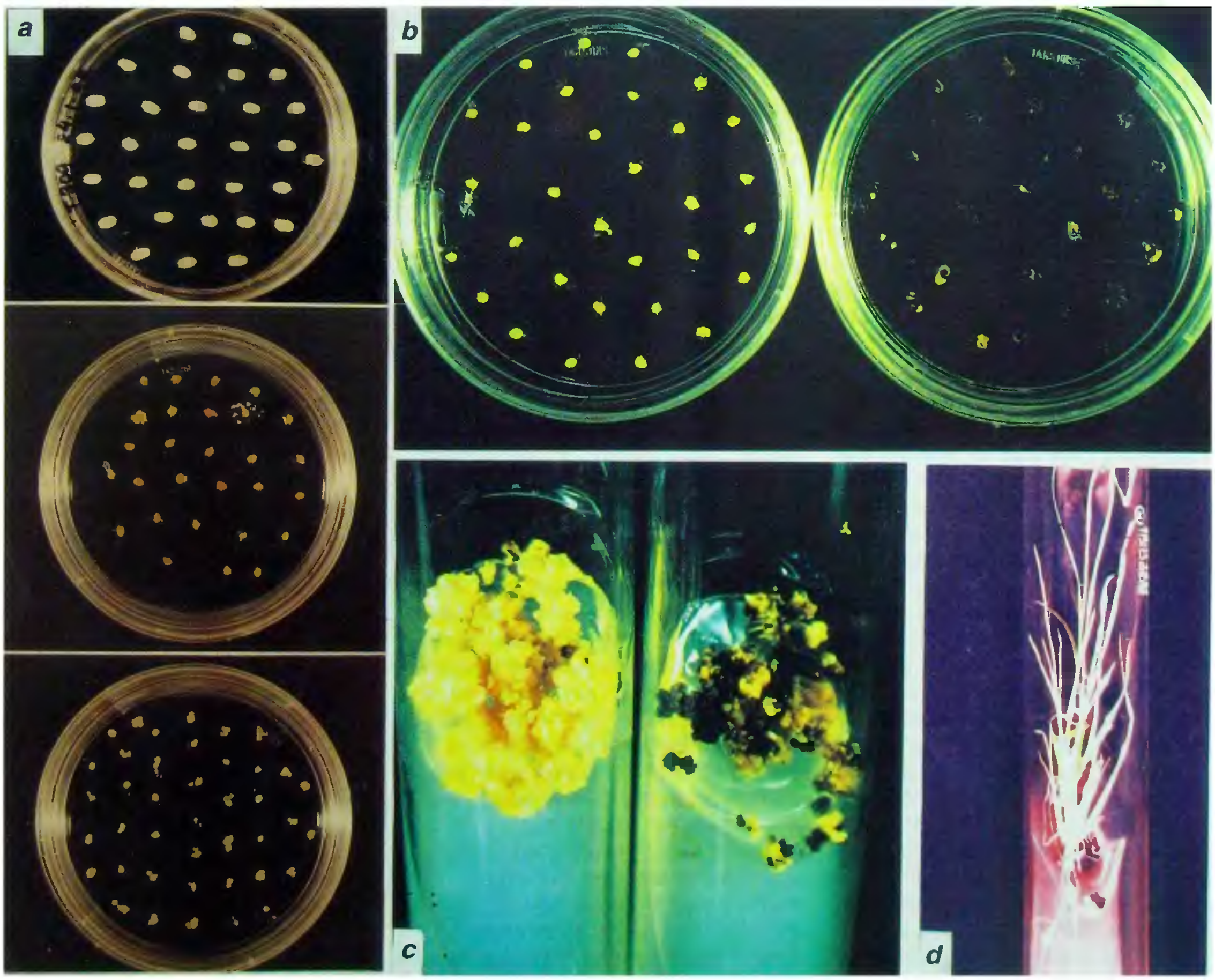

Figure 1. Tissue culture response of different rice cultivars. *a*, Stages in procuring scutellar calli for use in *A. tumefaciens*-mediated transformation of rice. Seeds of cultivar Taipei 309 (*top panel*), Excised scutella showing initiation of callus formation (*middle panel*), 1–2 mm-sized calli pieces employed as explants for co-cultivation with *A. tumefaciens* cells (*bottom panel*). Seeds, scutella and calli were inoculated on NB callus induction medium; *b*, Scutella of Taipei 309 (*left*) and IR 54 (*right*) after 7 days of inoculation on NB callus induction medium. Note that the scutellar portions of IR 54 have become necrotic upon subculturing whereas no such browning response is seen in Taipei 309 scutella; *c*, Response of Pusa Basmati 1 (*left*) and IR54 (*right*) calli after 15 days in secondary culture. Three-week-old primary calli (initiated from the mature seeds) were used for secondary culturing. MS callus induction medium was used; *d*, Regenerants of IR 54 from one-month-old primary callus.